Capturing drought stress resilience in grass through UAV Hyperspectral imaging

Stijn Kant

s.j.p.kant@students.uu.nl

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Supervisor: dr. Smigaj
Professor: prof.dr.ir. Bregt

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List of Abbreviations

EM	Electromagnetic
HS	Hyperspectral
PCA	Principal Component Analysis
VI	Vegetation Index
VWC	Volumetric Water Content
WUR	Wageningen University & Research

	Latin name	*Grass species*
Lp di	Lolium perenne L. diploid	Perennial Ryegrass diploid
Lp te	Lolium perenne L. tetraploid	Perennial Ryegrass tetraploid
Fa	Festuca arundinacea Schreb.	Tall Fescue
Pp	Poa pratensis L.	Kentucky Bluegrass
Frc	Festuca rubra L. Commutate	Red Fescue Commutate
Frt	Festuca rubra L. Trichophylla	Red Fescue Trichophylla
Frr	Festuca rubra L. Rubra	Red Fescue Rubra
Fo	Festuca ovina L.	Hard Fescue

Abstract

The primary aim of this research is to analyse the impact of drought stress on different turfgrass species, with the objective of identifying those that exhibit resilience under drought conditions. This contributes to the development of adaption strategies, an urgent need given rising frequency of drought events. Drought stress is experienced more frequently and severely by turfgrass species due to climate change in the last 10 years. This stress induces significant changes in vegetation health, which can be effectively analysed by hyperspectral remote sensing. This research focuses on evaluating the impact of drought stress on different turfgrass species and mixtures, considering two different mowing height, all intended for sports/events, park/recreation, and roadside applications. Hyperspectral reflectance data were collected using a Headwall Nano-Hyperspec camera of turfgrass subjected to drought conditions on four dates: 2nd and 26th of June and 18th and 30th of August. The study was conducted at the Nergena experimental field, located just North of Wageningen, Netherlands, where one group of grass plots were exposed to drought conditions and a control group of grass plots received irrigation. Reflectance data were used to calculate fifteen vegetation indices found in literature as successful in detecting drought stress in turf grass, focusing on five different sensitivities. These values were analysed, ranking the grasses on drought resistance and identifying similarities through Principal Component Analysis and hierarchical clustering. The study found significant differences between mowing height, with the plots mown on 6cm height performing better under drought conditions than the plots mown at 3cm height. Species with deeper root systems, such as Tall Fescue and Hard Fescue, exhibit the highest drought resistance, whereas species with open sods, Perennial Ryegrass tetraploid, showed the least drought resistance.

The influence of these specific species on the drought resistance of mixtures is notable; mixtures containing Tall Fescue and Hard Fescue exhibit the highest drought resistance, where those including Perennial Ryegrass tetraploid are the least resistant. These results provide insight into which species, mixtures and mowing heights can best be implemented across different use specifications to adapt to the climate change. This addresses the knowledge gap regarding the drought resistance of different cool season turfgrass species, mixtures and mowing height in already established sods.

Keywords: Drought resilience, hyperspectral remote sensing, turf grass species, vegetation indices, climate change adaption, clustering

Acknowledgments

First and foremost, I extend my deepest gratitude to Prof. Smigaj and Kooistra for their invaluable guidance. Their support was pivotal as I navigated through the unfamiliar territories of remote sensing and coding, areas in which my knowledge was initially limited. Their mentorship not only provided me with direction but also allowed me to connect my passion for nature with the focus of my thesis. This journey led to an in-depth analysis of the turfgrasses used in our study, a topic I found both fascinating and enriching. Additionally, I must express my appreciation to my loved ones for their patience and encouragement, enduring my endless discussions on drought stress in turfgrass.

1. Introduction

The ongoing effects of climate change are leading to more frequent droughts, a phenomenon observed globally, including in Europe. A study by Klijn (2012) examining climate change projections for the Netherlands indicates an anticipated increase in both the frequency and severity of droughts. These extended drought periods in Europe have damaging effects, both socially and economically (Ionita et al., 2016). Sectors such as agriculture and forestry experience damaging long-term effects on vegetation health resulting from these drought events (Hari et al., 2020). This includes yield losses, forest mortality and declining reservoir levels. Given the wide-ranging impact of drought vegetation stress across various sectors, numerous studies have been conducted to detect drought stress and examining the effects on plants (Katuwal, Yang & Huang, 2023; Hong, Bremer & van der Merwe, 2019; Bayat, van der Tol & Verhoef, 2016)

1.1 Evaluating Drought stress: Methods and Limitations

Governments are actively engaged in making their cities resilient as the effects of climate change persist, with a particular focus on climate-proofing public spaces, including parks (Albers et al., 2015). The strategic placement of drought-resistant vegetation is one approach to achieving this resilience. Urban parks, as well as sports clubs such as football and golf clubs, are increasingly interested in incorporating drought-resistant grass varieties into their turfs (Reiter et al., 2017). Determining which grass varieties can cope with these drought conditions is of great interest and practical relevance.

Various methodologies are employed to acquire knowledge about drought stress. One commonly used methodology in plant breeding is visual rating, which involves the assessment of vegetation quality and condition through visual inspection. Human evaluators examine the grass and assigning a score based on the quality, colour, texture and patterns of stress symptoms in the grass species (Haghverdi et al., 2021; Sherwood, 1983). However, this method relies on subjectivity and tends to be less dependable.
Another method involves the use of ground measurements, assessing parameters such as soil moisture, leaf water potential, stomatal conductance, chlorophyll content or leaf temperature. Nonetheless, these methods are labour-intensive and, being point measurements, do not offer a comprehensive analysis of entire research areas.

1.2 Advancements in Drought Stress Detection: The Role of Remote Sensing

Remote sensing techniques offer substantial advantages compared to these methods. The use of remote sensing with appropriate imaging technology offers fast, objective and consistent method of collecting data across larger research area. On an extensive scale, remote sensing techniques provide a more precise and effective method of detecting and monitoring drought stress than what can be obtained through ground measurements (Katuwal, Yang & Huang, 2023). Consequently, remote sensing is widely employed in various fields for vegetation stress detection, including precision agriculture (Hong, Bremer & van der Merwe, 2019), forest management (Le, Harper & Dell, 2023) and biodiversity conservation (Munné-Bosch & Villadongos, 2023).

One of most commonly used remote sensing techniques is hyperspectral (HS) remote sensing. This method involves the capture of reflected light at over more than 200 contiguous spectral bands within the visible to near infrared regions (Hong, Bremer and van der Merwe, 2019). The variation in vegetation reflectance across different wavelengths can be linked to various factors related to vegetation aspects, including pigment content, plant structure, water levels and biochemicals (Figure 1). These fluctuations in reflectance can be quantified through vegetation indices (VIs). When environmental conditions, such as drought stress, affect these vegetation factors, it is possible to analyse these impacts by examining the variations in the vegetation index (VI) values (Badzmierowski, McCall & Evanylo, 2019; Kim et al., 2011; Marshall et al., 2016).

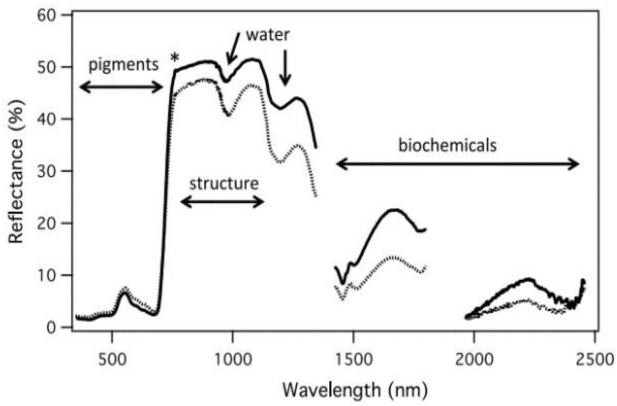


Figure 1: Canopy reflectance spectra showing spectral regions with information on different plant traits
Source: Gamon et al., 2019

1.3 Enhancing Drought Resilience: Hyperspectral Sensing in Turfgrass Management

In the domain of precision agriculture, there is a notable emphasis on assessing drought stress in turfgrass using remote sensing techniques. The increasing temperatures and precipitation variability driven by climate change have subjected an increasing area of turfgrass to drought stress conditions (Zarch, Sivakumar & Sharma, 2005). Consequently, the (early) detection of drought stress in grass species has gained significance. This has led to an increasing number of studies focusing on detecting and monitoring drought stress in cool season turfgrass species using different VIs or methods (Katuwal, Yang & Huang, 2023; Bayat, van der Tol & Verhoef, 2016; Hong, Bremer & van der Merwe, 2019). The majority of these studies sought to specify which spectral bands or remote sensing techniques are most suitable for detection of drought stress, largely focussing on single cool-season turfgrass species. There is still a relative scarcity of studies focussing on comparing the drought resilience of different cool season turfgrass species. Moreover, knowledge about the performance of different turfgrass species when grown in a mixture, as well as the performance of single species and mixtures in already established turfs, remains limited. This study aims to bridge this knowledge gap by providing valuable insights into the drought stress responses of cool season turfgrass species, mixtures and mowing heights in already established turfs.

2. Research Objectives

The aim of this research is to identify drought-resistant turfgrass species, mixtures and mowing height, to eventually implement these in public parks and sport fields to enhance their resilience to climate-related challenges. The research will focus on identifying drought resistance in the researched grass species using HS remote sensing techniques. As a result, this research aims to fill the existing knowledge gap by providing insights on which grass species, mixtures and mowing heights are drought resilience in already established turfs, thereby enabling their implementation to enhance resilience to climate fluctuations. With this, the main question of this study is:

"*MQ: To what extent can drought resilience of different grass species, mixtures and mowing heights be detected using hyperspectral remote sensing?"*

To answer the main research question, this study addresses four sub-research questions:

RQ1: Which vegetation indices preciously proved to be most effective in detecting drought stress in turfgrass species?

RQ2: What is the impact of drought stress on vegetation index values calculated for the grass varieties and compositions throughout the experiment?

RQ3: What is the impact of drought stress on the vegetation index values calculated for different mowing heights of the different grass varieties and compositions throughout the experiment?

RQ4: To what extent can similarities in drought responses be found between and across the turfgrass species, mixtures and mowing heights?

<u>Thesis outline</u>

This research begins with a comprehensive literature review in Chapter 3, aimed at identifying the most effective spectral bands and VIs for HS remote sensing in detecting drought stress in cool season turfgrass. This review will lead to the selection of 15 VIs which will be used to analyse the drought responses for the grass species, mixtures and mowing heights. In Chapter 4, the methodology is outlined, detailing the research setup and the selection of the single varieties and mixtures. This chapter also delves into the plant traits of the used species that are crucial for drought resistance and describes the drought treatment for the non-irrigated groups, alongside the methodological workflow used in this study. Chapter 5 present the results, showing the spectral signatures, calculated VI values and drought resistance ranking of the species, mixtures and mowing heights. It further explores the outcomes of Principal Component Analysis (PCA) and hierarchical clustering to find patterns and relationships in the data. The discussion in chapter 6 analyses these results within the context of existing literature, evaluating the implications of these findings and their significance in the broader field of drought stress management in turfgrass. Finally, Chapter 7 concludes the thesis by summarizing the key findings and contributions of the study, emphasizing the implications for the selection and management of turfgrass under the challenges of climate change and drought stress resilience.

3. Literature chapter

To address RQ1, a literature review was performed to identify a selection of usable VIs in detecting drought stress in the different turf grasses. Studies featuring these VIs are found by using keyworks like "hyperspectral indices drought stress turfgrass" and employing snowball sampling techniques. The results of these studies are summarized and form the basis of the selection of the VIs. The selection and equations of the indices are shown in Table 1.

3.1 Detecting drought stress with hyperspectral remote sensing

Drought effect on turf grass

Low volumetric water content (VWC) causes drought stress in grass, a condition that arises when a region undergoes a drought. Drought is defined as an extreme weather phenomenon, caused by the lack of precipitation (Paulo and Pereira, 2006). This phenomenon leads to a deficit in VWC and is observed in both areas with large and small amounts of precipitation. Drought is not restricted to specific seasons and can occur with different intensities (Staniak & Kocon, 2015).

Drought stress in grass, which limits growth, development and yield, can be categorized in two types. Moderate water stress reduces the growth and speed of cell division in leaves, occurring when the water content falls below tissue's saturation in the plant (Staniak & Kocoń, 2015). Bayat & Verhoef (2016) refer to this as short-term drought stress. Severe water stress is experienced after a prolonged water shortage, negatively effecting plant metabolism, especially the photosynthesis process (Staniak & Kocoń,, 2015). A decline in photosynthesis results in reduced stomata conductance, RuBisCo activity (the process of assimilating CO2 into the biosphere) and availability of CO2 (Hura et al., 2007; Jones, 1980). Bayat & Verhoef (2016) refer to this as long-term drought stress.

To which extent a plant can response and cope with drought is dependent on the plant's resistance. Some plants exhibit greater capacity in managing specific conditions, maintaining higher yields. This resistance is based on three characteristics of the plant: properties that determine its capacity to handle the stress factor, the ability to repair, and adaptation or acclimatization (Staniak & Kocoń, 2015). Plant properties include traits such as the depth of the root system or the level of stomatal conductance. The ability to repair damage concerns how fast and to what extent a plant can recover from a damaging period and restore its health. Adaptation relies on evolution and on the development of traits that enable vegetation to more effectively cope with changing conditions; breeding and mutations serve as driving factors for this evolutionary process. Acclimatization involves structural and functional modifications of plant traits as a response to the changing environment with modifications not being inherited unlike in adaptation (Staniak & Kocoń, 2015).

The processes of adaptation and acclimatization arise from strategies to avoid stress and cope with the effects of drought. These processes include traits that play a role in coping with drought, such as reducing transpiration and increasing efficient water uptake, conduction and storage (Blum, 2009). Considering the root system, most grasses have a root system situated in the upper soil (0-20cm), allowing them to extract a significant amount of water from a relatively small volume. However, grasses with a deeper root system, such as Festuca Arundinacea, reaching a depth of 2 meters, are considered more drought-resistant due to these characteristics (Carrow, 1996; Wilman, Gao

& Leitch, 1998). Another trait helping to cope with drought is the ability to reduce transpiration. Some grasses differ in their capacity to regulate stomata, thicken the cuticle (a protective layer covering the plant) or reduce the size or number of leaves (Staniak & Kocoń,, 2015; Chen & Zu, 2005; Cui et al., 2020).

Detecting drought stress in turf grass in crucial for effective vegetation management. Traditionally, the identification relies on the parameters Leaf Relative Water Content and Turf Quality (Katuwal, Schwarts & Jespersen, 2020; Liang et al., 2009; Leinauer et al., 2014), both of which respond to declines in VWC (Katuwal, Yang & Huang, 2023; Hu, Wang & Huang, 2013). While prior studies have demonstrated the utility of Leaf Relative Water Content in detecting drought stress in grass (Katuwal, Yang & Huang, 2023; Hu, Wang & Huang, 2013; Rahimi et al., 2010), the conventional method of measuring Leaf Relative Water Content is often destructive, involving the destruction of plant material. On the other hand, Turf Quality measurements are subjective, relying on the observer, impacting the precision and objectivity of the measurements. This is where remote sensing can play a crucial role.

Hyperspectral Remote Sensing

Remote sensing is defined as the extraction of information regarding objects, areas or phenomena based on their radiance, which can be acquired without the necessity to physically make contact (Camps-Valls et al., 2011). The origins of remote sensing trace back to 1903 when pigeons from the Bavarian Pigeon Corps were equipped with small, weighted cameras. These cameras, featuring a timer, took a picture every 30 seconds (Figure 2) (Colomina & Molina, 2014). Remote sensing has since evolved and become a key instrument for many monitoring applications, including the evaluation of drought stress in complex systems like landscapes and ecosystems (Avetisyan, Borisova & Velizarova, 2021).

Figure 2: One of the pigeons of the Bavarian Pigeon Corps with the mounted camera, shooting areal photos
Source: Remote Sensing tutorial Overview, n.d.

In optical remote sensing, sensors capture information within the wavelengths range of 400 nm to 2500 nm, including the visual, Near Infrared and Short-Wave Infrared spectrums. These spectrums are integral components of the total electromagnetic spectrum, which forms the solar energy emitted by the sun (Figure 3). The sensors measure the radiance, described by Shaw & Burke (2003) as the amount of light which is reflected by objects, areas, or phenomena. This can be achieved through passive sensor systems that collect electromagnetic (EM) radiance from the sun, or active sensors that transmit signals and measure their own reflectance (Bioucas-Dias et al., 2013).

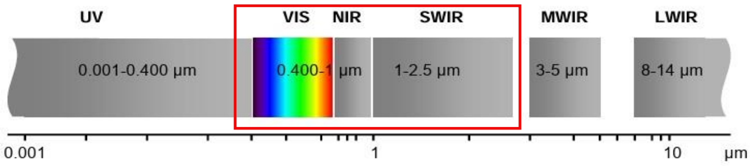


Figure 3: Electromagnetic spectrum, which exists of several spectrums. From small to large, Ultra-Violet, visual spectrum, Near-Infrared, Short Wave Infrared, Mid-Wave Infrared and Long Wave Infrared
Source: Pabich, 2002

When the sun's emitted energy encounters a surface, it undergoes three key interactions: reflection, absorption and transmission. Together, these interactions account for the total EM energy that reaches the earth. The way an object handles with this EM radiance depends on its characteristics, such as colour and reflectivity. Since colour and reflectivity effect the way this radiance is handled, the radiance and reflectance can be used to indicate the material compositions. Different materials react namely differently to the light in all different wavelengths because of its characteristics (Figure 4). As the reflectance spectrum remains constant regardless of illumination conditions, be it sunny or cloudy, it is a stable and reliable signature. The patterns within this signature can be utilized for the identification of materials and their properties (Cooke & Harris, 1970). This forms the foundation of optical remote sensing.

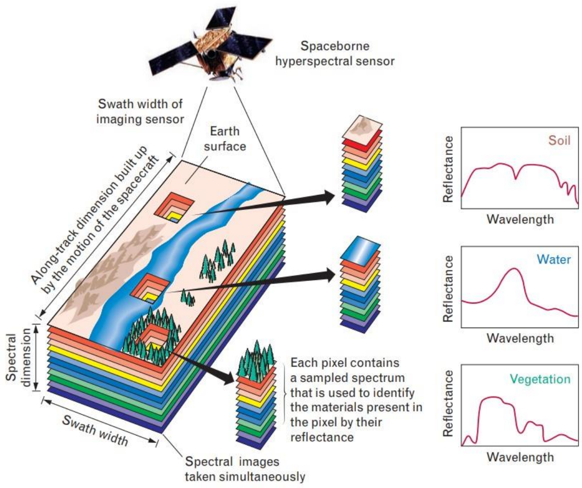


Figure 4: The concept of hyperspectral remote sensing
Source: Shaw & Burke, 2003

There are two main types of optical remote sensing sensors: multispectral sensors capture information in three to ten bands across the EM spectrum and HS sensors that capture information in more than two hundred continuous narrow bands. HS sensors offer significant advantages due to their operation with higher spectral resolution. In multispectral remote sensing, spectral information is averaged over broad bandwidths, leading to the loss of critical details in specific narrow bands, including absorption features (Blackburn, 1998; Thenkabail, Smith & Pauw, 2000). HS remote sensing has demonstrated its superiority over multispectral broadband-based remote sensing by providing crucial information for identifying both biophysical and biochemical parameters (Sahoo, Ray & Manjunath, 2015). Obtaining reflectance information at these specific narrow wavelengths offers for instance insights into chlorophyll or nitrogen levels, which, in turn, can be correlated with plant productivity, stress levels, or nutrient availability (Sahoo, Ray & Manjunath, 2015). This observation aligns with the findings of Katuwal, Yang and Huang (2023), who compared the efficacy of HS and multispectral VIs in detecting drought stress in turfgrass. Their study reveals that HS VIs demonstrated a higher predictability for Leaf Relative Water Content and Turf Quality compared to indices derived from multispectral imaging.

HS Remote sensing capturing information on vegetation

Transition from the broad concept of remote sensing to the nuanced insights of HS remote sensing, the focus shifts towards implementing HS remote sensing for deeper understanding of plant health. HS remote sensing is applicable in a wide range of agricultural uses, since different vegetation types have their own unique spectral reflectance signature of various types of vegetation captured by large continuous narrow bands (Sahoo, Ray & Manjunath, 2015). The amount of reflected and absorbed EM radiance in these bands can be linked to specific vegetation traits and functions (Moss & Loomis, 1952; Li, Zhang & Huang, 2014; Zhang et al., 2021). This information not only provides insights into the biochemical and physical properties of plants, but also enables studying various aspects of plant health and environmental interactions (Kureel et al., 2022; Blackburn, 2007; Bayat, Van der Tol & Verhoef, 2016)

Different plant traits influence the reflectance at specific wavelengths as shown in Figure 5. Reflectance in the visible region provides insight on the leaf pigments, such as chlorophyll (Hadoudane et al., 2002). Reflectance in the near-infrared region provides insights into the cell structure, linking to the scattering in the spongy mesophyll. The spongy mesophyll, rich of chloroplasts, is the internal tissue of a leaf. This is the site where the photosynthesis takes places, converting light energy into chemical energy. Finally, the reflectance of the shortwave infrared provides insights into the leaf water content (Champagne et al., 2003). Figure 5 also shows different absorption bands, which have an effect on the measured reflectance. In the visible region vegetation's reflectance signature include 2 chlorophyll absorption bands, one in the blue region around 450 nm and one in the red region around 670 nm and several water absorption bands caused by the atmospheric such as the water absorption bands at wavelength 1.45 µm and 1.94 µm. These water absorption bands can provide advantages as they can be used to collect more information through thick canopies and have a preferential sensitivity to thinner than to thicker tissues (Sims & Gamon, 2003).

