HIGH-THROUGHPUT FIELD PHENOTYPING OF ASCOCHYTA BLIGHT DISEASE SEVERITY IN CHICKPEA USING MULTISPECTRAL IMAGING

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ABSTRACT

Ascochyta blight (AB) caused by *Ascochyta rabiei* (Pass.) Labr. is an important and widespread disease of chickpea (*Cicer arietinum* L.) worldwide. The disease is particularly severe under cool and humid weather conditions, leading to crop losses at all stages of chickpea growth. Screening for resistant cultivars remains the most effective, economic and ecological method of disease management. However, traditional phenotyping methods that relying on trained experts are slow, costly, labor-intensive, subjective, often involve destructive sampling. The development of high-throughput phenotyping methods for Ascochyta blight disease holds promise for precise and rapid data. In this study, 216 chickpea genotypes were screened in field trials to investigate the use of digital imaging to implement reliable phenotyping of Ascochyta blight resistance. An unmanned aerial system equipped with a 5-band multispectral camera was used to capture imagery of the tested genotype plots. Digital image processing was employed to extract the NDVI index. Our aim was to explore the correlation between the NDVI index and visual disease severity ratings for Ascochyta blight. Results revealed a consistent correlation between the NDVI index extracted from image features and disease severity with R^2 of 0.936. Genotypes were classified into resistant (R), moderately resistant (MR) susceptible (S) and highly susceptible categories based on their responses. These differences in genotypes response were utilized to developpe a predictive model for monitoring Ascochyta blight. Our findings highlight that rapid and precise image-based, high-throughput phenotyping can effectively differentiate responses to Ascochyta blight across many chickpea genotypes.

Keywords: Drone imaging, Ascochyta blight, severity, phenotyping, chickpea, NDVI,

INTRODUCTION

Chickpea (*Cicer arietinum L.*) is a highly valuable crop, providing an important source of protein and improving soil health through nitrogen fixation. However, its production is severely affected by various abiotic and biotic stresses, including drought and diseases. Among these, Ascochyta blight (AB), caused by *Ascochyta rabiei* (Kovatsch.) Arx, 1962, is a major biotic threat that significantly limits chickpea yield [1]. AB primarily affects the plant's foliar parts, causing lesions and tissue necrosis that reduce seed quality and overall crop productivity. The disease often begins in small patches within the field but can rapidly spread under favorable conditions of temperature and rainfall [2-3]. Weather plays a crucial role in AB development, particularly in cooler (15-

25°C) and humid environments (>70%) during the growing season. Additionally, factors such as inoculum type, virulence, concentration, and the plant's growth stage and resistance level influence the severity and spread of the disease [3].

Due to the polycyclic nature of AB, control often requires multiple fungicide applications, which are costly and pose risks to human health, wildlife, and ecosystems [4, 5]. Additionally, the overuse of fungicides may lead to contamination and the development of resistant pathogens. Consequently, the sustainable method for managing Ascochyta blight is based on breeding to select resistant cultivars. However, the traditional methods of phenotyping disease resistance, relying on human expertise, are often time and labor consuming, not cost-effective, and sometimes requires destructive sampling of plants. In this context, the use of high-throughput phenotyping (HTP) methods for Ascochyta blight is promising for developing precise and rapid disease assessment digital tool. The HTP based on using digital imaging, such as drones mounted thermal, multispectral (MSI), or hyperspectral (HIS) cameras, offers a non-invasive and consistent imaging process to monitor plant stresses and disease severity. Use of drone technologies, capturing high-resolution spectral data showed great opportunities to detect both biotic and abiotic stresses in different crops [6,7]. Different Indices like NDVI (Normalized Difference Vegetation Index) and GNDVI (Green Normalized Difference Vegetation Index), are commonly used to assess disease severity among different plant stresses due plant hydric state or plant health state.