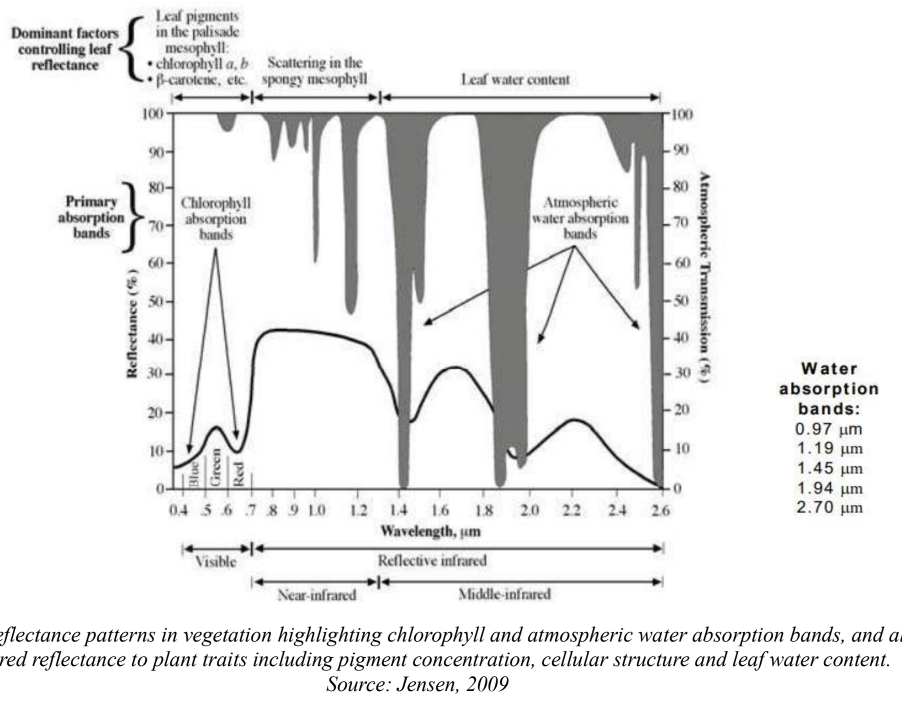


Figure 5: Spectral reflectance patterns in vegetation highlighting chlorophyll and atmospheric water absorption bands, and also attributes measured reflectance to plant traits including pigment concentration, cellular structure and leaf water content.
Source: Jensen, 2009

Shifts in level of leaf pigment or cell structure affect the ability of plants to absorb specific wavelengths, which can be observed in changes of vegetation's spectral reflectance. Based on these changes in measured reflectance vegetation stress can be observed, such as drought stress (Badzmierowski, McCall & Evanylo, 2019; Kim et al., 2011; Marshall et al., 2016). Figure 6 illustrates this phenomenon, showing the reflectance of grass under different soil moisture levels and presenting the percentage changes observed across different wavelengths. As soil moisture decreases, the grass experiences drought stress, resulting in changing reflectance signature. This occurs since the drought stress symptoms gradually change over time (Kumar et al., 2021). In the visible range, lower soil moisture levels correlate with reduced absorption in the red, blue and green bands. It's important to note that vegetation health can be affected by a variety of stress factors, both abiotic – such as temperature, CO2 levels, radiation, water and nutrients – and biotic, including weeds pests and diseases (Tittonel & Giller, 2013). This fluctuation can lead to either reversible or irreversible disturbances in vegetation functioning and structure, further influencing the observed changes in spectral reflectance.

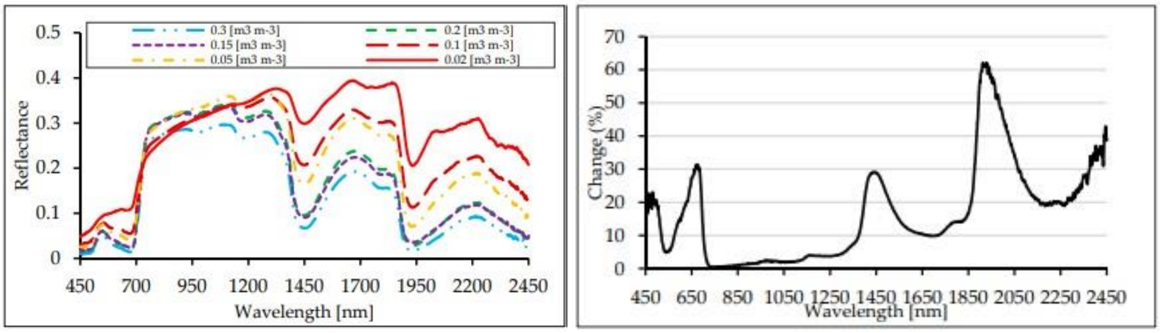


Figure 6: Change of reflectance spectra of grass in responses to drought & Percentage of change observed per wavelength
Source: Bayat, van der Tol & Verhoef, 2016

VIs exploit this information by capturing variations in measured reflectance. As reflectance at different wavelengths linked to distinct traits of the vegetation, combinations of VIs are employed for a comprehensive analysis. The use of remote sensing has become increasingly important, as they enable the capture of detailed spectral information from plants without causing damage. The spectral reflectance serves as a tool for rapid, non-destructive objective and consistent monitoring drought stress in vegetation (Mishra et al., 2019; Damm et al., 2014). VIs can vary in their sensitivity to detect, as their calculation use the reflectance of different wavelengths (Bayat, van der Tol & Verhoef, 2016).

3.2 Literature review: Vegetation indices

The focus is on HS VIs within the range of 0 – 1000 nm, which corresponds to the reflectance range used in this study. VIs beyond this range are excluded from consideration. Within this literature review, twelve studies were examined, each conducted research on the detection of drought stress on turfgrass through the use of HS data (Table 9 in Appendix A). Among these, ten studies conducted research on the relationship between drought stress and VIs, while two focused on the correlation with specific spectral bands. In these studies, drought stress was identified due to decreasing SWC. The majority of the articles conducted research on irrigated plots with a constant VWC level and non-irrigated plots undergoing drought or dry downs. Different parameters were measured to assess grass health, including Turf Quality (Badzmierowski, McCall & Evanylo, 2019; Caturegli et al., 2020; Jiang, Liu & Cline, 2009; Katuwal, Yang & Huang, 2023; McCall et al., 2017; Roberson et al., 2021; Jiang & Carrow, 2005;

Jiang & Carrow, 2007), Biomass (Badzmierowski, McCall & Evanylo, 2019), Tissue Nitrogen Accumulation (Badzmierowski, McCall & Evanylo, 2019), VWC (Badzmierowski, McCall & Evanylo, 2019; Caturegli et al., 2020; Jiang, Liu & Cline, 2009; McCall et al., 2017; Roberson et al., 2021), Leaf Relative Water Content (Caturegli et al., 2020; Katuwal, Yang & Huang, 2023), soil moisture (Caturegli et al., 2020; Jiang, Liu & Cline, 2009), Chlorophyll pigments level (McCall et al., 2017), Leaf Firing (Jiang & Carrow, 2005; Jiang & Carrow 2007) and Wilt Percent (Roberson et al., 2021). Pearson correlations were calculated to assess relationships between the values of the VIs and these parameters. Studies that did not calculate correlations with these parameters investigated whether significant changes in VI values were observable during the drought treatment (Bayat, Van der Tol & Verhoef, 2016; Dao et al., 2021; Hermans et al., 2021).

In total, 28 different VI's were identified in these studies, including 5 different sensitivities (Figure 7). The indices NDVI, WBI, SRI, PRI and GRI were most frequently employed, appearing 8, 6, 4, 4 and 3 times, respectively. The results regarding the ability of these indices in detecting drought stress have been compiled, and the corresponding Table 9 is available in Appendix B. Their effectiveness was evaluated through a comparative analysis of results from the relevant studies, selecting only those VIs identified by these identified as most proficient in detecting drought stress. Based on these results, the indices were categorized as either suitable, possibly suitable or not suitable for detecting drought stress in grass. A further distinction was made between the detection of early or long-term drought stress (Figure 7). For this research, the definition provided by Bayat & Verhoef (2016) was adopted. Their definition indicated that short-term drought could be detected after 11 days, while long-term drought stress could be identified after 36 days.

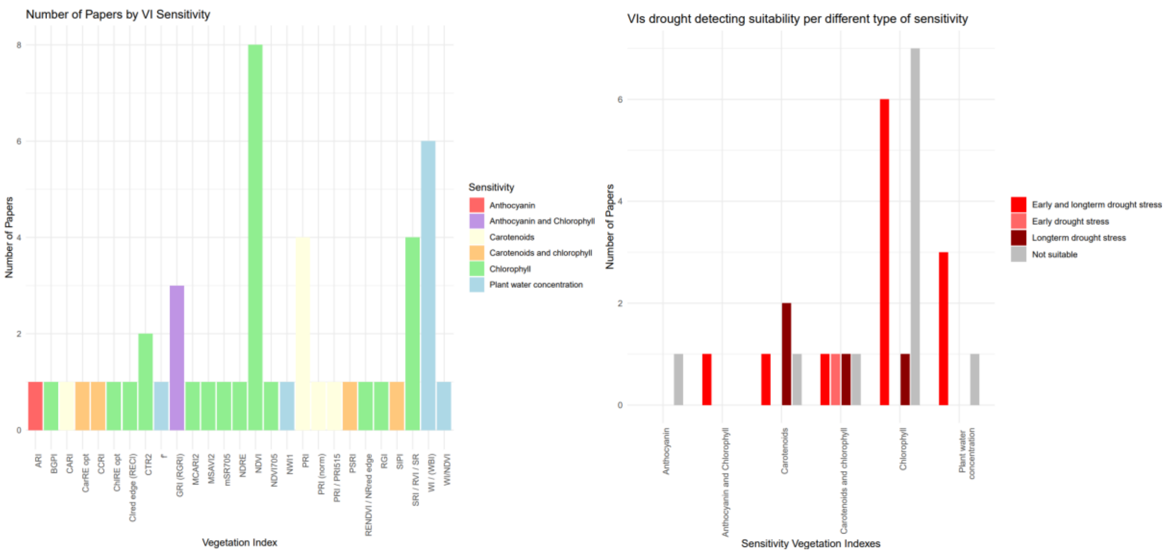


Figure 7: Number of citations for different VIs in the relevant articles & Categorization of VIs based on suitability and type of sensitivity

Subsequently, the VIs have been categorized based on their sensitivity to different plant characteristics. Within the 28 VIs identified, the following sensitivities were observed: Anthocyanin, Anthocyanin & Chlorophyll, Carotenoids, Carotenoids & Chlorophyll, Chlorophyll, and Plant Water Concentration.

Anthocyanin, Carotenoids and Chlorophyll are pigments that absorb radiance all at a different wavelength (Figure 8) and transport the light energy to other parts of the photosynthetic process (Hallik et al., 2017). Anthocyanin represents red, purple, blue and black pigments, Carotenoids represents yellow, orange and red pigments, and Chlorophyll represents green pigments. Both Anthocyanin and Carotenoids play a protective role when drought stress induces oxidative stress, leading to the overproduction of reactive oxygen species. These pigments counteract different forms reactive oxygen species to protect the plant's photosystems, resulting in higher levels of Anthocyanin and Carotenoids when turfgrass experiences drought (Shariatipour et al., 2022; Hallik et al., 2017). This research identified a single VI sensitive to Anthocyanin capable of detecting drought stress, which also exhibits sensitivity to Chlorophyll. VIs responsive to Carotenoids illustrate enhanced performance in detecting drought stress, with the majority effectively identifying drought stress, including those combined with Chlorophyll sensitivity. The VIs sensitive to Chlorophyll measure a decrease in level of the pigment when turfgrass experiences drought (Jazi, Etemadi & Aalipour, 2019). This decline is also linked to oxidative stress (Kato and Shimizu, 1985). This study found that most VIs were sensitive to Chlorophyll, with about half capable of detecting drought stress. Additionally, indices sensitive to plant water content, indicating the plant's hydration level, were notably effective in identifying drought stress.

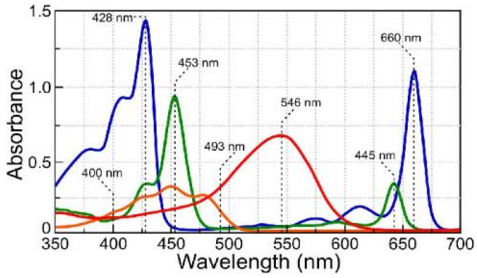


Figure 8: Overview of the absorption spectra of isolated chlorophyll a (blue), chlorophyll b (green), β-carotene (orange) and Anthocyanin (red)
Source: Barragán et al., 2018

In total, this research identifies 15 VIs capable of detecting drought stress, shown in Table 1. This excludes the widely used NDVI and PRI indices. Instead, modified versions, namely PRI(norm) and WI/NDVI, are used in this study for their better performance in detecting drought stress. A comparison of the wavelengths used in these VIs with the findings of Jiang& Carrow (2005 & 2007), who conducted research on the relationship between wavelength bands and drought stress parameters Turf Quality and Leaf Firing, reveals notable similarities. Wavelengths around 710 are associated with vegetation stress, around 660 nm, between 673 – 693 nm and around 900 nm are linked to TQ, while 667 – 687 nm is linked to LF. In the equations, the wavelengths 670 nm, 680 nm, 690 nm, and 695 nm are used, corresponding to Turd Quality and Leaf Firing, and 900 nm is linked to water content.

Table 1: Relevant VIs for this research

VI	Full name	Equations	Sensitive to	Suitable for short-and long-term drought stress
GRI	Green to red ratio index	GRI = R550 / R670	Anthocyanin and Chlorophyll	Short- and long-term
PRI (norm)	Normalized Photochemical Reflectance Index	PRI (norm) = (R570 – R531) / (R570 + R531) / [((R800 - R670)/sqrt (R800 +R670)) * R700/R670]	Carotenoids	Long-term
CARI	Carotenoids Index	CARI = (R720 – R521) / R521	Carotenoids	Short- and long-term
PRI512	Photochemical Reflectance Index 512	PRI512 = (R531 − R512)/(R531 + R512)	Carotenoids	Long-term
CarRE opt	Opt. carotenoid red edge index	CarRE opt = $(\rho_{510-530}^{-1} - \rho_{680-730}^{-1}) \times \rho_{760-780}$ *	Carotenoids and chlorophyll	Long-term
CCRI	Carotenoid/Chlorophyll Ratio Index	CCRI = ((R720–R521) / R521) / ((R750 +R705)/R705)	Carotenoids and Chlorophyll	Short- and long-term

SIPI	Structure Independent Pigment Index	SIPI = (R800 – R445) / (R800 + R680)	Carotenoids and chlorophyll	Short- term
BRI2	Blue/Red Pigment Index 2	BRI2 = R450 / R550	Chlorophyll	Short- and long-term
ChlRE opt	Opt. chlorophyll red edge index	ChlRE opt = (R680 –730^{-1} − R780–800^{-1}) × R755–780 *	Chlorophyll	Long-term
CTR2	Carter Index 2	CTR2 = R695 / R760	Chlorophyll	Short- and long-term
mSR705	Modified Simple Ratio	mSR705 = (R750 – R445) / (R705–R445)	Chlorophyll	Short- and long-term
RGI	Red/green pigment Index	RGI = R690 / R550	Chlorophyll	Short- and long-term
WI/NDVI	Ratio WI normalized difference vegetation index	WI/NDVI = (R900 / R970) / ((R800 − R680) / (R800 + R680)	Plant water concentration	Short- and long-term
NWI1	Normalized Water Index 1	NWI1 = (R970 - R900)/(R970 + R900)	Plant water concentration	Short- and long-term
WBI	Water Band Index	WBI = R900 / R970	Plant water concentration	Short- and long-term

* R520, R705, R770 are selected for CarRE opt and R705, R790 and R768 for ChlRE opt, as they fall at the central points of their ranges

4. Methodology

This methodology chapter serves as the structure of the study, presenting the experimental setup, drought treatment and the collection of HS remote sensing data critical for analysing drought stress responses in turfgrass. By outlining the methodological workflow, this chapter aims to guide the readers through the processes employed to address the research questions.

4.1 Experimental Design/Setup

Just north of the Wageningen Campus, Wageningen University & Research (WUR) established Nergena, an experimental field designated for several years observation on grass fields featuring specific compositions and under subjected to different maintenance regimes. This field underwent sowing on the 1st of September 2021.

The experimental fields cover a total area of 0.25 hectares and contains 48 plots measuring 4 by 4 meter and 192 plots measuring 2 by 2 meters (Figure 9). These plots have been sown with both single varieties and mixtures of species commonly found on sport/event fields and park/recreation areas. The research setup includes two different maintenance regimes, strips of grass measuring 3cm (left) and 6cm (right). Furthermore, the research setup includes two distinct irrigation methods: the right half of the field receives irrigation depending on the soil moisture level, while the left half remains non-irrigated. Additionally, three separate test groups for comparative analysis are incorporated, ensuring that any observed effects are attributed solely to drought stress. The research set-up is shown in Figure 9.

Figure 10 provides a comprehensive overview of the different grass species and mixtures sown in the research setup. A total of eight distinct species were sown, both in monocultures and in mixtures. Labels A to D were sown in larger plots measuring 4 x 4 meters, featuring mixtures primarily used in parks, sports and event fields. The remaining labels were planted in plots measuring 2 x 2 meters, featuring all the included species of the research in monocultures, labelled from E to M. Labels N to P feature mixtures used for sports/events, S to U for park/recreation and W for roadside applications.

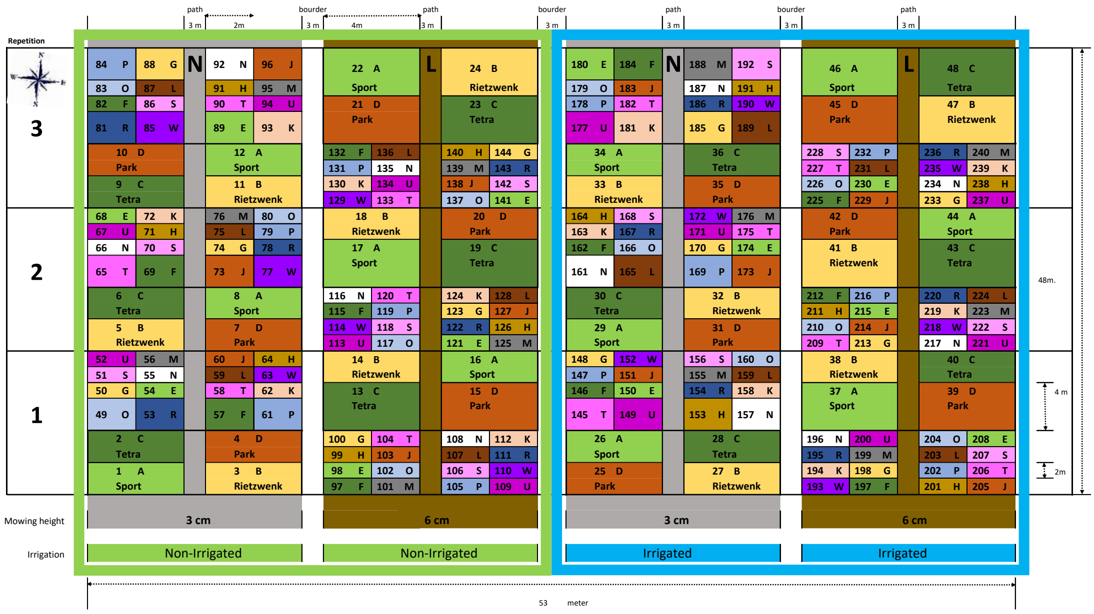


Figure 9: Overview of Research Set-Up

Species composition	Code	Plot	Lp di	Lp te	Fa	Pp	Frc	Frt	Frr	Fo
Perennial Ryegrass (sport)	A	4 x 4	50			50				
Tall Fescue	B	4 x 4			100					
Perennial Ryegrass Tetraploid	C	4 x 4		100						
Red Fescue (park)	D	4 x 4	20			20	30	30		
Perennial Ryegrass diploid	E	2 x 2	100							
Perennial Ryegrass tetraploid	F	2 x 2		100						
Tall Fescue	G	2 x 2			100					
Kentucky Bluegrass	H	2 x 2				100				
Red Fescue commutate	J	2 x 2					100			
Red Fescue Trichophylla	K	2 x 2						100		
Red Fescue Rubra	L	2 x 2							100	
Hard Fesue	M	2 x 2								100
Sport/events	N	2 x 2	50			50				
Sport/events	O	2 x 2		50		50				
Sport/events	P	2 x 2	50		50					
Sport/events	R	2 x 2			50	50				
Park/recreation	S	2 x 2	35			50		15		
Park/recreation	T	2 x 2	20		25	25	10	10	10	
Park/recreation	U	2 x 2	20			20	20	20	20	
Roadside	W	2 x 2					20	10	20	50

Figure 10: Overview of composition of species

4.2 Drought treatment

To accurately assess the effects of the drought treatment, it's crucial to gather detailed weather data that influenced the grasses' conditions. This approach enables attributing the observed stress in the experiment directly to drought conditions. The experiment involves data collection on four different dates, spanning from 2nd of June to 30th of August. During this period, temperatures exceeding 30 degrees were recorded on 10 days, with the KNMI officially declaring a heatwave from 9th to 16th of August (KNMI, 2023). The KNMI defines a heatwave as a sequence of at least 5 summer days (maximum temperature of 25.0 C° or higher) in De Bilt, of which at least three are tropical (maximum temperature of 30 C° or higher). Table 2 contains the measured temperatures on the research field.

Table 2: Temperature Measurements weather station Veenkampen
Source: Wageningen University & Research, 2022

		Number of days temperature measured			Highest measured temperature	
Month	**Average temp C°**	**Above 25 C°**	**Above 27.5 C°**	**Above 30 C°**	**Temp C°**	**Date**
June	22.8	7	4	1	30.6	22-6-2022
Jule	25	9	5	2	36.6	19-7-2022
Aug	26.4	15	11	7	33.6	25-8-2022

Throughout the study, VWC measurements were taken on specific dates: June 16, 22, 23 and 28; July 18 and 19; and August 9, 18, 23, 24 and 30 (Figure 11a). These measurements, conducted on both irrigated and non-irrigated plots, revealed a decline in VWC within the non-irrigated plots between 23rd of June and 18th of July. Similarly, a decrease in VWC beneath grass surface was recorded at the Veenkampen weather station, sited 4 kilometres from the research site (Figure 11b). This decline is associated with a lack of precipitation and the increase in averaged and maximum temperature from the 1st of July (Figure 11 c and b). The combination of these observations conclusively attributes the noted stress to drought conditions, indicating that the turf began experiencing drought stress around the 14th of July, as indicated from data at both the research site and the Veenkampen weather station.

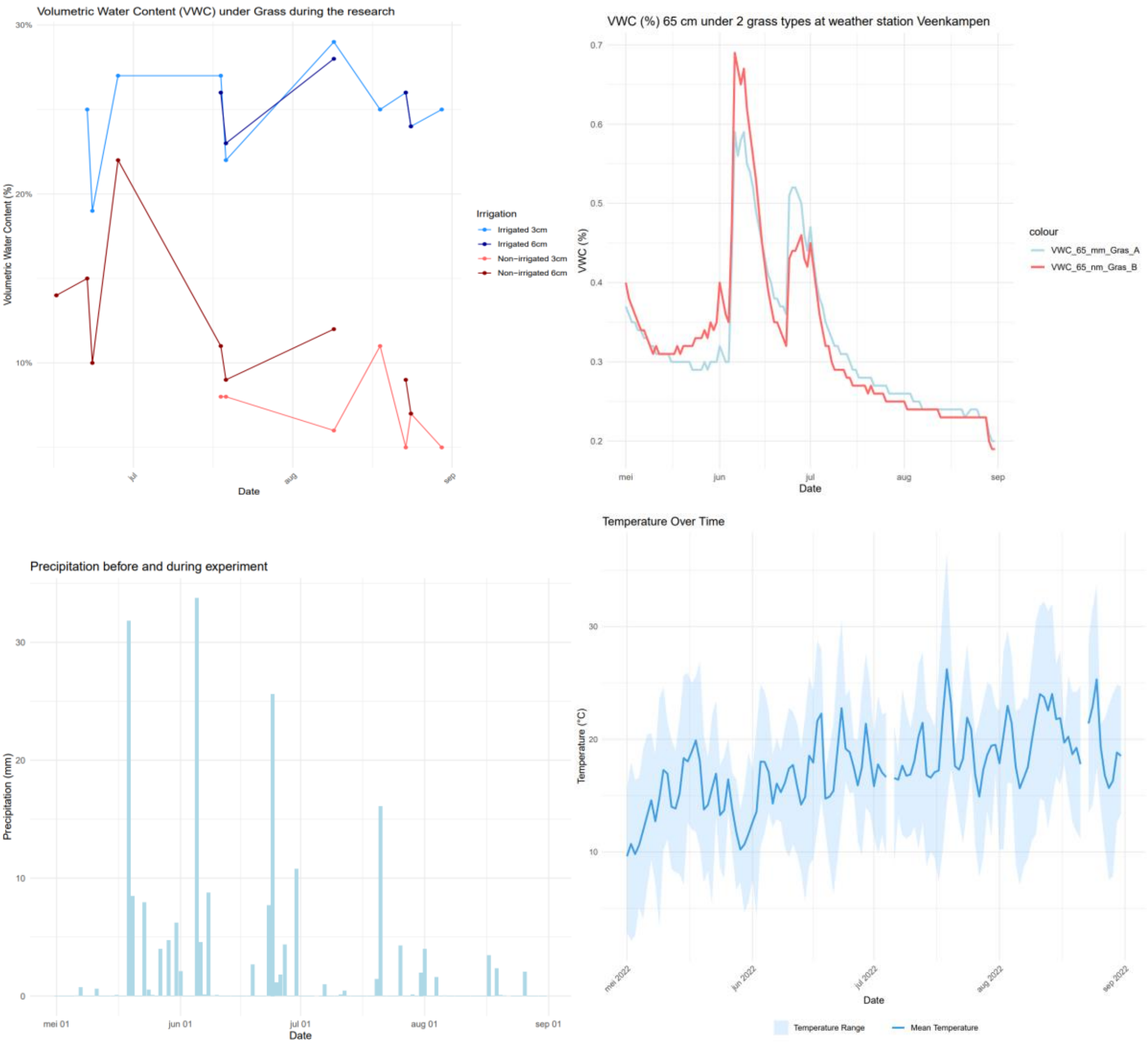


Figure 11: (a) Measured VWC (%) on the research plots, (b) measured VWC (%) 65 cm under 5 grass types at weather station Veenkampen, (c) precipitation before and during experiment measured at weather station Veenkampen, and (d) temperature measured at weather station Veenkampen
Source: Wageningen University & Research, 2022

4.3 Plant characteristics

The study includes eight turfgrass species, researched in single variety and in mixtures. The following section delves into a comparative analysis of the plant characteristics that impact drought tolerance, aiming to provide knowledge on the differences in these plant traits and connect them later to the observed results. This will offer insights into how specific plant traits contribute to the drought resilience measured.

<u>Roots</u>

Root analysis is conducted based on different characteristics, including plant height, rooting depth, root mass, and root distribution. Brown et al. (2010) conducted research on these traits across turf grass species. This research results, illustrated in Figure 12, show plant height, root depth and root mass distribution of those 5 grasses. The average root depth and root mass are presented in Table 3. Analysing the findings, indicates that Perennial Ryegrass and Tall Fescue exhibit the deepest roots, followed by Hard Fescue and Red Fescue, respectively. In contrast, Hairgrass, closely linked to Kentucky Bluegrass, produces the shallowest roots of these 5 grasses. Deeper roots enable the extraction of water from further beneath the surface, providing a substantial advantage during drought conditions and greatly influencing a species' resilience (Sheffer, Dunn & Minner, 1987).

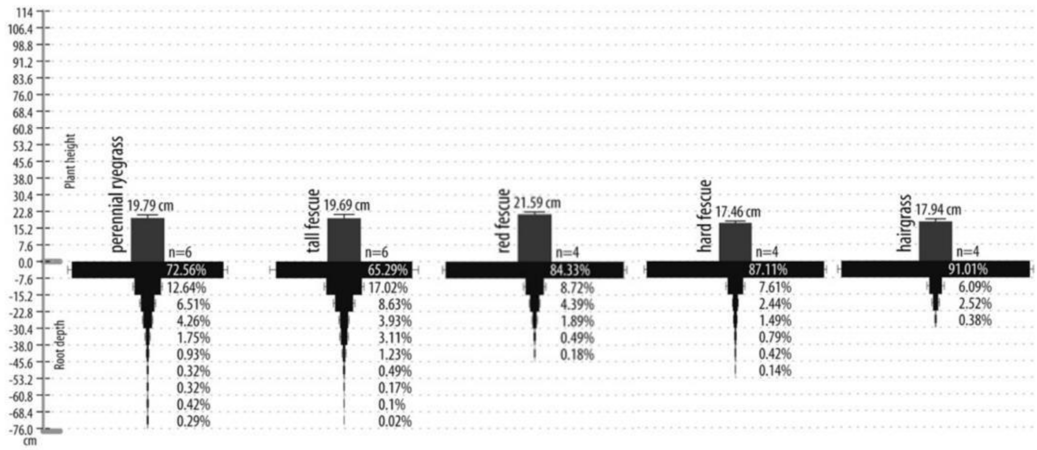


Figure 12: Plant height, root depth and root mass distribution for Perennial Ryegrass, Tall Fescue, Red Fescue, Hard Fescue and Hairgrass
Source: Brown et al., 2010

These proportional differences among the included species are also evident in other studies. Scheffer et al. (1987) conducted research on determining root distribution and soil moisture depletion by Kentucky Bluegrass, perennial ryegrass, and tall fescue. The results show that for Kentucky Bluegrass 75% of its roots are concentrated within the top 12 cm, while for Perennial Ryegrass and Tall Fescue, this percentage is 50%. This finding confirms the idea that Kentucky bluegrass has a shallower root system than Perennial Ryegrass and Tall Fescue. Moreover, Lin (1985) offers an overview of potential root depth. This overview reveals that Tall Fescue, in particular, can potentially develop deep roots, with a potential root depth from 45 to 150 cm. In conclusion, Tall Fescue is characterized by the deepest root systems, with Hard Fescue coming in a close second, and Perennial Ryegrass ranking third.

Table 3: Plant characteristics of 5 grasses based on different studies

	Average Rooting depth (cm) (Brown, 2010)	**Average Root mass (g)** (Brown, 2010)	**Root mass distribution 0-12 cm** (Scheffer, 1987)	**Potential root depth (cm)** (Lin, 1985)	**Leaf width (mm)** (Hannaway et al., 1999a; Hannaway et al., 1999b; Oliveira Prendes, 2008; Prendes & Palencia, 2015; USDA NRSC, 2004)	**Maximum Evaporation rate (mm per day)** (Huang, 2008)
Perennial Ryegrass	45,6	0,5	50%	20 - 45	2 – 6 .	8,5 - 10
Tall Fescue	50,7	0,5	50%	45 - 150	3 – 12 .	> 10
Red Fescue	34,2	0,3	75%	20 - 45	0,3 – 1,2 .	8 – 8,5
Hard Fescue	51,3	1,8	-		0,6 – 2,5 .	7 - 8,5
Hairgrass[1] / Kentucky Bluegrass[2]	20,9 [1]	0,2 [1]	-	20 – 45 [2]	2 – 5 [2].	> 10 [2]

Leaf width and evaporation

Two other characteristics associated with drought resistance are leaf width and evaporation. Research has indicated a significant relationship between these factors (Parkhurst & Loucks, 1972; Maylani, Yuniati & Wardhana, 2020), which is also reflected in the data on leaf width and evaporation rates for each grass type presented in Table 3. Leaf width is also linked to turf quality, as notably fine leaves contribute to a closed turf, whereas wider leaves results in a more open turf (Bals, z.d.). Additionally, evaporation can be linked to water use efficiency. For instance, Perennial ryegrass thrives in waterlogged soils since it has high moisture tolerance and tends to grow better in cool, moist soils (Hannaway et al., 1999), while red fescue is considered more drought-resistant (Bals, n.d.).

Varieties

This research includes not only different turfgrass species but also different varieties of the same species. Specifically, there are two variations of Perennial Ryegrass, Diploid and Tetraploid, and three variations of Red Fescue: Commutate (normal red fescue), Trichophylla (fine rhizomes) and Rubra (strong rhizomes). The distinctions between these variations are detailed in Table 4. Most importantly, the data shows differences among the varieties in terms of closed and open sods.

The specific characteristics between variations contribute to different performances. This is shown by the research of Demiroglu et al. (2010), who conducted research on the turf cover scores of the three red fescue variations over three years. The findings indicate that Rubra exhibit the highest average turf cover score, followed by Trichophylla and Commuta, respectively. However, when compared to other fescue species, more significant performance difference is seen. This pattern is consistent with findings from other studies, such as the research conducted by Ayan, Arslan & Acar (2020). Minor differences may be observed between varieties for thinning ratios, more substantial differences are evident comparing with other species like Festuca Arundinacea or Poa Trivalialis.

Table 4: Specific characteristics between variations Perennial Ryegrass & Red Fescue
Source: Hannay et al., 1999 & Bals, n.d.

Perennial Ryegrass		**Red Fescue**		
Diploid (Lp di)	*Tetraploid (Lp te)*	*Commutate (Frc)*	*Trichophylla (Frt)*	*Rubra (Frr)*
Faster growth rate More smaller tillers with thinner leaves, resulting in more closed sods	Less winter-hardy and persistent Less but larger tillers with wider leaves, resulting in more open sods	Extremely thin leaves, resulting in more closed sods	Produces short underground sprouts, enabling it to find open spaces in sods Extremely thin leaves, resulting in more closed sods	Produces long underground sprouts, enabling it to find open spaces in sods even better Wider leaves, resulting in more open sods

<u>Mixtures</u>

Turf serves a variety of purposes, each with its specific circumstances such as soil quality, sowing seasons, SWC, as well as variations in extent of use, maintenance, fertilization, and shading. The use of mixtures often presents greater possibilities for cultivating a healthy turf in such diverse conditions, as mixtures exhibit greater adaptability and are less prone to illnesses (Dunn, Ervin & Fresenburg, 2002). Hence, this research not only focuses on grass species but also emphasizes turf mixtures.

To meet the needs of different applications, specific usage categories were identified, and different mixtures were created for each set of conditions. These categories include sports/events, park/recreation, and roadside. Sports turfs are specifically created for field sports such as football, hockey, and korfball. Recreation/park turfs are commonly found in camping sites or city parks, with ornamental value considered secondary. Table 5 provides detailed information on the required operational and maintenance specifications.

Table 5: Different use specifications and maintenance for grass varieties
Source: Plantum, 2016

Grass varieties	**Use specifications**	**Maintenance**
Sport / Events	Dense and level sod Hardy and resistant Endure intense use	Spring: fast growth rate, require high share Perennial Ryegrass Late summer: With temperature high enough and moisture not being a problem for the slow establishment of Kentucky Bluegrass. This species is preferred for a dense and level sod.
Park / recreation	Primarily used in summer and recovers in autumn and winter Recover after long-term covering, relying on underground shoots	When turf is walked on and mowed (3-5 cm) a lot, the growth conditions are suitable for Perennial Ryegrass and Kentucky Bluegrass Drought conditions are also suitable for Kentucky Bluegrass Less walked or poor, dry soil is still suitable for Hard Fescue Less mowing is best for Red Fescue and Kentucky Bluegrass
Roadside	Must settle quickly, stand firm, strong sod, fast regrowth open spaces	Frequent mowing is not possible as the mixture requires a slow growth rate

Several mixtures can be used for the same use specification, as their characteristics align with these needs. The selection for mixtures is beneficial because a greater diversity of species improves genetic diversity and facilitates adaption to different microenvironments (Donald,1963; Steinke & Ervin, 2013). In terms of drought resistance, the genetic diversity also means that mixtures with different proportions of grasses exhibit different drought resistance. For instance, Non et al. (2010) found that turfgrass mixtures with a high proportion of Hard Fescue were more drought-resistant than those with a high proportion of Slender Creeping Red Fescue. The study of Reiter et al. (2017) also demonstrated that genetic diversity within turfgrass mixtures does not inherently enhance drought resistance, as there were no significant differences observed between monocultures and mixtures regarding retaining green cover rates. Moreover, this research identified the influence of specific species on mixtures performance, noting that Sheep Fescue and Slender Creeping Red Fescue negatively affect recovery. These findings challenge older ideas that diverse mixtures were necessary to preserve a healthy vegetative cover (Watschke & Schmidt, 1992). While genetic diversity in turf grass mixtures might offer benefits against diseases and other stresses, it is not necessarily the case for increasing drought tolerance. To improve drought resistance, the variety of species in a mixture is less crucial than selecting the most appropriate species. Specifically, species that feature deep roots, closed sods from thin leaves, and low evaporation rates are essential. These characteristics ensure that grasses maximize water uptake and minimize water loss, effectively increasing drought tolerance.

4.4 UAV-based remote sensing and image analysis

The aerial HS imagery was acquired using a Headwall Nano-Hyperspec camera mounted to a DJI M300 RTK drone. Aerial data was captured on four distinct dates: the 2nd and 16th of June, and the 18th and 30th of August 2022. This camera utilized a 14 mm lens and records radiance across 270 bands in the visible and near-infrared spectrum, spanning from 400 to 1000 nm. The data is collected at a spectral resolution of 2.2 nm and a spatial resolution of 4 cm. The drone's flight speed was configured at 3 m/s and the flight altitude was set to 50 meters (later refined by pixel size). A 40% overlap was employed during data collection, which took place within a time frame of approximately ±2 hours from solar noon.

4.5 Methodological workflow

To address the main research question, the research follows a methodological workflow outlined in Figure 13. This workflow describes clear stages associated with the research questions, which are further discussed.

<u>Selection of VIs capable of detecting drought stress</u>

In this research first a comprehensive literature review is conducted. Studies are identified using specific terms on Google Scholar, including "Hyperspectral indices drought stress turfgrass". Only studies conducted on cool season turfgrass and using HS data are considered, leading to a selection of 12 relevant studies. The VIs found with a sensitivity to drought stress are evaluated through a comparative analysis of results from the relevant studies, selecting only those VIs identified by these identified as most proficient in detecting drought stress. Based on these results, the indices were categorized as either suitable, possibly suitable or not suitable for detecting drought stress in grass. These selected indices form the basis for constructing a dataset providing insight into the grasses health status during the experiment. The equations of the indices are shown in Table 1.

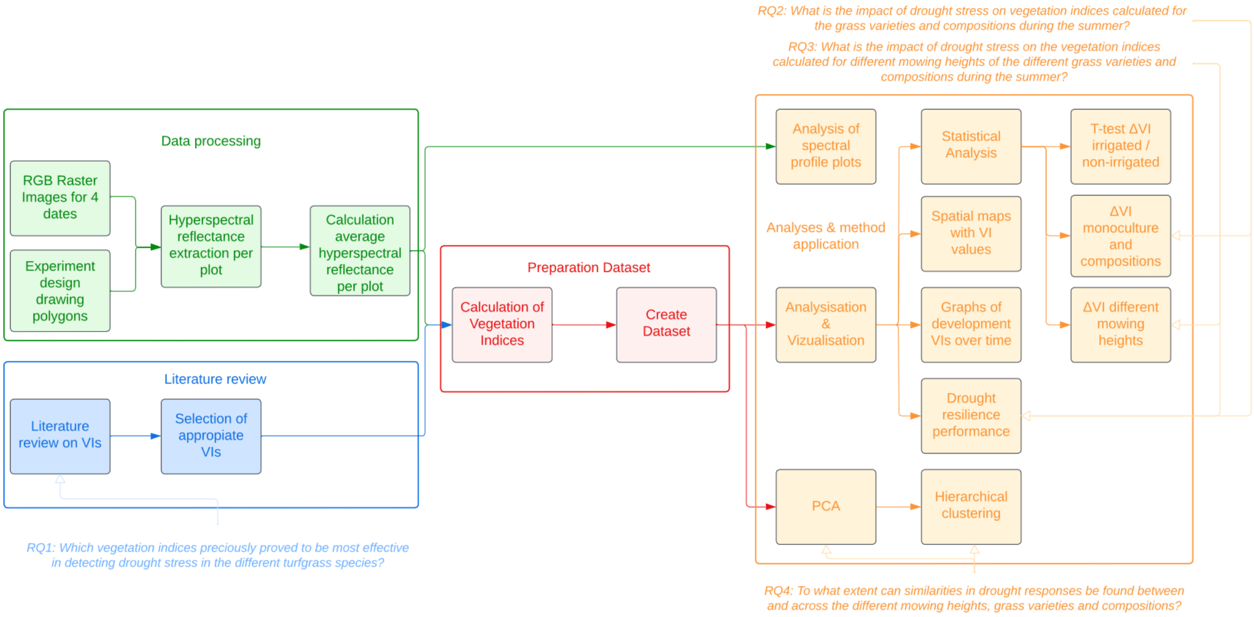


Figure 13: Research methodology workflow

Extraction of plot-level spectral responses

Data processing involves creating Polygon shapefiles for all 240 plots, each of which includes a 10 cm buffer to account for potential imperfections in the imagery and co-registration. These polygons contain information of the research overview in figure 9, linking the plots to the corresponding numbering and the codes of the species. The average measured reflectance for different spectral bands within each plot is calculated based on the four aerial images. This approach aligns with the practices observed in several studies (Wang et al., 2015; Bayat, van der Tol & Verhoef, 2016; Klein et al., 2008), as selecting average values offers a balanced representation of the data.

In this research, not all plots were included due to an issue observed during the experiment. The northernmost non-irrigated plots unintentionally received irrigation because of a slope in the experiment site (Figure 14). These plots are excluded from the calculations and the dataset. Therefore, species A, B, C, and D each three plots included for the 3cm height, while species A, B, C, D, G, H, L, and S, have two plots included for the 6cm mowing height. As a results, certain species are represented by three plots for comparison, while others represented by two.

Compilation of dataset for analysis

The selection of suitable VIs are calculated for the averaged HS reflectance per plot. This calculation involves applying the equation of each VI, which is calculated based on the reflectance values for the specific spectral bands required per plot. For instance, the Structure Independent Pigment Index is calculated using the following equation: SIPI = (R800 – R445) / (R800 + R680). The 15 VI values are calculated for the plots on the four collection dates, resulting in a dataset of 60 VI values columns for all the observation groups, totalling 215 plots.

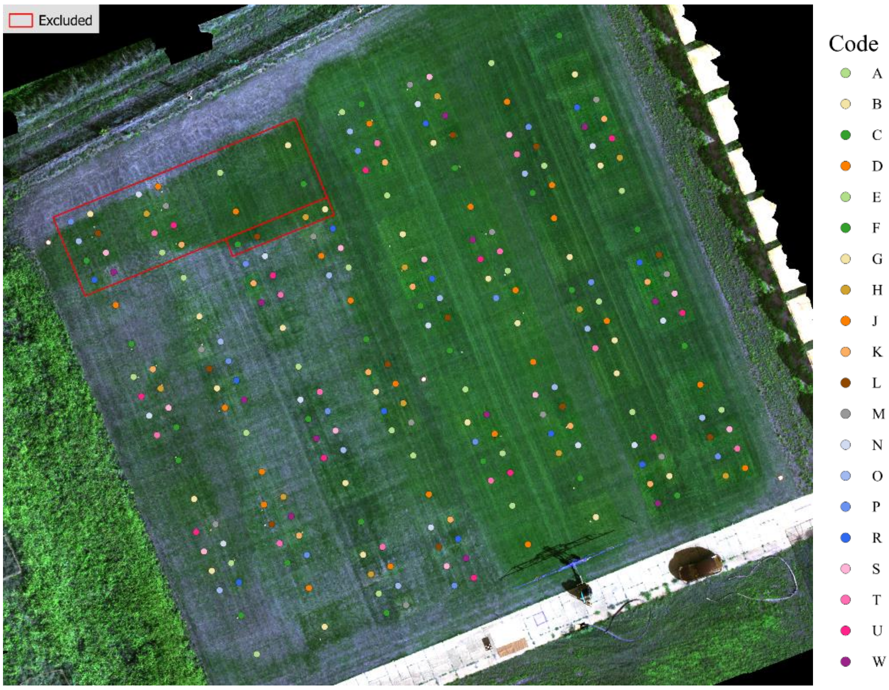


Figure 14: Excluded plots from the research due to a slope causing water flow to non-irrigated area, also shown by greener plots

Analysis of VI values across collection dates for drought stress

The dataset is analysed with a wide range of methods. First the signature reflectance is analysed. This is done by plotting the signature reflectance of the irrigated and non-irrigated plots separately for the four different dates. The resulting graphs show the general development of the measured reflectance for the two groups. Additionally, the VI values for the grass species, mixtures and mowing heights are analysed. This is done by visualizing the development over time by graphs and by maps spatially over time. In both the differences between the observation groups can be analysed.