Several authors [8-10] have been used digital methods for evaluating plant disease severity to provide greater accuracy, repeatability, and reproducibility compared to traditional techniques. This digital process involves image acquisition, analysis, processing, and validation through specialized software [8-10]. In fact, over the past three decades (1990-2020), significant advancements have been made in using digital tools for evaluating plants diseases severity. In the 1990s, cameras were first used to distinguish between healthy and diseased plants, such as in studies on Fusarium in corn [11] and maize streak virus (MSV) in resistant corn [12]. The 2000s saw the development of image analysis software like Assess [13] and ImageJ [14, 15], which improved the precision of disease quantification. By the 2010s, advanced imaging techniques, including thermal, hyperspectral (HIS), and multispectral (MSI) imaging, became widely used, offering early disease detection and more effective management compared to traditional visible spectrum imaging [16,17]. These imaging technologies, often mounted on drones, detect plant stress or disease by capturing temperature variations and multispectral data [18-19]. MSI cameras calculate spectral indices such as NDVI, which have been shown to strongly correlate with disease severity and plant health [20,21]. For instance, NDVI exhibited a strong negative correlation with disease severity in pineapple (-0.83 to -0.88) [22], and in chickpea, it correlated with leaf area index, chlorophyll content, and biomass [23]. Additionally, the correlation between visual disease ratings and NDVI in chickpea increased from -0.61 to -0.66 after 58 days, with NDVI's correlation with yield ranging from 0.76 to 0.92 [24].

According to the short review stated above, an early and accurate disease detection remain essential for implementing timely management strategies. Furthermore, the digital methods require more improvements as it is often difficult to discriminate between biotic and abiotic stresses that may cause similar symptoms, making visual diagnosis challenging [25]. In fact, the use of these digital indices cannot differentiate between biotic and abiotic stresses without efforts from agronomic experts of relying on the indices data information to the main occurring stress and

avoiding spatial and temporal interference between two different stresses that can be potentially expressed in the same digital data taken from one image process acquisition [26, 27].
This study explores the use of digital imaging for reliable phenotyping of Ascochyta blight resistance in chickpea. Specifically, our innovative digital method aims for testing the correlation between disease severity and NDVI and boosting this correlation trough use of different plants/genotypes as checks for showing a gradient of resistance to AB severity and using it as reference model to predict the disease severity among a large sample of plants/genotypes that can be potentially tested for selection with reference to AB severity using NDVI information. This innovative digital method aims to developing a precise, automated phenotyping process for an effective disease management.

MATERIAL AND METHODS

Evaluation of AB disease severity using classic method of visual scale

A field trial was conducted in 2020-2021 at the Sidi El Aidi experimental station of the INRA Settat. A total of 216 chickpea genotypes with varied resistance to AB was tested, using a randomized complete block design (RCBD) augmented with nine blocks and four checks. Spores of *A. rabiei* was inoculated via foliar spraying, during vegetative stage. Disease severity was evaluated visually using a 0-9 scale [28]. The visual reading of AB disease severity was taken for comparison with digital method using drone multispectral imaging to promote as rapid evaluation of crops health by the plant pathologist.

Evaluation of AB disease severity using digital imaging and NDVI

The disease severity was evaluated also digitally using drone multispectral imaging. NDVI values were computed from multispectral data to assess plant health. The NDVI values were used to find potential correlation with the visual reading.

Reference model for calibration and prediction of disease severity

To test the response of the 216 chickpea genotypes to AB disease severity, four checks of chickpea genotypes were used as references to show a gradient of different responses from low to high resistance to AB disease severity. A tuning curve is generated from a gradient of response (4 checks, 9 levels of disease severity) to serve as a reference model for testing the responses of 216 genotypes. This model is then used to assess the potential for predicting AB disease severity among the tested genotypes.

RESULTS

Evaluation of AB disease severity using visual assessment and NDVI

Use of decision-making tools is essential for phytopathologists to effectively manage disease interventions by utilizing digital NDVI information to evaluate disease severity. To develop this tool, we assessed the correlation between Ascochyta blight (AB) disease severity and NDVI based on data from 216 plots of tested genotypes and 36 plots of reference genotypes. Where C1 represents the resistant check, C2 represents the susceptible check, and C3 and C4 represent the moderately resistant checks (Fig. 1)

Since AB is characterized by color changes, resulting from lesions on plant leaves, the use of digital imaging data has significantly distinguished the infected plants. The NDVI results of four checks clearly illustrate the varying responses of chickpea genotype plots, showing a gradual transition in leaf color from vibrant green (resistant check C1) to yellow, orange (moderately resistant C3 and C4), and ultimately red (susceptible check C2). This decrease in green coloration correlates with the severity of AB, highlighting the relative foliar changes in the infected plants (Fig. 2).