Statistical t-tests are used to research whether the VI values indicate significant differences between irrigated and non-irrigated plots for each label linked to the mowing height. These t-test are performed on the VI values of all four collection dates. Based on these tests, it can be shown which VIs are able to detect drought stress and to what extent.

To understand the extent to which drought affects the health status of the grasses, the percentage of change in the VI values is calculated. This is done by determining the difference between the calculated VI values on the four dates and the percentage of change relative to the value of the first date, 2nd of June, is calculated. A lower percentage of change indicates that the grass was less affected in its health status due to drought. Ranks from 1 to 40 are assigned to the labels and mowing height per VI value as a normalisation method. The labels and mowing height are ranked relative to each other, with the observation group experiencing the least change ranked as 1 (most drought-resistant) and the one with the most change ranked as 40 (least drought-resistant). The final ranking is determined by the average score of these 15 rankings. The results of this ranking are shown in Table 7. This is done for the periods 2-6 to 18-8 and 2-6 to 30-8.

Cluster analysis

Clustering have proven to be a valuable tool in data science. In fields were users and research work with large datasets of spectral data and images, clustering algorithms prove to be a valuable approach for efficiently reducing vast amount of multi-dimensional data (Ralambondrainy, 1995). These methods allow discovering cluster structures within a data set by creating clusters that link data characterized by the greatest similarity and distinguishing clusters by the greatest dissimilarity (Sinaga & Yang, 2020). In this research, the nonparametric hierarchical clustering approach is employed, which avoids makings assumptions about the underlying data distribution and aims to identify patterns or structure within the data (McLachlan & Basford, 1988). In this research, PCA analysis is combined with hierarchical clustering. PCA is used to recognize patterns by dimensional reduction and this statistical method provides with these advantages to be conducted with clustering methods as hierarchical clustering (Jafarzadegan, Safi-Esfahani and Beheshti, 2019; Kaufman & Rousseeuw, 1990).

Principle Component Analysis (PCA) analysis and hierarchical clustering are employed in this research to identify similarities in drought stress responses among the observation groups. The dataset created provides information on drought stress responses. The combination of PCA with hierarchical clustering is used to analyse the extent to which similarities in drought responses can be found between and across the different mowing heights, grass varieties and compositions.

The PCA analysis is conducted with the packages 'FactoMineR' and 'factoextra' in R and is done before the clustering. The PCA analysis improves the clustering results, since it helps with noise reduction (Ding & He, 2004) and transforms high dimensional data into lower dimensional data, helping to detect coherent patterns more easily (Jolliffe, 2002). Additional hierarchical clustering is used. Hierarchical clustering methods create clusters by organizing the data into levels that resemble a hierarchy (Reddy & Vinzamuri, 2013). This clustering is performed top bottom, with the package 'hclust' and in cut in eight clusters with the package 'cutree'.

5. Results

Building on the methodologies outlined earlier, this chapter present the outcomes of the conducted analysis into drought stress responses of turfgrass. Through a systematic analysis of spectral signatures and calculated VI values across single varieties, mixtures, and mowing heights, this section provides crucial information into the drought resilience of the selected turfgrass species.

5.1 Spectral signatures of grass undergoing drought stress

To analyse the influence of drought on the measured reflectance, the signature reflection of the irrigated and non-irrigated plots is analysed for the four collection dates. In the figure 16 below, the averaged signature reflectance's for the irrigated plots on the four different collection dates are presented. The signature reflectance's evolve over the summer, with the measured reflectance under the 700 nm remaining relatively consistent, while the measured reflectance above 700 nm diverges more significantly. Particularly noticeable are the more diverged patterns observed in the graphs for the dates 18-8 and 30-8.

Figures 17 present the averaged signature reflectance's for the non-irrigated plots on the four different collection dates. Distinct patterns emerge here as well. Reflectance's measured under 700 nm flattens out due to less reflectance measured for green wavelengths from 500 to 600 nm and higher reflectance for red wavelengths from 600 to 700 nm. The measured reflectance above the 700 nm decreases for almost all observation groups. Additionally, the signatures exhibit more diverse results, indicating that certain species react differently to drought compared to others.

5.2 VI values calculated throughout the experiment

After obtaining the average reflectance values of each plot, the 15 VIs known for their ability in detecting drought stress are calculated for the species, mixtures and mowing heights. These VI values are analysed through the creation of spatial maps and graphs, enabling the analysis of their changes throughout the research period.

VI values through spatial maps

The figures 15 below show VI values calculated for the four collection dates. This shows clearly the differences in VI values calculated for the irrigated ad non-irrigated plots throughout the experiment. SIPI and CTR2 indexes were chosen due to their significant contribution to explaining variance in the subsequent PCA analysis (Appendix D). For the non-irrigated plots, these maps show a substantial decrease in SIPI and a substantial increase in CTR2. Conversely, the irrigated plots exhibit consistent values for both indexes during the experiment. This shows the impact of the drought treatment on the non-irrigated plots. Comparing the maps for the dates 16-6 with 18-8 reveals a substantial decrease in values for the non-irrigated plots, excluding the most northern plots where the research contains an error in slope that allows the water from the irrigated part to reach to these non-irrigated plots. This is the reason these plots were excluded from the analysis. Analysing the maps, we can see that the VI values for the 6cm plots keep more consistent across the experiment. Furthermore, it can be observed that the VI values of the same labels decrease more than others. This indicates that the different species, mixtures and mowing heights react differently to the drought.

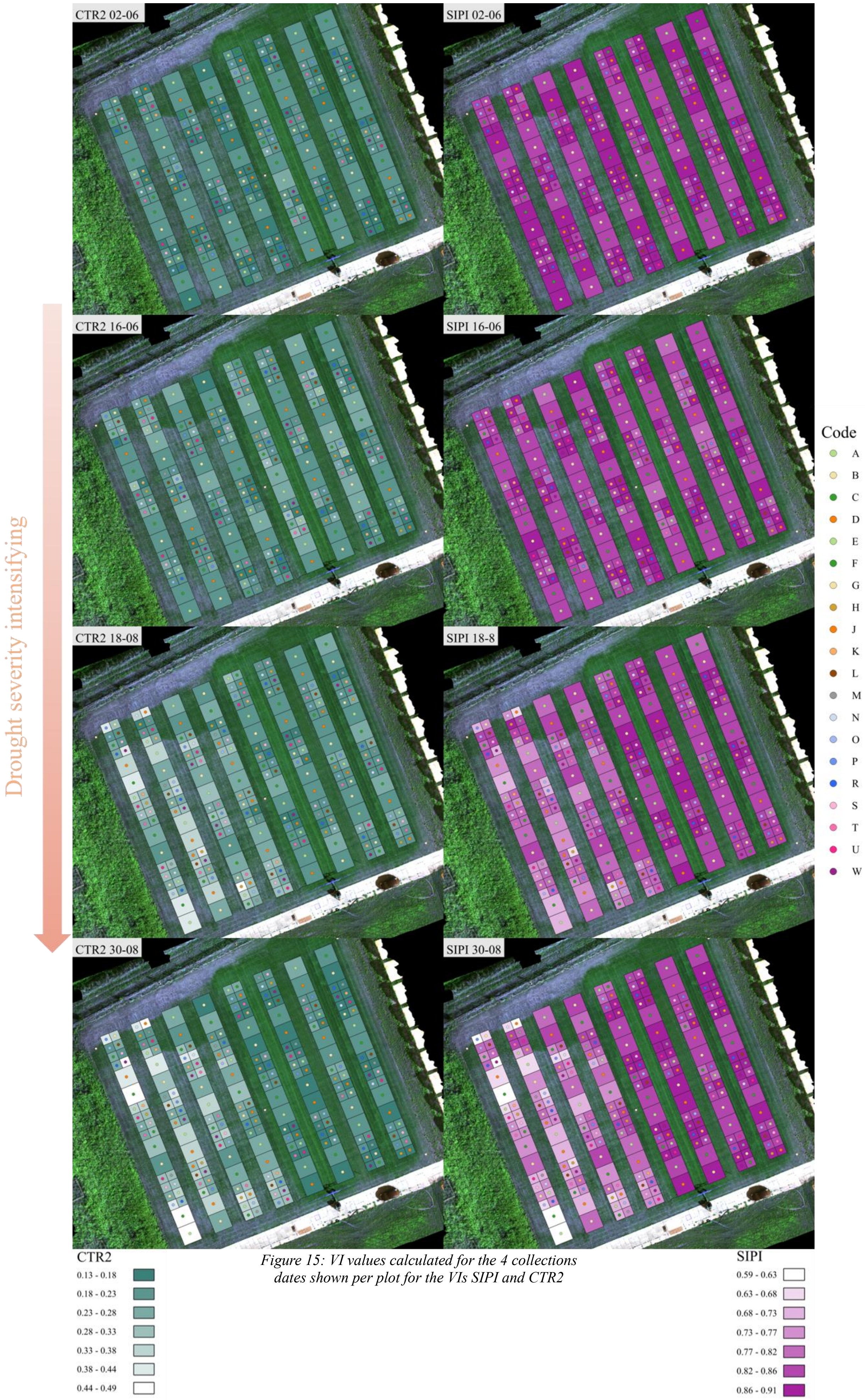


Figure 15: VI values calculated for the 4 collections dates shown per plot for the VIs SIPI and CTR2

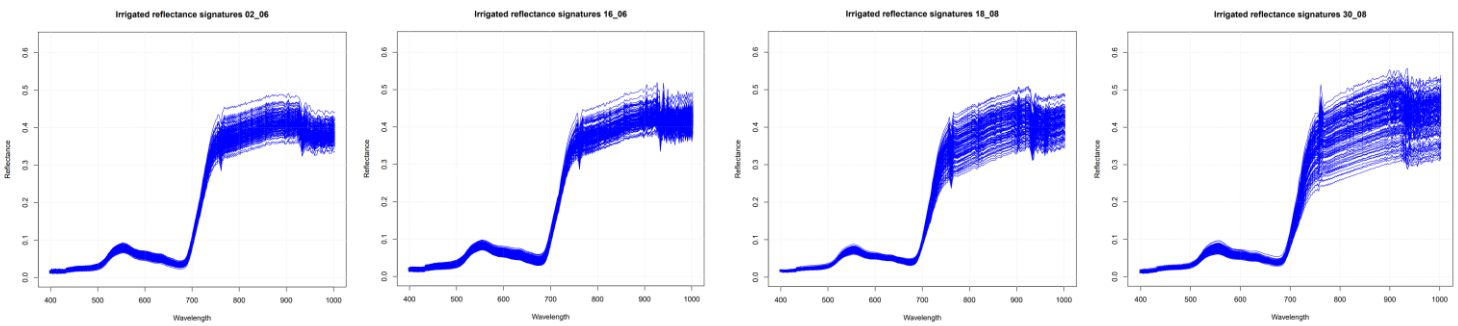


Figure 16: Signature reflectance for the irrigated species, mixtures and mowing heights

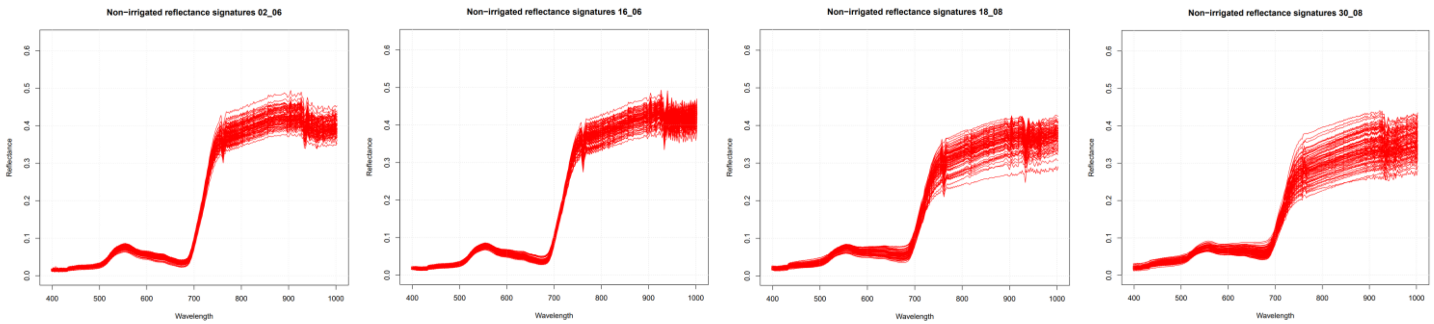


Figure 17: Signature reflectance for the non-irrigated species, mixtures and mowing heights

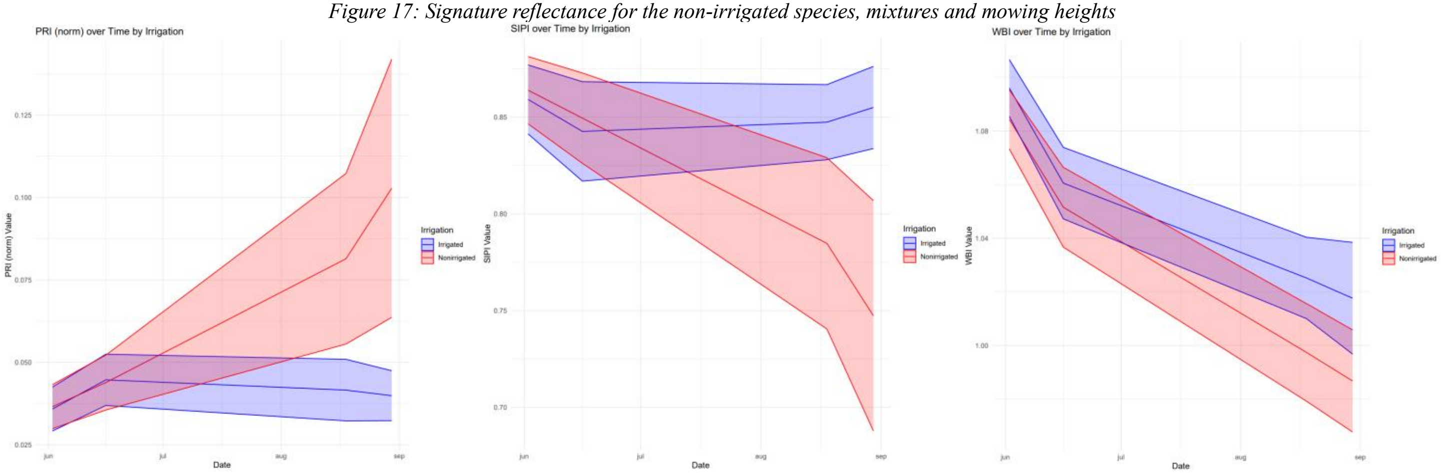


Figure 18: Development of VI values throughout the experiment for irrigated and non-irrigated species, mixtures and mowing heights

VI values through graphical plots

Figures 18 below depict the development from the indices GRI and PRI(norm), serving as examples (Figure 23 in Appendix E proves the development of all VIs). Analysing the average and standard deviation of the irrigated plots shows that the index values remain relatively consistent over time, with individual variations. This is expected since these plots were irrigated and not subjected to drought stress. Analysis of the average and standard deviation of the non-irrigated plots reveal significantly greater differences, especially between 16-6 and 18-8, indicating significant differences associated with experienced drought stress. The VWC (%) decreased during this period due to the high temperatures and lack of precipitation. This drought stress persisted from 18-8 to 30-8, resulting in further alterations in the index values. In conclusion, the graphs distinctly highlight the differences in the calculated VI values between the irrigated and non-irrigated grasses, demonstrating the impact of the drought treatment. The standard deviation shows the variance in response among the grass species.

5.3 Statistical analysis

Further analysis of the VI values involves conducting statistical tests. Statistical t-test have been used to research if the VI values for the irrigate plots differ significantly from the non-irrigated plots per species per mowing height. These t-test have been conducted for the VI values on all the four different collection dates. Subsequently, analyses were caried out to assess the variations among single varieties, mixtures and mowing heights.

Difference irrigated and non-irrigated

These results of the t-tests, presented in Table 6, highlight significant differences in five out of the fifteen VIs on the 2nd of June, indicating notable differences between the VI values of non-irrigated and irrigated plots. This is the case for the indexes SIPI, BRI2, WI.NDVI, NWII and WBI. The VIs SIPI and BRI2 show a significant difference with a P value < 0.05, while WI.NDVI, NWII and WBI show significant differences with a P value <0.001. Additionally, WI.NDVI, NWII and WBI show significantly differences for all the four collection dates. Notably, these are also the only three indexes which are sensitive to plant water concentration. The significance level of the difference decreased for all five VIs from the 2nd of June to the 16th of June. Additionally, all the VIs measured significant differences with a p value <0.001 on the collection dates 18th of August and 30th of August.

Table 6: Calculated differences between mean irrigated and non-irrigated VI values are depicted for each of the 15 VIs across four collection dates. T-tests were conducted to assess the significance of differences within the same observation groups.

VI's	GRI	PRI (norm)	CARI	PRI512	CarRE opt	CCRI	SIPI	BRI2	ChlRE opt	CTR2	mSR705	RGI	WI/NDVI	NWI1	WBI
Sensitive to:	*Anthocyanin and Chlorophyll*	*Carotenoids*	*Carotenoids*	*Carotenoids*	*Carotenoids and Chlorophyll*	*Carotenoids and Chlorophyll*	*Carotenoids and Chlorophyll*	*Chlorophyll*	*Chlorophyll*	*Chlorophyll*	*Chlorophyll*	*Chlorophyll*	*Plant water concentration*	*Plant water concentration*	*Plant water concentration*
2-6-2022	0.004	0.000	0.111	0.005	0.247	0.015	0.007 *	-0.011 *	0.069	-0.006	0.049	-0.002	-0.023 ***	0.005 ***	-0.011 ***
16-6-2022	0.036	-0.002	0.115	0.006	0.236	0.009	0.009	-0.009	0.099	-0.012	0.093	-0.013	-0.028 **	0.003 **	-0.007 **
18-8-2022	-0.568 ***	0.043 ***	-0.590 ***	-0.059 ***	-0.986 ***	-0.062 ***	-0.068 ***	0.061 ***	-0.426 ***	0.082 ***	-0.417 ***	0.222 ***	0.156 ***	0.015 ***	-0.030 ***
30-8-2022	-0.795 ***	0.067 ***	-1.059 ***	-0.101 ***	-1.678 ***	-0.176 ***	-0.115 ***	0.131 ***	-0.550 ***	0.117 ***	-0.502 ***	0.309 ***	0.278 ***	0.016 ***	-0.033 ***

* p-value <0.05, ** p-value < 0.01, *** p-value <0.001

Difference between single variety, mixtures and mowing height

The dataset on the calculated VIs across the collection dates provides data on the drought resilience of single varieties versus mixtures, as well as the impact of mowing height comparing 3cm to 6cm. Figures 19 show the averaged response for each category with a shaded semi-transparent line showing the deviation. The findings show outcomes for the VI PRI(norm), the graphs for the other VIs are presented by Figure 24 in Appendix F. An analyse of the different mowing heights indicates that the 6cm plots exhibit less variance in the calculated VI values and thus are less impacted by drought than the 3cm plots, a pattern observed in both single varieties and mixtures. When comparing the outcomes of single varieties to mixtures, the data reveals broadly comparable outcomes. However, single varieties tend to exhibit a slightly larger range of deviation, particularly in the plots with a 6cm mowing height.

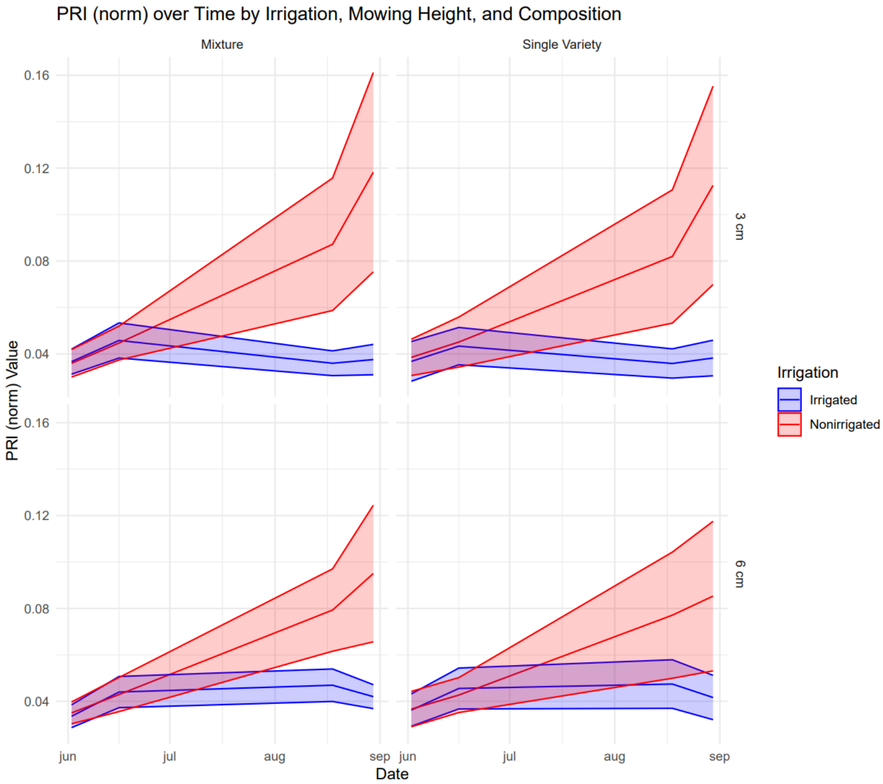


Figure 19: Average response with deviation for single variety and compositions and mowing height

5.4 Drought resilience performance

In order to analyse the drought resistance of all different grass species, mixtures and mowing heights, this study calculates the percentage of change in VI values across the collection dates. The tables in Appendix B display these percentage of changes. The results highlight differences between the non-irrigated observations, indicating varying responses of the observation groups, to drought stress. Substantial differences between irrigated and non-irrigated plots highlight the impact of drought stress, with non-irrigated plots showing substantial higher percentages of change. The table 7 below presents the ranking of the most drought-resistant species and mowing combinations, based on the averaged ranking score. All the explanations provided below refer to Table 7.

Mowing height

The results reveal interesting differences between the two mowing heights. In general, plots mowed at the height of 6cm perform better under drought conditions than plots mowed at 3 cm. The 6cm of each label also perform better overall than the 3 cm plots. This trend is emphasized when comparing the rankings of 2-6/18-8 with the rankings of 2-6/30-8. 6cm plots exhibit better performance than the 3cm plots for longer severe drought durations. An exception to this trend is observed in label J, where Frc 3cm performs better than Frc 6cm.

Species

Analysing the single variety that are most drought resistant shows that labels B6cm, M6cm, and G6cm are highest ranked. These labels include species Fa and Fo, making them the most drought-resilient species. Other high- ranked labels are E6cm, B3cm, K6cm and G3cm, indicating the drought resilience of the species Lp di and Frt mown at 6cm and mown at Fa 3cm height. The comparison of rankings across the two periods reveals minor differences, with only species only changing a few positions. Throughout both periods, Fa6cm consistently emerges as the most drought-resilient, closely followed by Fo6cm. The least drought-resilient labels are C3cm and H3cm, indicating that Lp te and Pp mown at 3cm height are most affected by drought. Other low-ranked labels are L3cm, K3cm and J6cm, indicating that the species Frr and Frt mown height at 3cm and Frc mown at 6cm are also substantially impacted by drought. A comparison of rankings between the two periods shows that Frt 3cm experienced more pronounced effects from drought in the final two week of the study.

Single varieties and mixtures

A comparison of rankings between single varieties and mixtures reveals that Label R6cm, including 50% Fa and 50% Pp, is the only mixture ranked in the top 6, suggesting that mixtures are not as drought resilience as single varieties. Other mixtures ranked high include D6cm and A6cm, suggesting that Pp6cm and Lp di6cm, following closely behind Fa, excel in mixtures compositions. Moreover, Frc 6cm and Frt 6cm also show strong performance in mixtures. Notably, Fa performs well in both single variety and in mixtures, whereas Fo is primarily performing well in a single variety.

Mixtures most affected by drought stress include labels O and A, including Lp te 3cm, Pp 3cm and Lp di 3cm. Notably, the single variety Lp te 3cm and Pp 3cm are also highly influenced by drought. Other low-ranked labels are D3cm and U3cm, containing Lp di 3cm, Pp 3cm, Frc 3cm, Frt 3cm and Frr 3cm. Pp 3cm performs poorly in mixtures, while Pp 6cm performs well in mixtures.

Differences in drought resilience ranking: June 2 – August 18 vs. June 2 – August 30

The grass experienced the drought two weeks shorter for the period 2-6 to 18-8 than the period from 2-6 to 30-8. The first period, lasting 36 days, is considered long-term drought stress, matching the definition for such conditions. In contrast, short-term drought stress refers to duration of up to 11 days. This research includes two different assessments of long-term drought conditions. As mentioned earlier, there is minimal variation between the scores of the two rankings. Most of the top 10 remains the same, with only three rankings changing. A more notable decrease in ranking is seen for labels M3cm and K3cm, indicating Fo3cm and Frr3cm were more influenced by drought compared to the other species. And a more notable increase is seen for labels J 3cm and L 6 cm, showing that Frc3cm and Frr6cm perform better under the longer drought conditions.

Table 7: Ranking non-irrigated for the periods 2-6 to 18-8 and 2-6 to 30-8. Mixtures are color-coded: blue for sports/events, pink for park/recreation, and purple for roadside. Additionally, shading differentiates between the two mowing heights and distinguishes single varieties from mixtures and the observation groups are categorized into three resilience level groups of equal size

Ranking 2-6 / 30-8	Averaged score 2-6 / 30-8	Ranking 2-6 / 18-8	Averaged score 2-6 / 18-8	Species	Species name	Mowing height	Single variety / mixtures	Composition of species	Resilience level
1	2	1	2	B	Tall Fescue	6 cm	SV	100% Fa	+++
2	5	2	4	M	Hard Fesue	6 cm	SV	100% Fo	+++
2	5	5	8	R	Sport/events	6 cm	M	50% Fa, 50% Pp	+++
4	7	3	6	G	Tall Fescue	6 cm	SV	100% Fa	+++
4	7	6	9	E	Perennial Ryegrass diploid	6 cm	SV	100% Lp di	+++
4	7	3	6	B	Tall Fescue	3 cm	SV	100% Fa	+++
7	8	10	13	D	Red Fescue (park)	6 cm	M	20% Lp di, 20% Pp, 30% Frc, 30% Frt	+++
8	9	6	9	A	Perennial Ryegrass (sport)	6 cm	M	50% Lp di, 50% Pp	+++
9	10	9	12	K	Red Fescue Trichophylla	6 cm	SV	100% Frt	+++
9	10	8	10	G	Tall Fescue	3 cm	SV	100% Fa	+++
11	12	13	14	S	Park/recreation	6 cm	M	35% Lp di, 50% Pp, 15% Frt	+++
12	13	13	14	C	Perennial Ryegrass Tetraploid	6 cm	SV	100% Lp te	+++
13	16	13	14	T	Park/recreation	3 cm	M	20% Lp di, 25% Fa, 25% Pp, 10% Frc, 10% Frt, 10% Frr	+++
13	16	16	16	P	Sport/events	3 cm	M	50% Lp di, 50% Fa	+++
15	17	10	13	T	Park/recreation	6 cm	M	20% Lp di, 25% Fa, 25% Pp, 10% Frc, 10% Frt, 10% Frr	++
15	17	17	17	W	Roadside	3 cm	M	20% Frc, 10% Frt, 20% Frr, 50% Fo	++
17	18	19	19	P	Sport/events	6 cm	M	50% Lp di, 50% Fa	++
17	18	10	13	M	Hard Fesue	3 cm	SV	100% Fo	++
19	19	19	19	U	Park/recreation	6 cm	M	20% Lp di, 20% Pp, 20% Frc, 20% Frt, 20% Frr	++
20	20	18	18	W	Roadside	6 cm	M	20% Frc, 10% Frt, 20% Frr, 50% Fo	++
21	23	31	29	J	Red Fescue commutate	3 cm	SV	100% Frc	++
21	23	25	24	R	Sport/events	3 cm	M	50% Fa, 50% Pp	++
21	23	21	20	F	Perennial Ryegrass tetraploid	6 cm	SV	100% Lp te	++
24	24	25	24	N	Sport/events	6 cm	M	50% Lp di, 50% Pp	++
24	24	23	23	N	Sport/events	3 cm	M	50% Lp di, 50% Pp	++
26	25	35	33	L	Red Fescue Rubra	6 cm	SV	100% Frr	++
26	25	32	30	H	Kentucky Bluegrass	6 cm	SV	100% Pp	++
28	27	22	22	E	Perennial Ryegrass diploid	3 cm	SV	100% Lp di	+
28	27	28	28	S	Park/recreation	3 cm	M	35% Lp di, 50% Pp, 15% Frt	+
28	27	27	27	F	Perennial Ryegrass tetraploid	3 cm	SV	100% Lp te	+
31	28	28	28	J	Red Fescue commutate	6 cm	SV	100% Frc	+
31	28	23	23	K	Red Fescue Trichophylla	3 cm	SV	100% Frt	+
33	30	28	28	O	Sport/events	6 cm	M	50% Lp te, 50% Pp	+
34	31	32	30	L	Red Fescue Rubra	3 cm	SV	100% Frr	+
35	34	34	31	U	Park/recreation	3 cm	M	20% Lp di, 20% Pp, 20% Frc, 20% Frt, 20% Frr	+
36	35	35	33	D	Red Fescue (park)	3 cm	M	20% Lp di, 20% Pp, 30% Frc, 30% Frt	+
36	35	39	38	H	Kentucky Bluegrass	3 cm	SV	100% Pp	+
38	38	37	36	A	Perennial Ryegrass (sport)	3 cm	M	50% Lp di, 50% Pp	+
39	39	38	37	C	Perennial Ryegrass Tetraploid	3 cm	SV	100% Lp te	+
39	39	39	38	O	Sport/events	3 cm	M	50% Lp te, 50% Pp	+

5.5 PCA Analysis & Hierarchical clustering

In this section the results from the PCA analysis and hierarchical clustering are shown on non-irrigated plots over the period from June 2nd to August 30th.

PCA analysis

A PCA analysis was conducted to reduce the dimensions of the dataset and find similarities within the data. Following the elbow method, two principal components where chosen, which collectively accounted for 88.8% of the total variance. This significant level of explained variance suggests that these components effectively capture the underlying structure of the dataset. The variable contributions to these components were analysed, showing that the indices SIPI, PRI512, CCRI, CARI, PRI(norm), RGI, CTR2 and ChREopt most important in explaining the variance. These variables were the main factors distinguishing data from one another. The results of the scree plot and the contribution of the variables are included in Appendix C.

Figures 20 show three biplots made, categorized on the drought resistance ranking (discussed in last section), Mixtures & Single variety and 3cm and 6cm mowing height. For explanation on coding, refer to Table 7. The analysis of these biplots indicates that the groups with higher resistance, such as B6cm and M6cm, appear on the right side of the X-axis, while the less resistant groups, O3cm and C3cm, appear on the left side. The biplots also effectively cluster the groups by mowing height, indicating that mowing height results in similar characteristics in means of drought stress response. Contrary to this pattern, labels F, J and N cluster opposite, with 3cm plots aligning closer to 6cm plots and other way around.

The biplots also reveal that single varieties are more widely spread, indicating substantial differences in drought stress response among them. The labels B, G and M, representing the most drought resistant species Fa and Fo, cluster together for both 3cm and 6cm plots. Label C3cm, representing the least drought resistant Lp te, does not cluster with other single varieties. Among Red Fescue varieties, labels J, K and L show similarities in the 3cm plots. However, at 6cm, these labels are plotted further apart, with labels K and L being more similar to each other compared to J.

When comparing single varieties to mixtures, the mixtures are clustered more closely together, with sport/events and park/recreation mixtures forming distinct groups. In the sports and events mixtures, labels R, P and N cluster together, with R being the most drought resistant. Labels A3cm and A6cm, along with O3cm, are outliers. For park/recreation mixtures, S and T exhibit many similarities, whereas U is less drought tolerant and plotted further from S and T.

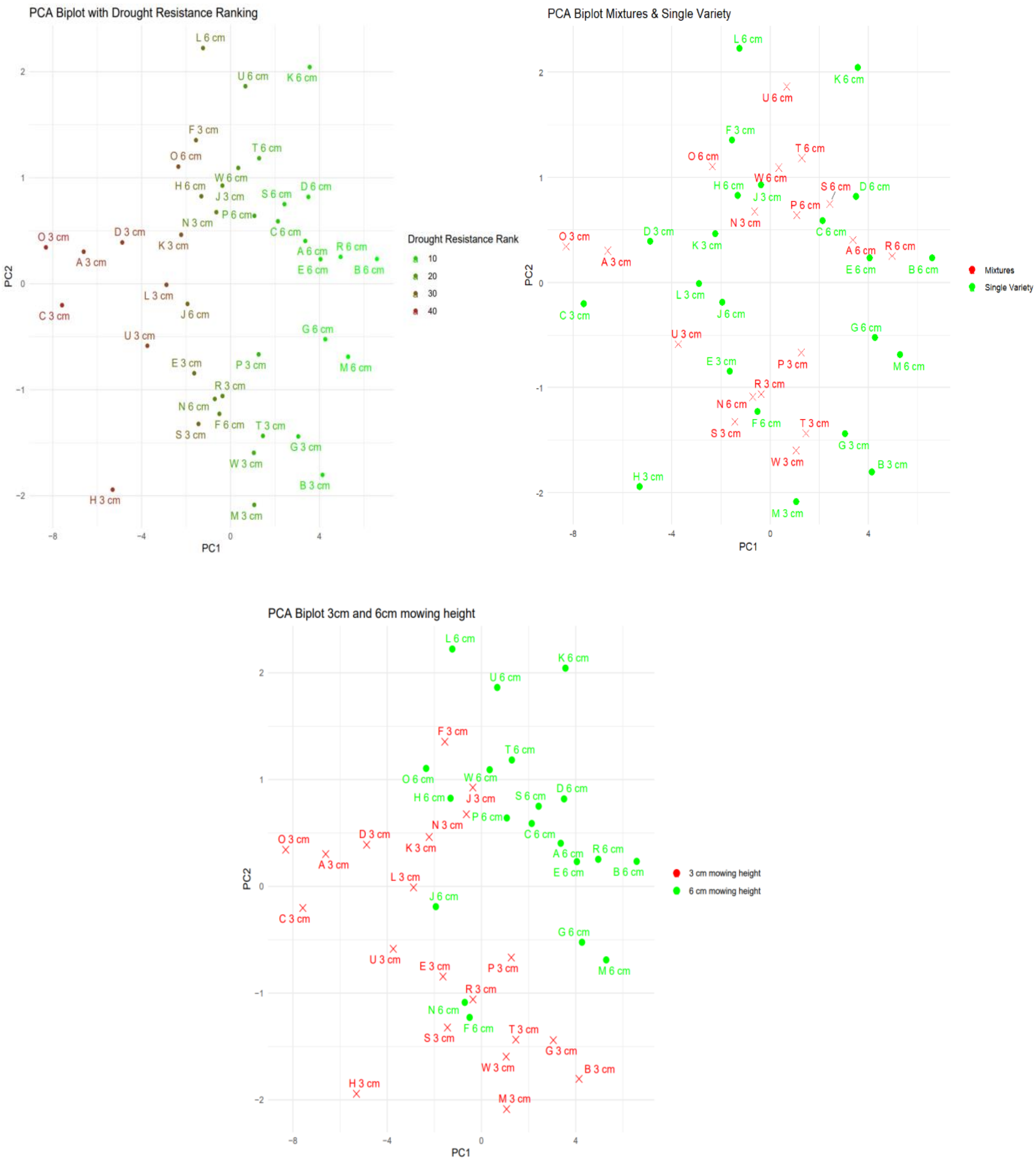


Figure 20: Biplots categorized on Drought Resistance Ranking, Mixtures & Single Variety and two mowing heights

Hierarchical clustering

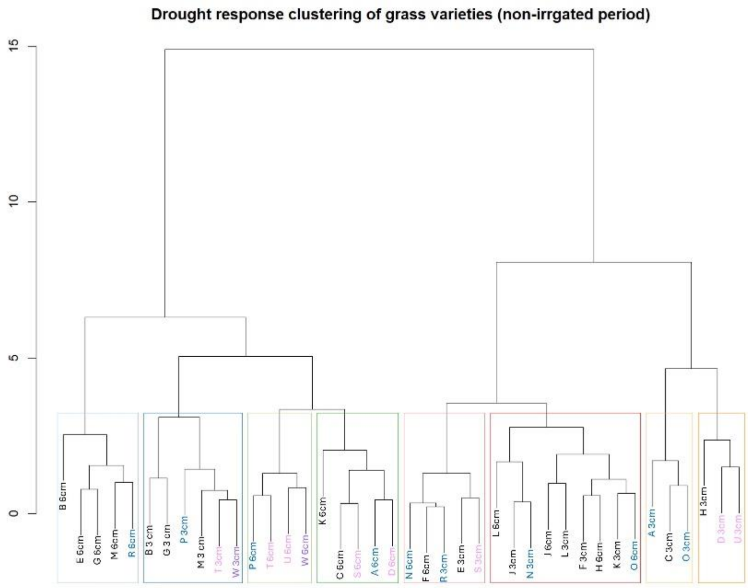
To get a better understanding how the observation groups can be clustered together, a hierarchical clustering has been conducted. The tree has been cut in 8 clusters (Figure 21). Looking at the hierarchy, the left clusters is the best performing cluster under drought conditions, including the drought resilience ranking from 1 to 20. The right cluster is the least drought resilience cluster, including the rankings 21 to 40.

Left side cluster – Best performing under drought conditions

The grey cluster groups the species that are most resistant to drought, with B6cm standing out as the most drought-resistant among these. Interestingly, all the observation groups in this cluster are mown to a height of 6 cm, with R6cm being the only mixed variety. In the dark blue cluster, the most drought-resistant species mown at 3cm are shown. B3cm and G3cm, both single varieties, are most drought resilient and grouped together. The light green cluster consists exclusively of 6cm plots that are all mixtures. The dark green cluster includes a mix of 6cm mixtures and single varieties, which is the second most resistant cluster. This cluster also contains the most resistant mixtures after R6cm.

Right side cluster – Worst performing under drought conditions

The pink cluster includes lower-ranked 6cm plots as well as the second-highest clustered 3cm plots, including both mixtures and single varieties. The red cluster, which is the largest, contains nine observation groups and includes the lowest-ranked 6cm plots. Within this cluster, L6cm is hierarchically the most distant from the other 6cm plots, leaving J6cm, H6cm and O6cm closely grouped as the least drought-resistant 6cm plots. The light orange cluster, marked by the worst performance in drought resilience, includes exclusively 3cm plots, with C3cm and O3cm scoring the lowest in the ranking and clustered together. And as last, the orange cluster ranks as second worst in terms of performance, also consisting solely of 3cm plots.



Code	Mowing height	Species name	Single variety / mixtures	Ranking score	Resilience level	Averaged ranking cluster
B	6 cm	Tall Fescue	S V	1	+++	3
E	6 cm	Perennial Ryegrass diploid	S V	5	+++	3
G	6 cm	Tall Fescue	S V	4	+++	3
M	6 cm	Hard Fesue	S V	2	+++	3
R	6 cm	Sport/events	M	2	+++	3
B	3 cm	Tall Fescue	S V	6	+++	13
G	3 cm	Tall Fescue	S V	9	+++	13
P	3 cm	Sport/events	M	14	++	13
M	3 cm	Hard Fesue	S V	18	++	13
T	3 cm	Park/recreation	M	13	+++	13
W	3 cm	Roadside	M	15	++	13
P	6 cm	Sport/events	M	17	++	18
T	6 cm	Park/recreation	M	15	++	18
U	6 cm	Park/recreation	M	19	++	18
W	6 cm	Roadside	M	20	++	18
K	6 cm	Red Fescue Trichophylla	S V	9	+++	9
C	6 cm	Perennial Ryegrass Tetraploid	S V	12	+++	9
S	6 cm	Park/recreation	M	9	+++	9
A	6 cm	Perennial Ryegrass (sport)	M	8	+++	9
D	6 cm	Park/recreation	M	7	+++	9
N	6 cm	Sport/events	M	23	++	25
F	6 cm	Perennial Ryegrass tetraploid	S V	23	++	25
R	3 cm	Sport/events	M	22	++	25
E	3 cm	Perennial Ryegrass diploid	S V	27	+	25
S	3 cm	Park/recreation	M	29	+	25
L	6 cm	Red Fescue Rubra	S V	26	++	28
J	3 cm	Red Fescue commutate	S V	21	++	28
N	3 cm	Sport/events	M	23	++	28
J	6 cm	Red Fescue commutate	S V	29	+	28
L	3 cm	Red Fescue Rubra	S V	34	+	28
F	3 cm	Perennial Ryegrass tetraploid	S V	29	+	28
H	6 cm	Kentucky Bluegrass	S V	27	+	28
K	3 cm	Red Fescue Trichophylla	S V	32	+	28
O	6 cm	Sport/events	M	33	+	28
A	3 cm	Perennial Ryegrass (sport)	M	37	+	39
C	3 cm	Perennial Ryegrass Tetraploid	S V	39	+	39
O	3 cm	Sport/events	M	40	+	39
H	3 cm	Kentucky Bluegrass	S V	37	+	36
D	3 cm	Park/recreation	M	36	+	36
U	3 cm	Park/recreation	M	34	+	36

Figure 21: Biplots categorized on Drought Resistance Ranking, Mixtures & Single Variety and two mowing heights

6. Discussion

This study examines the response of cool season turfgrass species, mixtures to drought conditions, aiming to identify drought-resistant varieties suitable for use in sports and events, parks and reaction areas, and roadside environments. This chapter includes a discussion section, where the observed patterns in the findings are analysed and connected with existing literature.

6.1 Different grasses and mixtures, different responses to drought

Reflectance patterns in vegetation offer valuable insights into plant health and stress levels, serving as an important tool for monitoring drought stress on turfgrass. The signature reflectance patterns, shown in Figures 16 and 17, exhibit distinct responses between the irrigated and non-irrigated plots. Specifically, irrigated plots maintain consistent reflectance value below 700 nm and exhibit consistent averaged measured reflectance around 0.4 across the different collection dates. In contrast, non-irrigated show an increase in the averaged reflectance around 700 nm and a decrease in the measured averaged reflectance above 700 nm. This leads to a more flattened out signature reflectance curve below 700 nm and a drop in average reflectance measured in the second part. These changes align with drought stress indicators as identified by Bayat, van der Tol & Verhoef (2016), suggesting that our findings confirm with existing literature on drought stress impacts. Another pattern seen is the wide range of reflectance measured across the different observation groups, attributable to the study's diversity in grass species compositions and mowing heights. Previous studies showed that grass species and mowing heights significantly influence reflectance signatures (Caturegli et al, 2014; Lee et al., 2011), which explains the observed variance at the start of the experiment and the continuation as drought impacts the grasses.

This study used these differences in measured reflectance across the collection dates to analyse responses to drought stress. The results of the t-test revealed that five of the fifteen explored VIs showed a significant difference on the 2nd of June. While the maps do not indicate clear differences, Figure 23 Appendix E suggest that for VIs BRI2, SIPI and WI/NDVI the irrigated plots appear closer to drought stress conditions than the non-irrigated plots on June 2nd. Conversely, WBI and NWI1 show the opposite trend. The significance observed on this date cannot be directly attributed to drought stress experienced by the grasses, as drought conditions did not begin until the 14th of July. Additionally, all graphs show a change in VI values for all plots between June 2nd and 16th while there was no drought stress experienced. The differences in the calculated VI values can be contributed to other processes, such as biomass accumulation and / or phenological changes, which influence these values (Masin, Zuin and Zanin, 2005; Meyer, Hoffman & Bonos, 2017).