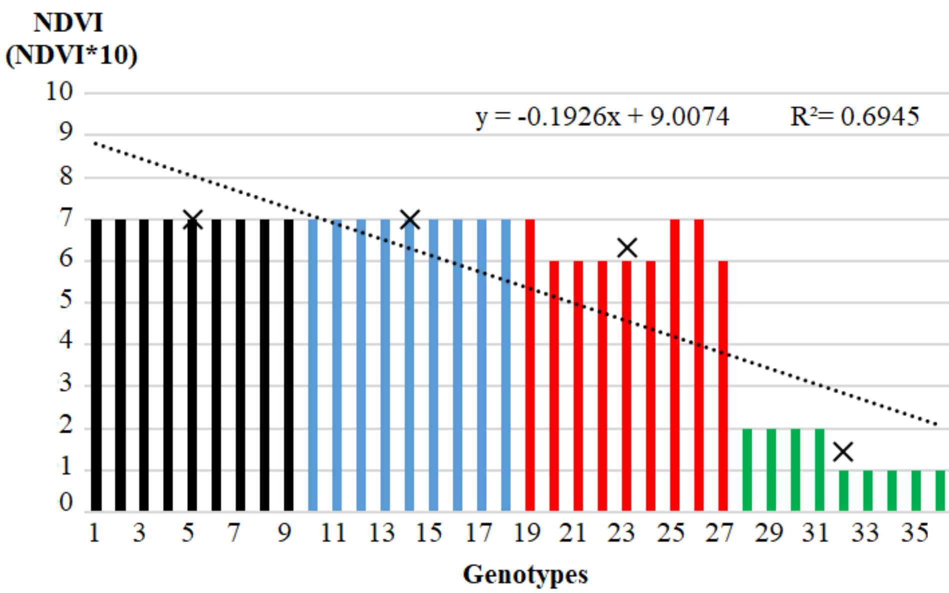

Figure 1. NDVI values relatives to 4 checks having different reaction to AB, resistant, moderate resistant or susceptible (36 plots relative to four checks with nine repetitions).

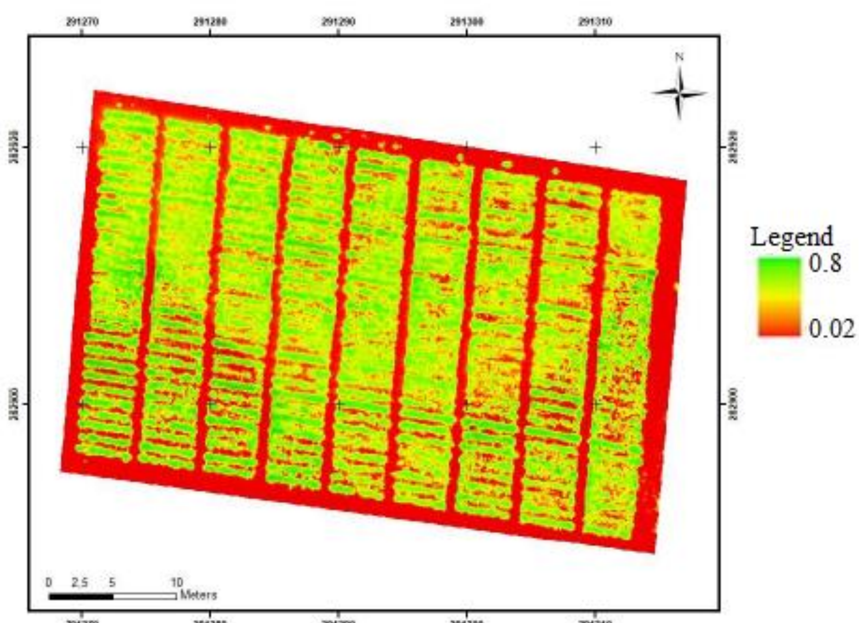

Figure 2. NDVI image illustrating 216 genotype plots with visual notation of 4 checks resistance to AB disease (S: susceptible, MR: Moderately Resistant and R: Resistant).