The analysis of graphs for the different mowing heights and compositions (Appendix F) indicate that mowing height substantially influences drought performance. Specifically, VI values for the 6cm plots show less variation compared to the 3cm plots over the course of the experiment. Additionally, no substantial differences are observed between mixtures and single varieties, suggesting that having a mixture or single variety does not enhance performance under drought conditions. This finding is also supported by the study of Reiter et al. (2017), which reported no observed differences between mixtures and single varieties in retaining green cover rates. It appears that the advantage lies not in the diversity of grass species sown but in the choice of specific species. The variability in standard deviation across the varieties and mixtures can be attributed to the species sown. Essentially, some species outperform others under drought stress, as evidenced by the varied performance results.

6.2 Linking drought-resistance to plant characteristics

The rankings in Table 7 clearly indicate that various species, mixtures and mowing heights each respond uniquely to drought stress, as previously discussed. Primarily, the rankings clearly demonstrate the impact of mowing height on drought tolerance. Grasses mowed at a height of 6 cm consistently outperform those mowed at 3 cm under drought conditions. These results align with previous research indicating that taller turfgrass exhibits increased drought resistance (Braun et al., 2022; Shaba, Abbas & Alshammary, 2014). The longer grass length can be linked to increased carbon fixation and root production. Additionally, taller grass supports the accumulation of proline, a compound that helps in maintaining cellular water balance during stress and nutrient storage, further improving the drought resistance (Shaba, Abbas & Alshammary, 2014). Secondly, the ranking reveals the drought resilience of specific species. The observed drought resistance for each species is linked to specific plant characteristics, such as root systems, leaf width and evaporation rates, as detailed in the literature review.

The literature review indicates that different grass species exhibit different root systems traits, including root depth and root mass distribution. As shown in Figure 12, Perennial Ryegrass and Tall Fescue developed the deepest roots, succeeded by Hard Fescue and Red Fescue. Kentucky Bluegrass exhibited the shallowest roots. Table 3 further presents root depth, root mass, and distribution findings from additional studies, indicating Hard Fescue and Tall Fescue have the deepest average rooting depths (51,3 cm and 50,7, respectively), followed by Perennial Grass (46,6 cm), Red fescue (34,2 cm) and Kentucky Bluegrass (20,9 cm). These findings align with Scheffer (1987) and Lin (1985), who reported that Red Fescue's root mass distribution is shallower compared to Perennial Ryegrass and Tall Fescue, with Tall Fescue rooting deeper than Perennial Ryegrass.

When relating these root characteristics to drought resistance observed in this research, it's evident that species with deeper roots exhibited advanced drought tolerance. Hard Fescue and Tall Fescue, with the deepest roots and substantial higher root mass, were the most-drought resistant. This aligns with existing studies, which found that Hard Fescue is one of the most drought resistant species (Butler et al., 1987). Perennial Ryegrass diploid, mown on 6cm height, also performed well under drought conditions, ranking just below Hard and Tall Fescue. When mowed at 3cm, its performance declines, indicating a less developed root system compared to the other species. Conversely, Red Fescue and Kentucky Bluegrass, with shallower roots, demonstrated lower drought resilience, with Kentucky Bluegrass being the least resistant due to its shallow rooting depth and root mass. Perrenial Ryegrass tetraploid, similar to Kentucky Bluegrass, demonstrates the lowest drought resilience. Since this research does not provide information on root depth differences among variations of species, the observed differences are attributed to other plant characteristics, not root depth.

Leaf width and evaporation rate are also key factors influencing the drought resistance of grass species. According to the results summarized in Table 3, Tall Fescue has the broadest leaves (3-12 m), with Perennial Ryegrass (2-6 mm) and Kentucky Bluegrass (2-5 mm) following. Whereas Hard Fescue (0,6-2.5 mm) and Red Fescue (0.3-1.2 mm) exhibit the narrowest leaves. The order of evaporation rates mirrors this, with Tall fescue and Kentucky bluegrass experiencing the highest rate (>10mm per day), Perennial Ryegrass (8,5-10mm per day), Red fescue (8-8,5 mm per day) and Hard Fescue (7-8,5 mm per day) following. This correlation directly impacts resistance observed in this study. Hard Fescue, because of its narrower leaves and lower evaporation rate, stands out as highly drought-resistant, particularly in the 6cm plots which outperformed most Tall Fescue plots. The lower evaporation rate of Hard Fescue, relative to Tall Fescue, likely provides a subtle advantage, given their similar root depths. Tall Fescue, despite its high evaporation rate, remains among the most drought-resistant species, suggesting that a high rate of water loss does not necessarily disadvantage it greatly during drought conditions. Kentucky Bluegrass, characterized with a high evaporation rate, ranks as less drought resistant. This information contributes further insights alongside root system depth, which is the primary factor explaining drought resilience (Carrow, 1996; Wilman, Gao & Leitch, 1998).

6.3 Contrast among varieties and specifications

This study also explores the drought resilience of different species variations, including two variations of Perennial Ryegrass and three variations of Red Fescue. The drought resistance ranking analysis reveals a substantial difference between Lp di 6cm and Lp te 6cm. For the 3cm mowing height, both variations are more closely related, yet Lp di showing greater drought resistance than the Lp te variation. Furthermore, among Red Fescue variations, Commutata emerges as the most drought-resistant for 3cm plots, while Trichophylla excels at 6cm plots. These results also indicate differences in ideal mowing height for achieving most drought resistance sods.
This difference observed between the Perennial Ryegrass variations can be linked to differences in plant traits (Table 4); Lp di exhibits smaller tillers and thinner leaves, resulting in denser sods, while tetraploids have fewer but larger tillers with broader leaves, resulting in more open sods. Tetraploids are also known to be less persistent (Hannay et al., 1999). Similarly, the differences between the Red Fescue variations are explained by the plant traits of Commutata and Trichophylla, which have narrower leaves, resulting in denser sods, unlike Rubra’s broader leaves that result in more spaced sods (Bals, n.d.). This variation in plant traits suggests that denser dos tend to offer greater drought resistance compared to more open sods.

Lastly, this research focused on turfgrass usage specifications, including sports and events, park and recreation and roadside, each with unique characteristics and maintenance needs. Table 5 provides insight into the drought resistance competence of the mixtures used for specific use. Mixtures containing the species Lp te demonstrate the least drought resistance, indicating their unsuitability for inclusion in drought-resistant compositions. In contrast, Fa and Lp di 6cm show strong performance in mixtures, recommending its use.

The analysis of preferred turfgrass mixtures used for specific applications, such as sports/events and park/recreation, highlights PP as preferred choice due to its dense and even sods. However, cultivating this species has its difficulties leading to the exploration of compositions. Perennial Ryegrass (Lp), known for its rapid growth, is often used in combination with Kentucky bluegrass to enhance the sods performance (Plantum, 2016). Mixtures including Pp and Lp di show drought resistance, although requiring a mowing height of 6 cm. For lower mowing heights of 3cm, mixtures of Lp di and Fa are recommended. In park/recreation use, mixtures featuring Frt and Frr are preferred for their rapid recovery (Plantum, 2016). The analysis indicates that Frt-mixtures outperform the Frr-mixtures in terms of drought resistance.

6.4 Clustering similar drought responses

The PCA biplots and hierarchical clustering offer nuanced insight into the turfgrass drought response, helping to understand the responses beyond the rankings and enhance the understanding of the multidimensional dataset. The PCA biplots show in Figure 20 a distinct clustering based on mowing heights, confirming the found influence of mowing height on drought resilience, with 6cm plots generally outperforming 3cm plots. The wide scatter of single varieties in the biplots indicates substantial differences in drought stress response. This scattering is expected, given the 100% species composition, resulting in substantially larger differences in drought stress responses and a consequent increase in outliers. Among the varieties, substantial differences are observed between E and C & F, respectively Lp di and Lp te, attributable to the differences between closed and open sods. Varieties with open sods, like Lp te, perform worse than those with closed sods, like Lp di. For Red Fescue Varieties J, L and K, greater similarity is observed at the 3cm mowing height than at 6cm. This variation is likely due to more pronounced differences between open and closed sods at the higher mowing height, explaining the observed difference between L, with an open sod, and K and J, which have closed sods. The difference between J and K can be attributed to K being the most drought-resistance of the variations. These similarities and differences are clearer through biplots, whereas the strength of the relationships is less evident in the rankings.

This diversity in drought stress response highlights the critical role of species selection in turf management, especially regarding drought resistance. The clustering of mixtures in the biplot further suggests the strategic advantage of including drought-tolerant species, showing that the composition and proportion of species in mixtures are crucial. Specifically, mixtures for sports/events showed a substantial drought resilience, particularly when composed of the species Fa, Lp di 6cm and Pp, which were closely clustered in the analysis. It also highlights the similarities and differences between species variations, in single varieties as in mixtures. These results suggest a strategic selection of species for improving drought resilience of turfgrass mixtures.

Hierarchical clustering provides additional insights to these findings, clustering the turfgrasses into under drought performance-based groups. This clustering confirmed the importance of choosing the right species and mowing height, as shown by the PCA, and also emphasized how different turfgrass species, mixtures and mowing heights differ in drought resistance. The clustering highlights based on hierarchy the relationships, offering confirmation and additional insights in previously established relationships. For instance, the clustering result shows the distinct drought resistance of the single variety Fo at both 3cm as 6cm, the consistent drought resilience Fa across 3cm and 6cm including its presence in mixtures, the minimal impact of Pp and the Red Fescue varieties on the drought resistance of mixtures, and the notable vulnerability of Lp Te, disregarding included of its inclusion in mixtures or as a single variety. Furthermore, it identifies clusters of single varieties, mixtures and mowing heights, offering guidance for selecting the most drought-resistant turfgrass for different use specifications. Concluding, the clustering analysis indicates the multidimensional nature of drought stress response in turfgrass, emphasizing the species selection, mowing height and mixture composition. It confirms that while individual plant traits are foundational, different maintenance affects their drought resilience. These findings validate the pervious observations but also expand our understanding highlighting the added value of clusters.

6.5 Limitations and further work

This research offers valuable insights into the application of HS remote sensing for assessing drought resistance in grass species and mixtures. However, if faces several limitations worth noting. Firstly, the data collected only allow for the analysis of long-term drought stress, identifiable after 36 days, while short-term drought stress can be detected as early as 11 days according to Bayat, Van der Tol &Verhoef (2016). With drought conditions starting in early July and the subsequent data collection on August 18th, the focus is on grasses subjected to approximately 48 days of drought. This enables this research to study severe drought stress across the grasses, mixtures and mowing heights but limits the ability to research the early stages of drought stress. Further studies could align with this study and conduct the same methodology on early stages of drought, exploring the differences short-and long-term drought effects.

Furthermore, the research employs the Headwall Nano-Hyperspec camera, capturing HS data within the 400 to 1000 nm range. Although this range includes several crucial wavelengths for evaluating vegetation health, important wavelengths of the near-infrared (NIR) water absorption bands around 1440 and 1930 nm are not included (Bayat & Verhoef, 2016). Previous research indicates that the Short-wave infrared wavelengths (0.9 to 1.7 µm) within the optical section of the EM spectrum is the most suitable for analysing water status in plant canopies. This suitability is due to the water band's capacity the incoming radiation is absorbed by the water content in plants (Tucker, 1980; Jiang & Carrow, 2005). Therefore, subsequent research might benefit from including these wavelengths to compare and enrich the findings of this study.

Additionally, applying different irrigation regimes within the same experimental field could provide insight into the specific water needs of different grass species, mixtures and mowing heights. This information could be applied to the maintenance of the grasses for the time they are planted and throughout their use. Previous studies have laid the groundwork for this approach. For example, Bastug & Buyuktas (2003) and Alshehhi et al. (2010) analysed the drought resilience of turfgrass under several irrigation levels, including 100%, 88%, 75% and 50% standard irrigation. The standard irrigation can be attributed to specific applications, such as sports/events and park/recreation. By systematically varying irrigation levels and closely analysing the physiological responses of the grass, this approach could provide insights for efficient water management specifical to the needs of each turfgrass species, mixture and mowing height.

7 Conclusion

This research utilized UAV hyperspectral imagery to identify drought stress and analyse drought resilience across different cool season turfgrass species, contributing to valuable insight into the precision agriculture field.

A thorough literature review identified 28 VIs sensitive to drought stress across 12 relevant studies focused on cool season turfgrass. Of these, 15 VIs were choses based on their proven effectiveness. This research validated the selected VIs' ability to detect and quantify drought stress effects among different grass species, mixtures, and mowing heights under drought conditions. The analysis of VI values across the collection dates reveals that: 1) higher mowing height typically improves drought resistance, 2) negligible differences in drought resistance are observed between single varieties and mixtures, 3) similarities in drought response are primarily associated with root depth and sod structure, 4) mixtures with a higher percentage of drought resistance species exhibit generally the greatest drought resistance, and 5) the species Fa and Fo, at both 3cm and 6cm mowing heights, along with Lp di at 6cm, exhibit the highest drought resistance, while Lp te, at both mowing heights, showed the least resilience, regardless of whether in mixtures or single varieties.

With these findings, this study bridges a knowledge gap by providing results on the drought resilience of turfgrass species, mixtures and mowing heights in already established turfs. This knowledge is of increasing importance as climate change leads to more frequent and severe droughts in the Netherlands and across Europe. The implementation of drought resistant grasses will help maintain its function for specific uses as sports/events and park/recreation during drought conditions.

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9 Appendix A: Relevant articles on detecting drought stress in cool season turfgrass

Table 8: Relevant articles on detecting drought stress response in turfgrass

	Used articles	Duration research	Study location / climate	Type of grass	Width spectral bands -range
1	Badzmierowski, M. J., McCall, D. S., & Evanylo, G. (2019). Using hyperspectral and multispectral indices to detect water stress for an urban turfgrass system. Agronomy, 9(8), 439.	18 May to 26 July 2017 & 6 june to 12 july 2018	Fieldplots Virginia, United States	Tall fescue	320 - 1100 nm 1.4 nm bandwidth 3 nm spectral resolution
2	Bayat, B., Van der Tol, C., & Verhoef, W. (2016). Remote sensing of grass response to drought stress using spectroscopic techniques and canopy reflectance model inversion. Remote Sensing, 8(7), 557.	10 August 2010 & 27 October 2014	Greenhouse, Twente, Netherlands	Poa pratensis Kentucky bluegrass	350 - 2500 nm 1.4 nm bandwidth
3	Caturegli, L., Matteoli, S., Gaetani, M., Grossi, N., Magni, S., Minelli, A., ... & Volterrani, M. (2020). Effects of water stress on spectral reflectance of bermudagrass. Scientific Reports, 10(1), 15055.	1 to 31 May 2018	Greenhouse, Pisa, Italy	hybrid bermudagras	350 - 2500 nm 2 nm interval 3 nm spectral resolution
4	Dao, P. D., He, Y., & Proctor, C. (2021). Plant drought impact detection using ultra-high spatial resolution hyperspectral images and machine learning. International Journal of Applied Earth Observation and Geoinformation, 102, 102364.	17 June to 18 July 2019	Greenhouse, Ontario, Canada	Bromus inermis	400 - 1000 nm 1.8 nm interval
5	Haghverdi, A., Reiter, M., Singh, A., & Sapkota, A. (2021). Hybrid bermudagrass and tall fescue turfgrass irrigation in central California: II. Assessment of NDVI, CWSI, and canopy temperature dynamics. Agronomy, 11(9), 1733.	4 may to 11 Septermber 2018 & 22 June to 26 Augustus 2019	Field plots, Central California, United States	hybrid bermudagras & tall fescue	blue: 430 - 470 nm red 530 - 570 nm NIR 720 - 740 nm
6	Hermanns, F., Pohl, F., Rebmann, C., Schulz, G., Werban, U., & Lausch, A. (2021). Inferring grassland drought stress with unsupervised learning from airborne hyperspectral vnir imagery. Remote Sensing, 13(10), 1885.	7 may 2018 & 23 April 2019	Fieldplot, Saksen-Anhalt, Germany	mixed grassland, not specified	409-989nm 3.2 nm
7	Jiang, Y., Liu, H., & Cline, V. (2009). Correlations of leaf relative water content, canopy temperature, and spectral reflectance in perennial ryegrass under water deficit conditions. HortScience, 44(2), 459-462.	May to Augustus 2007 & June to Augustus 2008	Indiana, United States	Perennial Ryegrass	880 and 650 nm collected with Crop Circle ACS-210
8	Katuwal, K. B., Yang, H., & Huang, B. (2023). Evaluation of phenotypic and photosynthetic indices to detect water stress in perennial grass species using hyperspectral, multispectral and chlorophyll fluorescence imaging. Grass Research, 3(1).	20 days (no month specified)	Controlled environmental growth chamber New Jersey, United States	Kentucky bluegrass (Poa pratensis L.)	400 - 1000 nm 1.9 nm interval
9	McCall, D. S., Zhang, X., Sullivan, D. G., Askew, S. D., & Ervin, E. H. (2017). Enhanced soil moisture assessment using narrowband reflectance vegetation indices in creeping bentgrass. Crop Science, 57(S1), S-	1 to 11 December 2015 & 1 to 11 February 2016	Green house, Virginia, United States	creeping bentgrass	320 - 1100 nm 1.4 nm interval
10	Ervin, E. H., Askew, S. D., & McCall, D. S. (2021). Improving soil moisture assessment of turfgrass systems utilizing field radiometry. Agronomy, 11(10), 1960.	June to September 2018	Greenhouse, Virginia, United States	creeping bentgrass & Hybrid bermudagrass	320 - 1100 nm 1.4 nm interval
11	Jiang, Y., & Carrow, R. N. (2005). Assessment of narrow-band canopy spectral reflectance and turfgrass performance under drought stress. HortScience, 40(1), 242-245.	10 July to 25 July 2001, 20 to 31 Augustus 2001 & 10 to 21 September 2001	Field plots, Georgia, United States	Bermudagrasses, Seashore paspalums, zoysiagrass, st. augustinegrass & tall fescues	400 - 1100 nm 3 nm interval
12	Jiang, Y., & Carrow, R. N. (2007). Broadband spectral reflectance models of turfgrass species and cultivars to drought stress. Crop science, 47(4), 1611-1618.	10 to 25 july, 20-31 Augustus, & 10-21 September	Field plots, Georgia, United States	Bermudagrass, seashore paspalum, zoysiagrass, St. Augustinegrass and tall fescue	10-15 bandwith centrered around 660, 710, 810, 900, 950, 1200 and 1480 nm

10 Appendix B: Identified VIs sensitive to drought stress

Table 9: Identified VIS sensitive to drought stress found in the 12 relevant studies

VI	Full name	Equations	Sensitive to	Results sumarry	Which papers	Suitable / not suitable	Suitable for
ARI	Anthocyanin Reflectance Index	ARI = 1/R550 − 1/R700	Anthocyanin	4. not detecting early drought stress 4. all VIs detected longterm drought stress - (dramatic change) (need research abilities VIs different stages)	4	Not suitable	Not suitable
GRI (RGRI)	Green to red ratio index	R550/R670	Anthocyanin and Chlorophyll	1. can determine water stress 9. significant relationship with TQ (0.001 & 0.01)), Chlorophyll (0.01 & 0.05), SWC (0.01 & 0.05) - not with TWC (0x) 10. 2nd strongest relationship to VWC 0.56	1, 10, 9	Suitable	Early & longterm drought stress
PRI	Photochemical Reflectance Index	PRI = (R570 – R531) / (R570 + R531)	Carotenoids	2. not able detect early drought 2. able to detecht long term drought 4. not detecting early drought stress 4. all VIs detected longterm drought stress - (dramatic change) (need research abilities VIs different stages) 8. for drought stress cool-season turfgrass 9. significant relationship with TQ (2x 0.01), Chlorophyll (2x 0.001), TWC (1 0.01) - not with SWC (0x)	8, 2, 4, 9	Not suitable	Not suitable
PRI (norm)	Normalized Photochemical Reflectance Index	PRI (norm) = PRI/[((R800 ´ R670)/ sqrt (R800 +R670)). R700/R670]	Carotenoids	2. best longterm drought detecting 2. not able detect early drought	2	Suitable	Longterm drought stress
CARI	Carotenoids Index	CARI = (R720–R521)/R521	Carotenoids	4. Could detect early drought stress 4. all VIs detected longterm drought stress - (dramatic change) (need research abilities VIs different stages)	4	Suitable	Early & longterm drought stress
PRI512	Photochemical Reflectance Index 512	(ρ531 − ρ512)/(ρ531 + ρ512)	Carotenoids	6. Best performing for carotenoid/chlorophyll ratio and light use efficiency (LUE) 6. Important for assessing severity of vegetation stress (pigment-related indices)	6	Suitable	Longterm drought stress
CarRE opt	Opt. carotenoid red edge index	$(\rho510–530^{-1} − \rho680–730^{-1}) \times \rho760–780$	Carotenoids and chlorophyll	6. Best performing for Carotenoid content 6. Important for assessing severity of vegetation stress (pigment-related indices) (6. strongest relationship with drought stress)	6	Suitable	Longterm drought stress
CCRI	Carotenoid/ Chlorophyll Ratio Index	CCRI = CARI/CIrededge	Carotenoids anc	4. Could detect early drought stress 4. all VIs detected longterm drought stress - (dramatic change) (need research abilities VIs different stages)	4	Suitable	Early & longterm drought stress
PSRI	Plant Senescence Reflectance Index	PSRI = (R680 –R500) / R750	Carotenoids and chlorophyll	8. for drought stress cool-season turfgrass	8	Not suitable	Not suitable
SIPI	Structure Independent Pigment Index	SIPI = (R800 – R445) / (R800 + R680)	Carotenoids and chlorophyll	8. detect drought stress earliest	8	Suitable	Early drought stress
BGPI	Blue/Green pigment Index 2	R450/R550	Chlorophyll	2. best early drought detection 2. able to detecht long term drought	2	Suitable	Early & longterm drought stress
ChIRE opt	Opt. chlorophyll red edge index	$(\rho680–730^{-1} − \rho780–800^{-1}) \times \rho755–780$	Chlorophyll	6. Important for assessing severity of vegetation stress (pigment-related indices)	6	Suitable	Longterm drought stress
CIred edge	Red edge chlorophyll index	(R750/R710)−1	Chlorophyll	4. could not detect early drought stress 4. all VIs detected longterm drought stress - (dramatic change) (need research abilities VIs different stages)	4	Not suitable	Not suitable
CTR2	Carter Index 2	CTR2 = R695/R760	Chlorophyll	2. second best long term drought detecting 2. able for early dourght detection 6. important predictor for drought stress	2, 6	Suitable	Early & longterm drought stress
MCARI2	Modified chlorophyll absorption ratio index 2	$\frac{1.5(2.5(p800 - p670) - 1.3(p800 - p550)}{\sqrt{(2p800+1)^2} - \left(6p800 - \sqrt[5]{p670}\right) - 0.5}$	Chlorophyll	6. no important predictor drought stress	6	Not suitable	Not suitable
MSAVI2	Modified soil-adjusted vegetation index 2	$\frac{(2pNIR + 1 - \sqrt{(2pNIR+1)^2 - 8(pNIR - pRED))}}{2}$	Chlorophyll	6. no important predictor drought stress	6	Not suitable	Not suitable
mSR 705	Modified Simple Ratio	mSR705 = (R750–R445)/ (R705–R445)	Chlorophyll	4. Could detect early drought stress 4. all VIs detected longterm drought stress - (dramatic change) (need research abilities VIs different stages)	4	Suitable	Early & longterm drought stress
RENDVI / NRred edge	Red edge normalised difference vegetation index / Red edge normalized ratio	(ρ750 − ρ705)/(ρ750 + ρ705) / ((R750–R710)/(R750+R710))	Chlorophyll	6. no important predictor drought stress	6	Not suitable	Not suitable
RGI	Red/green pigment Index	RGI = R690/R550	Chlorophyll	2. able for early dourght detection 2. able to detecht long term drought	2	Suitable	Early & longterm drought stress
SRI / RVI /	Simple Ratio Index / Simple Ratio Vegetation index	SRI = R800 / R675 RVI =NIR/R SR = R750/R710	Chlorophyll	1. can determine vegetation stress, not exact which type of stress 4. not detecting early drought stress 4. all VIs detected longterm drought stress - (dramatic change) (need research abilities VIs different stages) 8. detect drought stress earliest 9. significant relation with TQ (0.001 & 0.01), Chlorophyll (2x 0.001), TWC (1x 0.01) - not with SWC (0x)	8, 1, 4, 9	Maybe suitable	Early & longterm drought stress
MNLI	Modified Non-linear Vegetation Index	MNLI = 1.5* (R2 800–R680)/ (R2 800 + R680 + 1.5)	Chlorophyll	4. not detecting early drought stress 4. all VIs detected longterm drought stress - (dramatic change) (need research abilities VIs different stages)	4	Not suitable	Not suitable
NDRE	Normalized Difference Red Edge	(ρ790 − ρ720)/(ρ790 + ρ720)	Chlorophyll	8. use as vegetation density but not drought stress	8	Not suitable	Not suitable

NDVI	Normalized Difference Vegetation Index	NDVI = (R800–R680)/ (R800 + R680)	Chlorophyll	1. can determine vegetation stress, not exact which type of stress 3. not able for solely water content research - because narrow bands overlap with chlorophyll 4. not detecting early drought stress 4. all VIs detected longterm drought stress - (dramatic change) (need research abilities VIs different stages) 5. relationship with visual rating tall fesue, not for hybrid bermuda 7. leaf relative water content (RWC) highly correlated with NDVI 8. use as vegetation density but not drought stress 9. significant relation with TQ (0.01 & 0.05), chlorophyll (2x 0.05), SWC (1x 0.05), TWC (1x 0.05) 10. lowest relationship to VWC 0.47	8, 1, 10, 4 , 7, 9, 5, 3	Maybe suitable	Early & longterm drought stress
NDVI705	NDVI at 705 nm	(R750 − R705)/(R750 + R705)	Chlorophyll	9. Significant relation with TQ (0.01 & 0.05), Chlorophyll (2x 0.01), TWC (1x 0.05) - not with SWC (0x)	9	Not suitable	Not suitable
f'	First derivative @ 950.6 nm	First derivative @ 950.6 nm	Plant water concentration	6. no important predictor drought stress 6. almost perfectly collinear with WBI (-0.98)	6	Not suitable	Not suitable
NWI1	Normalized Water Index 1	NWI1 = (R970 ´ R900)/ (R970 + R900)	Plant water concentration	2. able for early dourght detection 2. able to detecht long term drought	2	Suitable	Early & longterm drought stress
WI / (WBI)	Water Band Index	WBI = R900/R970	Plant water concentration	1. Can determine water stress 2. able for early dourght detection 2. able to detecht long term drought 3. able as vegetation water stress indicator - lower values of WI indicate higher water stress both in leaves and soil 6. Best performing predictor for RWC 9. significant correlation to TQ (2x 0.01), Chlorophyll (2x 0.001), soil water content (2x 0.001), tissue water content (0.01 & 0.05) 10. strongest relationship to VWC 0.62	2, 6, 1, 10, 9, 3	Suitable	Early & longterm drought stress
WI/NDVI	Ratio WI normalized diference vegetation index	(R900/R970)/((R800−R680)/ (R800+R680)	Plant water concentration	3. able as vegetation water stress indicator - WI/NDVI at higher values correspond higher water stress levels	3	Suitable	Early & longterm drought stress

11 Appendix C: Calculate percentage of change in VI values between 2nd of June and 30th of August

Table 10: Percentage of change of the averages VI values per species per mowing height for the non-irrigated plots between 2-6 and 30-8

Species	Mowing height	Irrigation	GRI	PRI	CARI	PRI 512	CarRE opt	CCRI	SIPI	BRI2	Chl RE opt	CTR2	mSR705	RGI	WI/NDVI	NWII	WBI
A	3 cm	Non-irrigated	-58%	401%	-43%	-55%	-51%	-22%	-25%	76%	-57%	112%	-36%	96%	42%	-142%	-11%
A	6 cm	Non-irrigated	-39%	134%	-17%	-29%	-20%	-5%	-11%	37%	-27%	46%	-17%	51%	12%	-108%	-9%
B	3 cm	Non-irrigated	-33%	95%	-25%	-29%	-28%	-16%	-9%	33%	-22%	27%	-11%	37%	7%	-105%	-7%
B	6 cm	Non-irrigated	-30%	75%	-13%	-19%	-14%	-3%	-6%	15%	-18%	30%	-13%	33%	3%	-89%	-7%
C	3 cm	Non-irrigated	-57%	435%	-46%	-56%	-53%	-26%	-26%	85%	-56%	122%	-35%	97%	46%	-154%	-12%
C	6 cm	Non-irrigated	-45%	156%	-20%	-30%	-24%	-5%	-11%	41%	-31%	52%	-20%	59%	9%	-118%	-10%
D	3 cm	Non-irrigated	-55%	335%	-40%	-52%	-48%	-18%	-21%	69%	-54%	96%	-35%	89%	32%	-139%	-11%
D	6 cm	Non-irrigated	-40%	126%	-18%	-27%	-24%	-3%	-10%	37%	-32%	47%	-20%	50%	8%	-97%	-8%
E	3 cm	Non-irrigated	-50%	212%	-33%	-45%	-38%	-17%	-18%	68%	-42%	58%	-23%	73%	26%	-134%	-9%
E	6 cm	Non-irrigated	-39%	121%	-16%	-30%	-17%	-5%	-11%	40%	-24%	38%	-14%	50%	13%	-97%	-7%
F	6 cm	Non-irrigated	-54%	256%	-27%	-43%	-32%	-8%	-16%	52%	-40%	93%	-27%	83%	20%	-128%	-11%
F	3 cm	Non-irrigated	-48%	181%	-30%	-42%	-34%	-16%	-15%	62%	-33%	56%	-18%	65%	16%	-148%	-10%
G	3 cm	Non-irrigated	-34%	102%	-28%	-29%	-34%	-16%	-11%	36%	-30%	38%	-17%	40%	9%	-103%	-8%
G	6 cm	Non-irrigated	-34%	97%	-21%	-26%	-25%	-10%	-9%	27%	-25%	39%	-16%	39%	7%	-101%	-8%
H	3 cm	Non-irrigated	-53%	301%	-51%	-52%	-60%	-34%	-22%	67%	-51%	100%	-31%	75%	29%	-135%	-11%
H	6 cm	Non-irrigated	-45%	247%	-34%	-39%	-43%	-16%	-16%	39%	-44%	115%	-31%	66%	27%	-101%	-9%
J	6 cm	Non-irrigated	-50%	221%	-29%	-40%	-37%	-9%	-13%	58%	-42%	76%	-28%	77%	14%	-108%	-9%
J	3 cm	Non-irrigated	-52%	204%	-34%	-49%	-42%	-15%	-16%	73%	-44%	80%	-27%	78%	19%	-132%	-8%
K	3 cm	Non-irrigated	-45%	234%	-36%	-41%	-46%	-11%	-16%	54%	-53%	87%	-36%	68%	17%	-141%	-10%
K	6 cm	Non-irrigated	-34%	118%	-17%	-24%	-26%	5%	-9%	22%	-41%	66%	-29%	45%	8%	-105%	-8%
L	3 cm	Non-irrigated	-51%	238%	-36%	-47%	-45%	-15%	-17%	63%	-48%	90%	-31%	73%	20%	-140%	-11%
L	6 cm	Non-irrigated	-50%	275%	-30%	-37%	-40%	-5%	-14%	46%	-49%	104%	-34%	78%	18%	-107%	-9%
M	3 cm	Non-irrigated	-31%	116%	-34%	-42%	-42%	-19%	-14%	48%	-39%	53%	-22%	36%	15%	-119%	-8%
M	6 cm	Non-irrigated	-19%	72%	-19%	-25%	-26%	-6%	-8%	27%	-30%	32%	-18%	24%	4%	-94%	-8%
N	6 cm	Non-irrigated	-51%	221%	-26%	-41%	-30%	-9%	-16%	52%	-37%	73%	-24%	74%	21%	-130%	-10%
N	3 cm	Non-irrigated	-48%	188%	-34%	-40%	-40%	-18%	-16%	65%	-39%	57%	-22%	65%	17%	-121%	-9%
O	3 cm	Non-irrigated	-60%	486%	-48%	-57%	-56%	-26%	-26%	84%	-59%	126%	-38%	106%	40%	-145%	-13%
O	6 cm	Non-irrigated	-54%	306%	-31%	-42%	-38%	-11%	-16%	55%	-44%	87%	-29%	85%	19%	-127%	-12%
P	3 cm	Non-irrigated	-43%	171%	-24%	-37%	-27%	-12%	-14%	48%	-31%	38%	-17%	56%	15%	-130%	-10%
P	6 cm	Non-irrigated	-47%	181%	-22%	-38%	-25%	-7%	-14%	44%	-32%	62%	-21%	66%	18%	-120%	-9%
R	3 cm	Non-irrigated	-45%	195%	-36%	-40%	-43%	-20%	-14%	49%	-39%	65%	-24%	59%	14%	-115%	-9%
R	6 cm	Non-irrigated	-34%	90%	-19%	-24%	-25%	-5%	-8%	28%	-27%	40%	-17%	40%	5%	-79%	-7%
S	3 cm	Non-irrigated	-48%	201%	-36%	-42%	-42%	-20%	-17%	68%	-40%	61%	-23%	65%	18%	-126%	-10%
S	6 cm	Non-irrigated	-43%	159%	-19%	-31%	-23%	-4%	-12%	40%	-31%	51%	-20%	57%	12%	-112%	-9%
T	3 cm	Non-irrigated	-42%	144%	-29%	-35%	-33%	-17%	-13%	51%	-31%	40%	-16%	52%	11%	-118%	-9%
T	6 cm	Non-irrigated	-47%	209%	-22%	-34%	-27%	-5%	-12%	41%	-36%	60%	-24%	66%	14%	-109%	-9%
U	3 cm	Non-irrigated	-53%	300%	-40%	-47%	-48%	-21%	-19%	77%	-51%	74%	-31%	81%	24%	-130%	-11%
U	6 cm	Non-irrigated	-50%	206%	-22%	-37%	-28%	-2%	-13%	43%	-39%	79%	-26%	71%	15%	-109%	-9%
W	3 cm	Non-irrigated	-35%	135%	-29%	-39%	-36%	-15%	-12%	48%	-37%	37%	-21%	43%	10%	-136%	-10%
W	6 cm	Non-irrigated	-45%	166%	-26%	-40%	-35%	-6%	-13%	44%	-42%	86%	-28%	61%	14%	-113%	-9%

Table 11: Percentage of change of the averages VI values per species per mowing height for the irrigated plots between 2-6 and 30-8

Species	Mowing height	Irrigation	GRI	PRI	CARI	PRI512	CarRE opt	CCRI	SIPI	BRI2	ChlRE opt	CTR2	mSR705	RGI	WI/ NDVI	NWII	WBI
A	3 cm	Irrigated	-8%	10%	8%	2%	10%	9%	1%	1%	-3%	-13%	-3%	12%	-8%	-93%	-8%
A	6 cm	Irrigated	-16%	27%	0%	-3%	-1%	5%	-2%	6%	-10%	6%	-8%	16%	-4%	-85%	-7%
B	3 cm	Irrigated	4%	-12%	7%	3%	8%	8%	2%	-1%	-2%	-13%	-2%	3%	-8%	-63%	-6%
B	6 cm	Irrigated	1%	-5%	3%	4%	3%	7%	1%	-4%	-7%	-3%	-6%	4%	-6%	-68%	-6%
C	3 cm	Irrigated	-23%	46%	3%	-8%	4%	10%	-2%	9%	-12%	6%	-9%	27%	-5%	-97%	-9%
C	6 cm	Irrigated	-30%	96%	-4%	-7%	-6%	7%	-3%	8%	-20%	10%	-15%	34%	-5%	-126%	-11%
D	3 cm	Irrigated	-6%	-1%	9%	1%	11%	10%	1%	1%	-1%	-8%	-2%	10%	-6%	-68%	-6%
D	6 cm	Irrigated	-13%	24%	0%	-2%	-1%	6%	-1%	2%	-11%	7%	-9%	15%	-4%	-75%	-6%
E	3 cm	Irrigated	-18%	30%	5%	-3%	8%	9%	-1%	7%	-7%	-4%	-6%	22%	-6%	-102%	-9%
E	6 cm	Irrigated	-22%	26%	1%	-8%	2%	5%	-3%	12%	-6%	13%	-5%	21%	-2%	-81%	-7%
F	6 cm	Irrigated	-23%	65%	1%	-6%	0%	11%	-3%	5%	-20%	8%	-15%	30%	-5%	-107%	-10%
F	3 cm	Irrigated	-28%	57%	-9%	-13%	-12%	1%	-6%	18%	-19%	27%	-13%	31%	-1%	-106%	-10%
G	3 cm	Irrigated	6%	-22%	11%	3%	13%	9%	2%	0%	2%	-13%	1%	1%	-7%	-55%	-5%
G	6 cm	Irrigated	0%	-1%	2%	4%	-1%	8%	0%	-3%	-11%	6%	-10%	4%	-5%	-59%	-6%
H	3 cm	Irrigated	-8%	11%	-2%	-4%	-9%	8%	-1%	1%	-20%	6%	-15%	15%	-5%	-71%	-7%
H	6 cm	Irrigated	-6%	15%	0%	3%	-7%	13%	0%	-13%	-21%	7%	-18%	9%	-5%	-79%	-7%
J	6 cm	Irrigated	-16%	7%	-5%	-11%	-8%	1%	-2%	16%	-13%	5%	-8%	19%	-4%	-77%	-7%
J	3 cm	Irrigated	-11%	-8%	1%	-4%	1%	4%	-1%	6%	-3%	5%	-3%	11%	-3%	-45%	-4%
K	3 cm	Irrigated	6%	-10%	7%	4%	5%	11%	2%	-6%	-9%	-12%	-7%	4%	-9%	-89%	-7%
K	6 cm	Irrigated	2%	1%	6%	8%	2%	14%	1%	-16%	-14%	-2%	-13%	5%	-7%	-85%	-7%
L	3 cm	Irrigated	-11%	7%	-3%	-8%	-6%	4%	-1%	9%	-13%	5%	-9%	16%	-5%	-78%	-7%
L	6 cm	Irrigated	-22%	53%	-4%	-5%	-9%	8%	-3%	0%	-23%	19%	-18%	24%	-5%	-98%	-10%
M	3 cm	Irrigated	19%	-38%	11%	9%	10%	10%	3%	-2%	1%	-13%	1%	-8%	-9%	-54%	-4%
M	6 cm	Irrigated	21%	-36%	9%	15%	10%	7%	3%	-9%	4%	-16%	1%	-11%	-11%	-68%	-6%
N	6 cm	Irrigated	-7%	3%	10%	2%	13%	11%	1%	1%	-2%	-9%	-3%	11%	-7%	-77%	-7%
N	3 cm	Irrigated	-14%	32%	0%	-1%	1%	5%	-1%	3%	-10%	-6%	-7%	15%	-6%	-105%	-9%
O	3 cm	Irrigated	-26%	72%	-3%	-10%	-5%	10%	-4%	10%	-22%	15%	-17%	34%	-4%	-99%	-10%
O	6 cm	Irrigated	-23%	49%	-6%	-9%	-8%	3%	-3%	11%	-17%	13%	-12%	24%	-3%	-107%	-9%
P	3 cm	Irrigated	-6%	-1%	11%	3%	17%	9%	2%	2%	4%	-16%	1%	10%	-9%	-88%	-8%
P	6 cm	Irrigated	-19%	24%	-1%	-7%	-1%	4%	-2%	10%	-8%	10%	-6%	18%	-3%	-78%	-7%
R	3 cm	Irrigated	-3%	-5%	4%	-1%	1%	11%	0%	-2%	-11%	1%	-9%	8%	-5%	-54%	-5%
R	6 cm	Irrigated	-5%	22%	-3%	1%	-8%	8%	-1%	-7%	-21%	1%	-16%	11%	-5%	-67%	-6%
S	3 cm	Irrigated	-8%	11%	10%	1%	11%	14%	0%	0%	-7%	-3%	-6%	12%	-7%	-84%	-7%
S	6 cm	Irrigated	-19%	36%	-3%	-5%	-6%	5%	-2%	5%	-15%	10%	-11%	19%	-4%	-97%	-9%
T	3 cm	Irrigated	0%	-7%	12%	4%	18%	8%	3%	-2%	8%	-19%	4%	5%	-9%	-82%	-7%
T	6 cm	Irrigated	-13%	14%	-1%	-3%	-2%	5%	-1%	4%	-10%	6%	-8%	14%	-4%	-72%	-6%
U	3 cm	Irrigated	2%	-13%	14%	6%	19%	12%	2%	-4%	5%	-17%	2%	4%	-9%	-72%	-6%
U	6 cm	Irrigated	-23%	50%	-2%	-3%	-5%	7%	-2%	2%	-16%	16%	-13%	23%	-3%	-87%	-8%
W	3 cm	Irrigated	8%	-24%	8%	3%	7%	8%	2%	-2%	-2%	-10%	-2%	0%	-8%	-64%	-5%
W	6 cm	Irrigated	8%	-17%	4%	6%	4%	5%	1%	-6%	-4%	-16%	-3%	-2%	-8%	-74%	-6%