Reference model for calibration and prediction of disease severity

The correlation between NDVI and visual reading using data from the four checks used as reference genotypes facilitated the creation of an empirical model that can be used to predict AB disease severity using NDVI as an indicator (Eq. 1). The model equation is mounted as follows:

$$\textbf{Predicted Severity} = -0.9816 * \text{NDVI} + 9.983 \qquad (1)$$

The results showed existence of a strong correlation ($R^2 = 0.98$) between the actual visual rating scores and the predicted scores.

Prediction test of severity using the reference model

The severity of AB disease among the 216 genotype is predicted using the reference model to show how it is possible to find fitting between the predicted severity and the actual one.

Among the 216 genotypes, the correlation between measured severity and predicted severity showed a good fit using RMSE, MAE, and RE metrics (RMSE = 0.27, MAE = 0.15, RE = ±0.04), indicating its potential for practical application in assessing AB disease severity in crop fields based solely on NDVI data.

DISCUSSION

In this research, drone multispectral imaging was utilized to evaluate the severity of Ascochyta blight (AB) in 252 chickpea plots, comprising 216 genotypes and four check varieties. The NDVI (Normalized Difference Vegetation Index) was calculated to correlate with visual disease ratings, enabling field-scale assessment of AB severity. High-resolution orthophoto images revealed distinct differences between heavily infested and healthy plots, with consistent discoloration linked to increased plant mortality.

The study found a strong correlation between NDVI and AB severity, aligning with previous research indicating NDVI as a robust index for disease quantification. The relationship between NDVI and visual ratings showed an impressive R^2 value of 0.98. An empirical model was developed through linear regression, successfully predicting disease severity, validated by RMSE, MAE, and RE metrics.

The potential of NDVI as a decision-making tool for disease management was emphasized, facilitating timely interventions based on environmental conditions. Other studies highlighted the use of machine learning and decision support systems in disease detection and management, demonstrating significant accuracy in monitoring various crop diseases.

However, the accuracy of disease monitoring may be influenced by factors such as plant senescence, canopy density, and environmental conditions. Our study established a significant correlation between NDVI (Normalized Difference Vegetation Index) and the disease severity of four control genotypes with known reactions to Ascochyta blight. This finding highlights the potential of NDVI as a reliable tool for detecting biotic stress in crops. The use of NDVI for monitoring plant health has been widely reported as an effective indicator for assessing vegetation vigor and stress, particularly in response to pathogens [25]. By providing non-destructive, real-time monitoring capabilities, NDVI can serve as an early-warning system for managing disease outbreaks and guiding targeted interventions in precision agriculture [25]. Our results further validate the growing body of research that supports NDVI as a promising tool for assessing biotic stress in crops, allowing for efficient and sustainable crop management. In addition, the study underscored the need for calibrated images to increase accurate analysis and suggested further research to enhance the differentiation of disease symptoms using hyperspectral and multispectral sensing techniques.

CONCLUSION

This study showed that it is possible to adequately use NDVI derived from multispectral images and improve its fitting to effectively detect and assess the severity of Ascochyta blight on chickpeas. A strong correlation between NDVI and disease ratings allowed for the creation of an accurate predictive model. The prediction is greatly improved as the model calibration is referenced to a gradient of disease severity using four genotypes as checks to show a gradual response of disease severity.

The reference curve of the checks responses to AB disease showed that it is possible to implement robust predictive model for monitoring disease severity. In fact, the referencing of NDVI information to disease severity of known genotypes improved the model fitting. The digital monitoring of chickpea green cover can be greatly improved if the fitting of NDVI response to disease severity is calibrated with reference to use of checks gradient to assess AB disease severity. This innovative method based on calibration can potentially help the plant pathologists to overcome the problem of discriminating between NDVI responses to biotic and/or abiotic stresses by using specifically NDVI information to assess and control AB disease severity. The results highlight NDVI's potential for field-scale disease monitoring and high-throughput phenotyping, with future integration of deep learning offering further advancements in disease management.

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