12 Appendix D: Percentage of variance explained & Variable contributions in PCA analysis

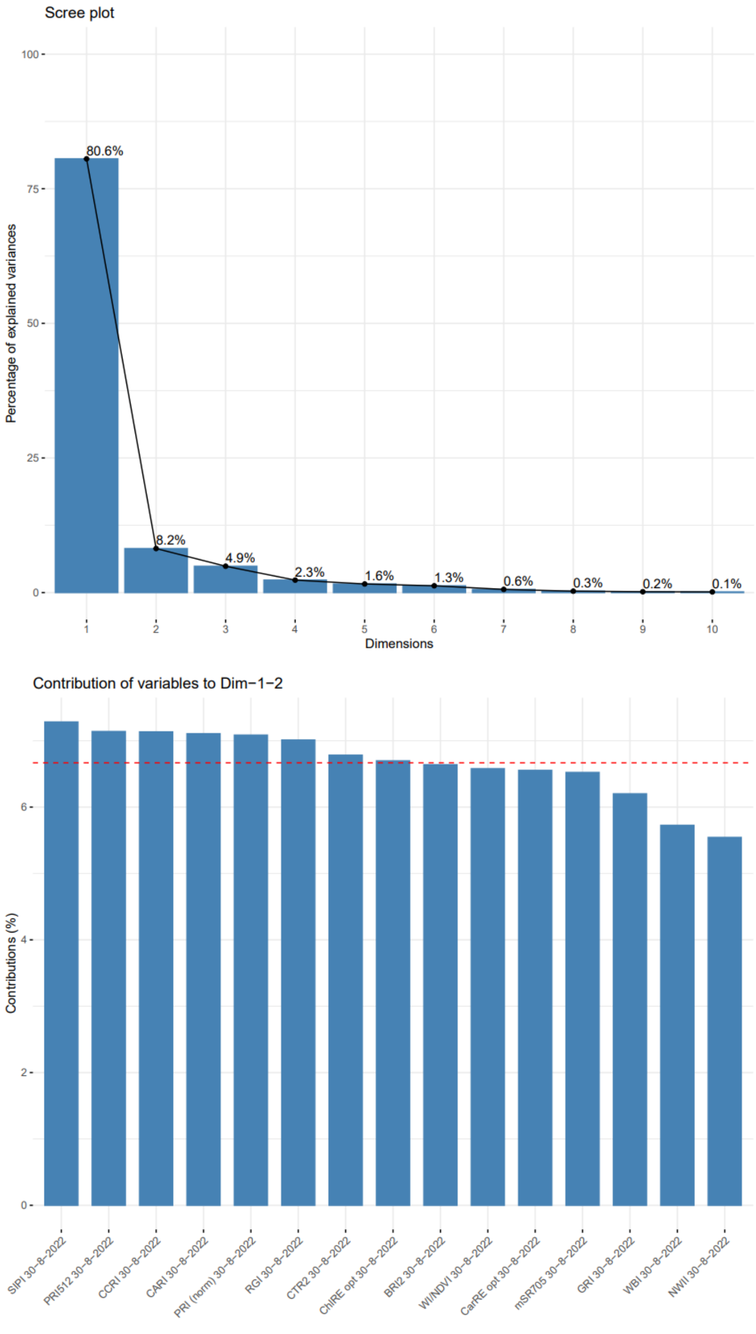
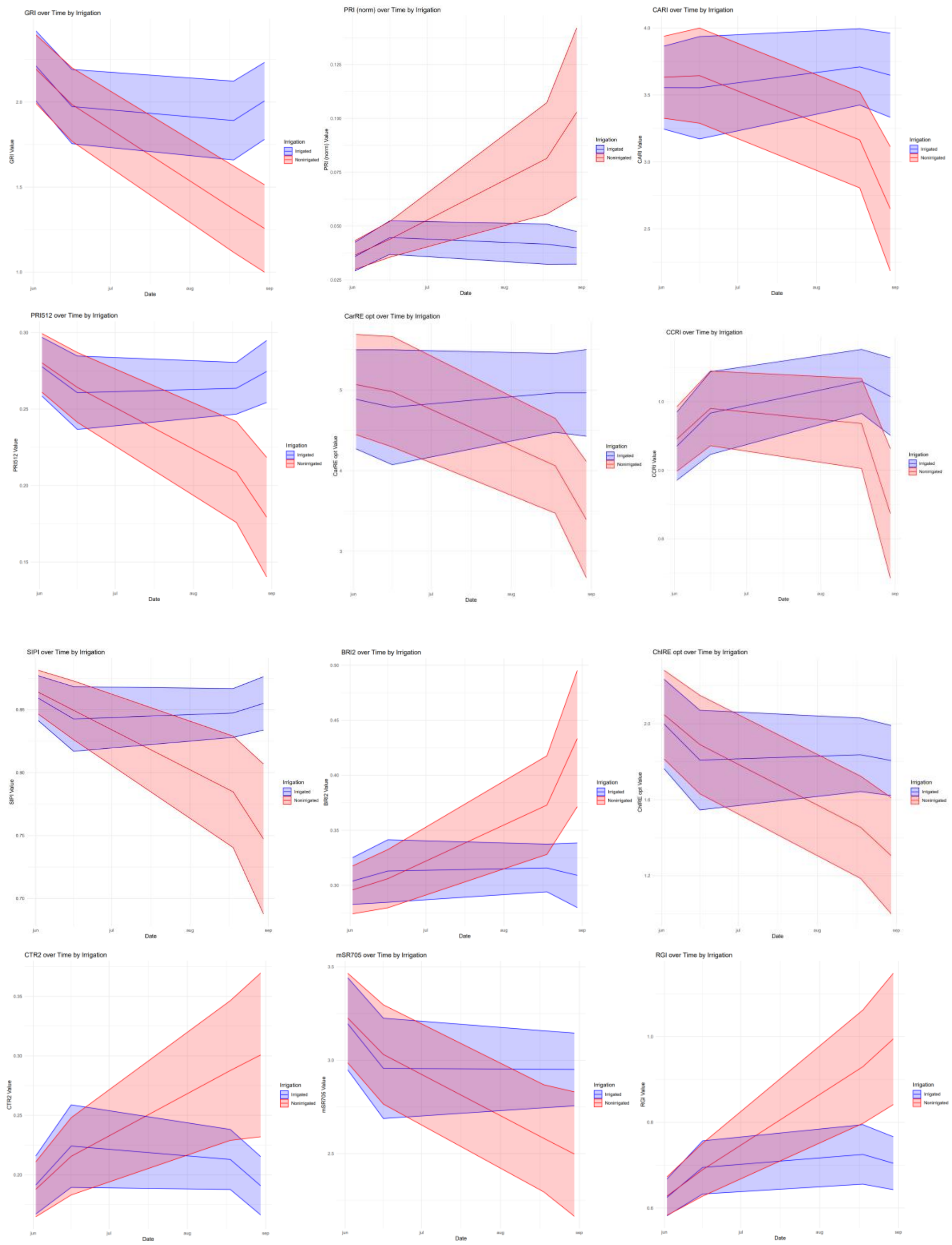


Figure 22: Results of PCA analysis, illustrating the percentage of variance explained and contribution of the variables

13 Appendix E: Development of VI values by irrigation throughout the experiment



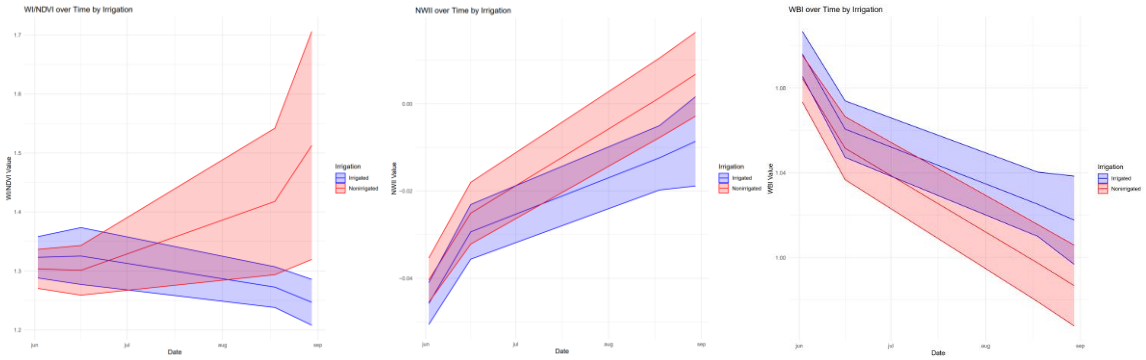
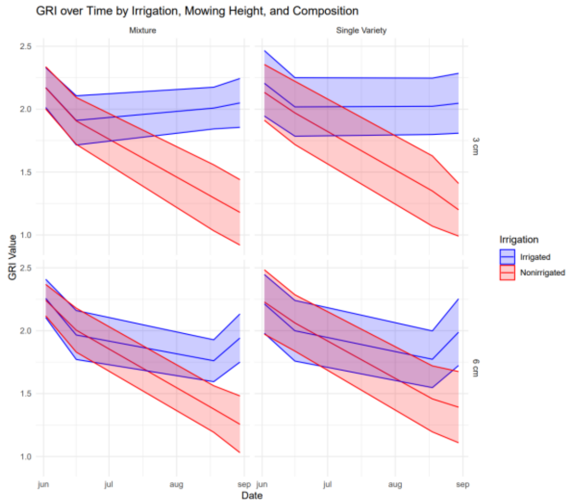
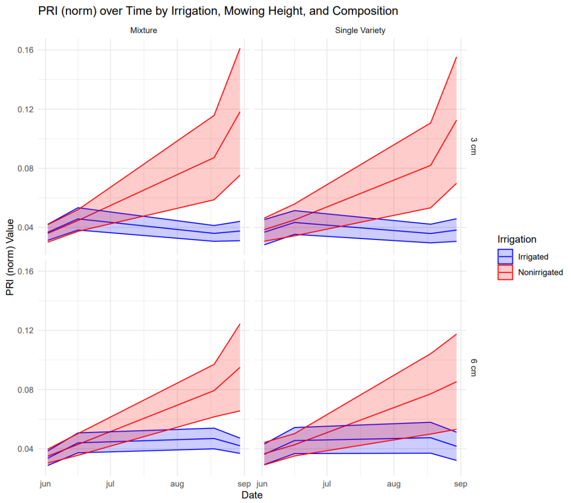
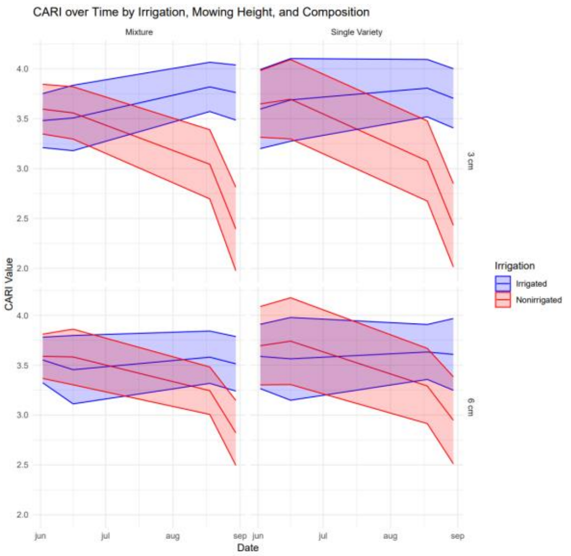


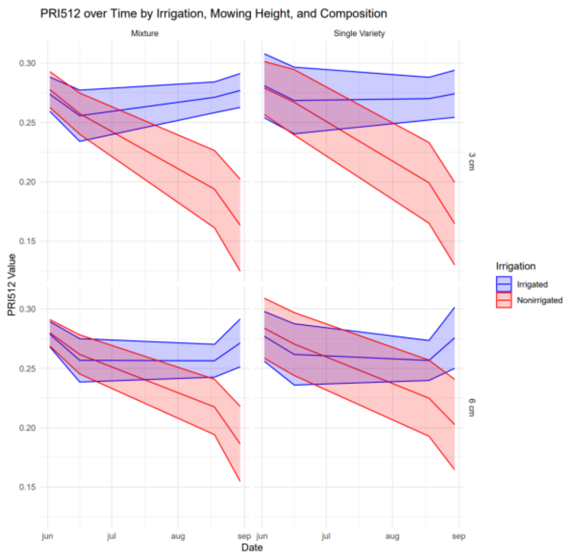
Figure 23: Development of VI values by irrigation throughout the experiment

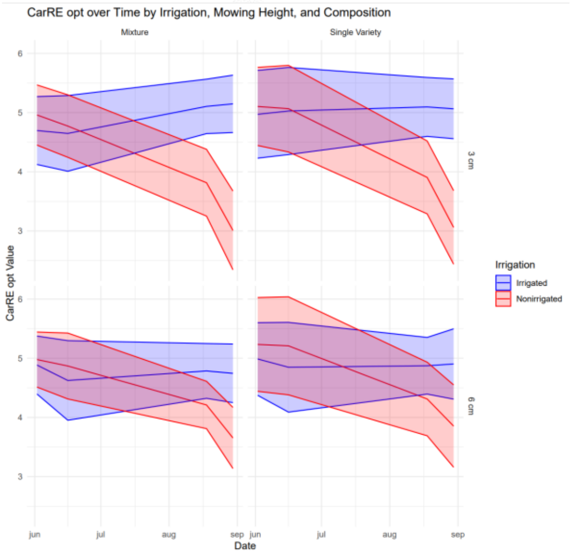
14 Appendix F: VI values by irrigation, mowing height and mixture throughout the experiment

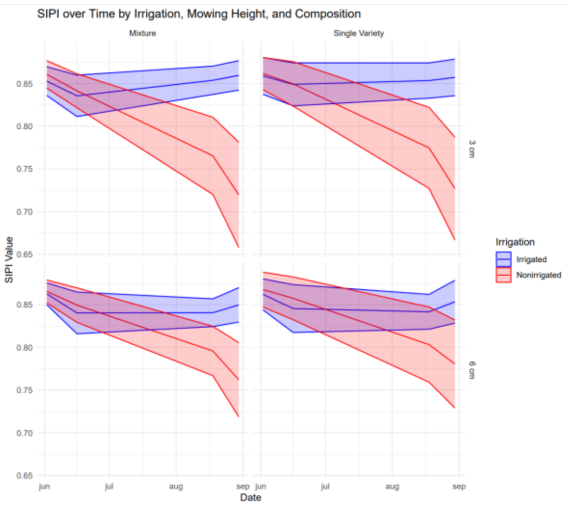


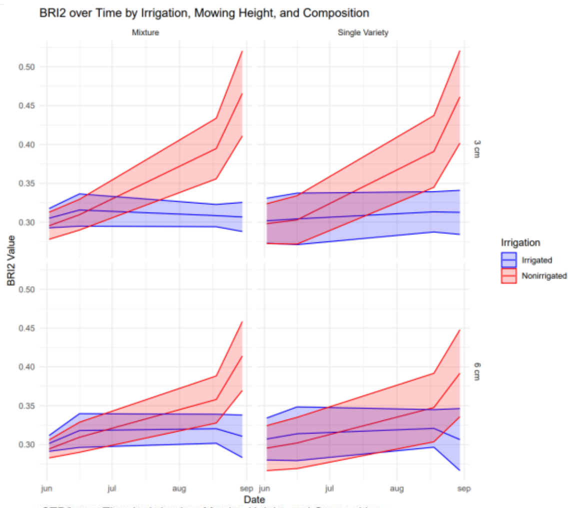


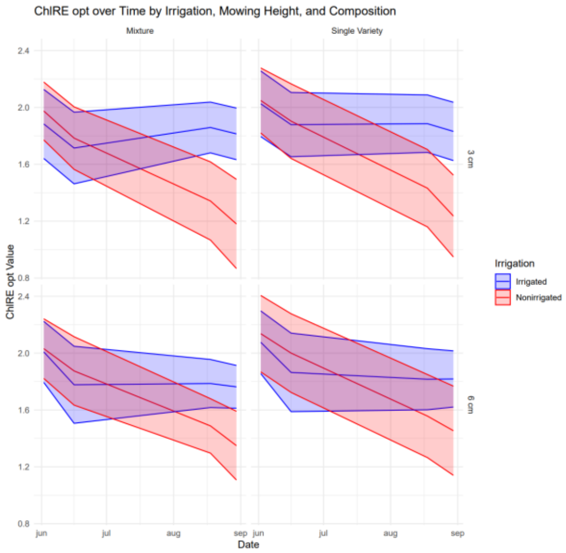


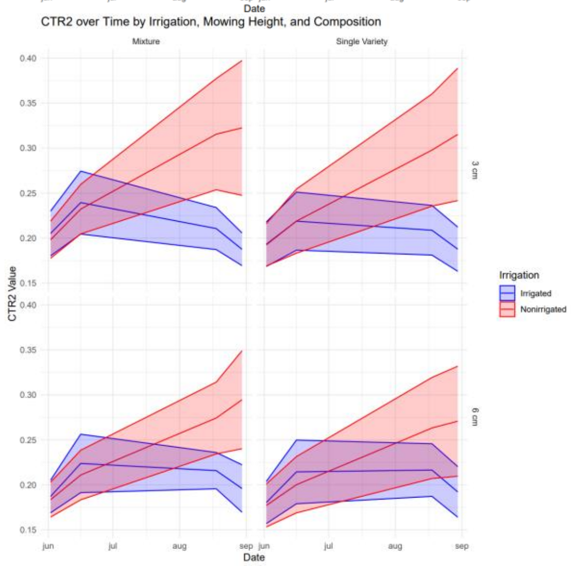


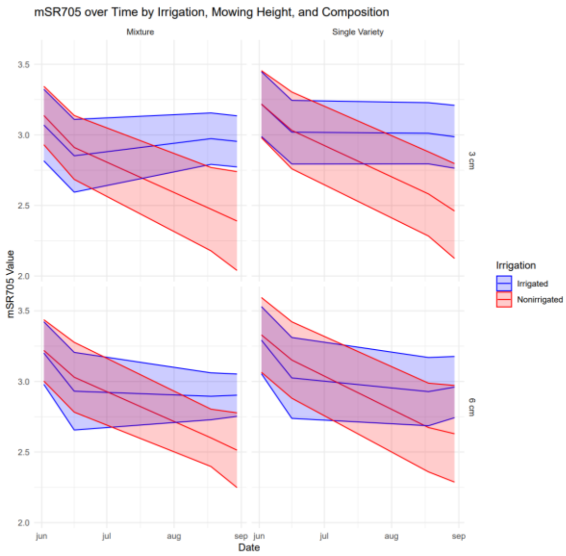


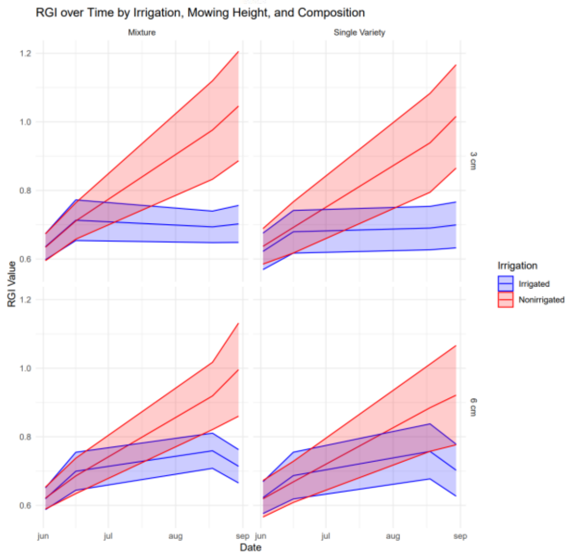

SIPI over Time by Irrigation, Mowing Height, and Composition
Mixture
Single Variety
3 cm
6 cm
SIPI Value
Date
jun
jul
aug
sep
Irrigation
Irrigated
Nonirrigated


BRI2 over Time by Irrigation, Mowing Height, and Composition
Mixture
Single Variety
3 cm
6 cm
BRI2 Value
Date
jun
jul
aug
sep
Irrigation
Irrigated
Nonirrigated


ChlRE opt over Time by Irrigation, Mowing Height, and Composition
Mixture
Single Variety
3 cm
6 cm
ChlRE opt Value
Date
jun
jul
aug
sep
Irrigation
Irrigated
Nonirrigated


CTR2 over Time by Irrigation, Mowing Height, and Composition
Mixture
Single Variety
3 cm
6 cm
CTR2 Value
Date
jun
jul
aug
sep
Irrigation
Irrigated
Nonirrigated


mSR705 over Time by Irrigation, Mowing Height, and Composition
Mixture
Single Variety
3 cm
6 cm
mSR705 Value
Date
jun
jul
aug
sep
Irrigation
Irrigated
Nonirrigated


RGI over Time by Irrigation, Mowing Height, and Composition
Mixture
Single Variety
3 cm
6 cm
RGI Value
Date
jun
jul
aug
sep
Irrigation
Irrigated
Nonirrigated

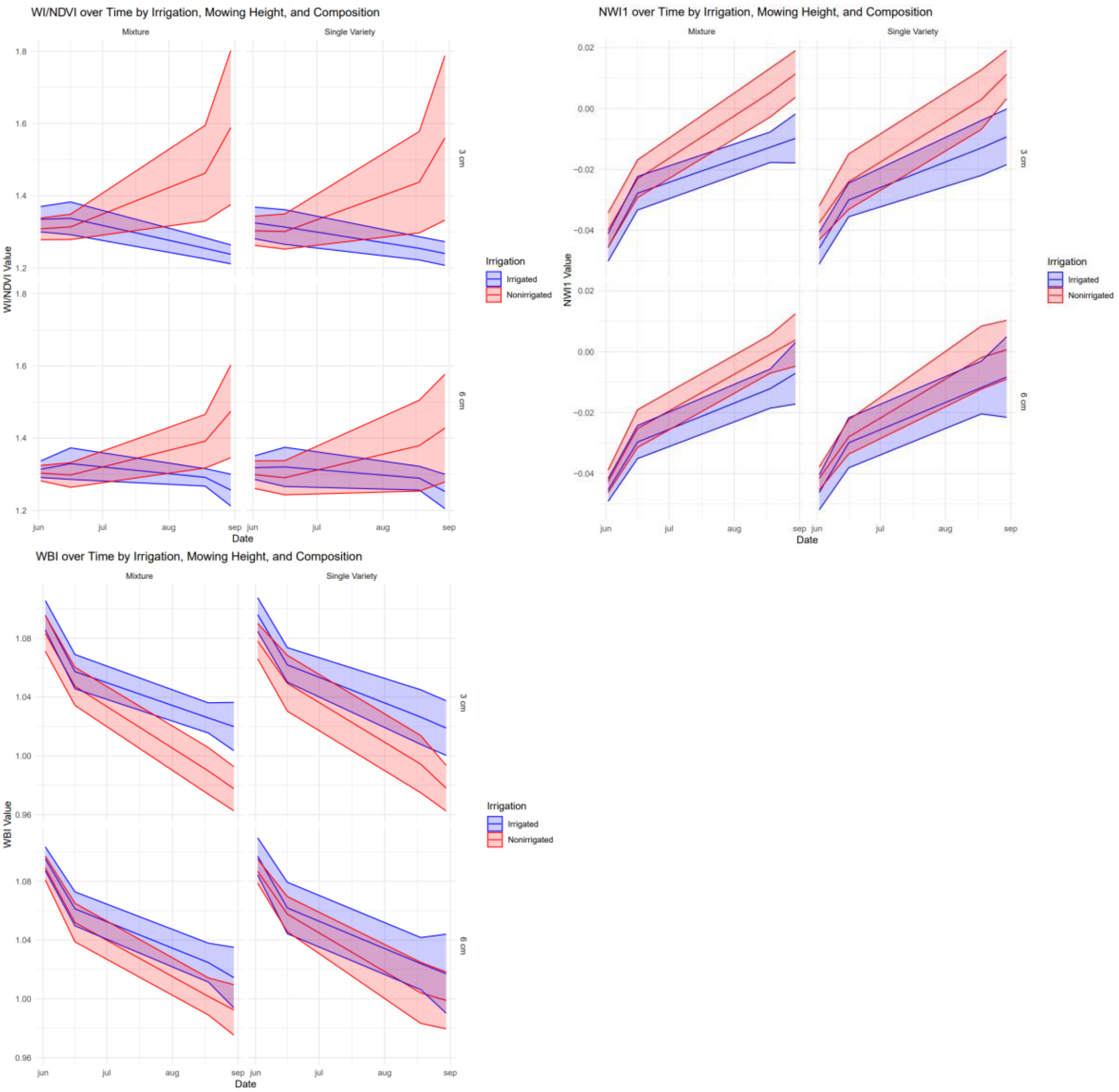


Figure 24: VI values by irrigation, mowing height and mixture throughout the experiment