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ARTICLE

Assessment of the efficiency of image analyzer applications in determining substrate quality for vegetable seedling development

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Abstract

The aim of this study is to evaluate the efficiency of images analyzer applications in determining substrate quality for vegetable seedling development. Two leaf coverage determiner apps using digital images, Canopeo and GreenTest, were tested and their values were compared with results obtained from NDVI, fresh mass, dry mass, and humidity in a substrate testing experiment for lettuce seedling development. The experiment, employing a completely randomized design with 4 treatments (control, witness, humus and organic compost) and 4 repetitions, took place in a greenhouse in Londrina, Paraná, Brazil, in April 2022. Results indicated that the Canopeo app demonstrated greater sensitivity in distinguishing between treatments and exhibited a very strong relation with other scientifically employed analyses. In contrast, GreenTest showed moderate relation and lower sensitivity in the analyses. In conclusion, Canopeo proves to be a reliable application for determining leaf coverage and analyzing the performance of vegetable seedlings. **Keywords**: Destructive Testing; Lettuce seedling; NDVI;

1. Introduction

The production of vegetable seedlings can be considered one of the most important stages in lettuce cultivation, as it directly influences the plant productivity, the quality on the product destined for the consumer market, and the profit of the producer (Castoldi et al., 2014).

In addition to the need to produce high-quality seedlings, horticulturists also face the necessity of costs reduction in their activities, and one of the alternatives that should contribute to reducing production-associated cost is the utilization of organic compost available in the production region (Moraes et al., 2021).

However, to replace commercial substrates, there is a need to test alternative substrates by evaluating seedlings based on parameters such as leaf number, plant size, fresh and dry mass, root size, and leaf area (Castoldi et al., 2014; Moraes et al., 2021). These techniques are time-

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consuming and manual, require qualified labor, are expensive, destructive, prone to human errors, and necessitate external devices such as scales, drying oven, laboratory glassware and calipers (Haider et al., 2021).

The assessment of leaf coverage and the normalized difference vegetation index (NDVI) are the techniques that allow for the evaluation of plant development and, consequently, of substrates. Commercial instruments for their indirect estimation, such as the GreenSeeker, prove to be an alternative to destructive methods, offering time savings, precision, reliability, ease of application, and the ability to estimate various parameters related to crops (Farias, el al, 2023).

The GreenSeeker is a commercial equipmente with an optical sensor that uses light-emitting diodes in the red (650nm) and NIR (770nm), which calculates, through internal microprocessing, obtaining the NDVI (Gomes et al., 2021; Reznick et al., 2021). On the other hand, it is an expensive instrument (Confalonieri et al, 2013).

Smartphones exhibit performance similar to computers, connectivity, are ubiquitous, and have an affordable price for a large percentage of the population. Equipped with a camera, memory, and processing power that make them suitable for the indirect determination of leaf coverage (Confalonieri et al., 2013), provided they are equipped with a dedicated application for this function, as is the case with Canopeo (Campana et al., 2023).

Therefore, the objective of this study is to evaluate the efficiency of image analyzer in determining leaf coverage related to the quality of substrates for the development of vegetable seedlings.

2. Materials and Methods

The experiment was conducted in a greenhouse at the Federal University of Technology of Paraná, located in Londrina, Paraná, Brazil. The location is at a latitude of -23.310° South, longitude of 51.16° West, and situated at an altitude of 559 meters above sea level.

The planting of lettuce seeds of the Crespa Grand Rapids TBR variety was carried out in the second half of April 2022. The seedlings were grown in 128-cell (8x16) polystyrene trays, using 4 different types of substrates.

The experimental design adopted was the Completely Randomized Design (DIC), which consisted of four treatments and four repetitions. The treatments were formed by the following substrates:

- Control: Commercial vegetable substrate;
- Witness: Topsoil + Vermiculite in a 5:1 ratio;
- Humus: Topsoil + Vermiculite + Humus in a 5:1:1 ratio;
- Compost: Topsoil + Vermiculite + Compost in a 5:1:1 ratio.

The treatments were selected not only to identify the substrates that demonstrate the best results bus also to generate variations in leaf coverage indices among them, enabling the attainment of distinct values that can be detected by seedling development assessment systems.

Once germination occurred, leaf area analyses were evaluated every 3 days using two apps on a smartphone: Canopeo and GreenTest.

Canopeo is an image analysis tool that uses the RGB system to classify image pixels. The app selects green coverage pixels based on the R/G, B/G ratios and the excess green index. The result is a binary image with white pixels representing green coverage and black pixels representing the lack of green coverage, with the fractional value expressed as a percentage.

GreenTest is an app developed by the authors of this study that allows users to select the image to be analyzed using two buttons: one that activates the camera to capture an image and another that accesses the photo gallery. Image analysis is performed by storing the image's height and width information in pixels. Then, the app counts the number of green and non-green

pixels in the image and indicates the ratio of green pixels to total pixels, expressed as a percentage.

As an example, the digital image presented in Figure 1 was used in the processing of the applications. Figure 2 depicts the Canopeo application (Figura 2a) and the resulting post-processing (Figure 2b). Meanwhile, Figure 3 shows the GreenTest application (Figure 3a) and its corresponding image processing (Figure 3b). The original language of the application was retained.



Figure 1 - Processed digital image used in the applications for illustration purposes.

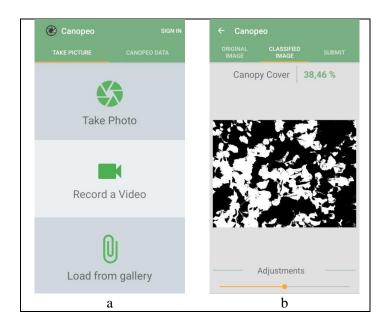


Figure 2 – Canopeo application (a) and its post-processing result (b).

Green Test	Green Test
Resolução original da imagem:	Resolução original da imagem: 3000x4000
CAPTURAR IMAGEM	Exibir paleta
Otimizar velocidade de análise:	Canais de análise:
Resolução otimizada:	Verde Vermelho Xerde
Porcentagem verde:	Limitador mínimo canal verde: 127
ANALISAR	Limitador máximo canal verde: 192
SALVAR IMAGEM	Otimizar velocidade de análise: Resolução otimizada: 400x400 Porcentagem verde: 39,20%
	ANALISAR
a	b

Figure 3 – GreenTest application (a) and its post-processing result (b).

The results obtained from Canopeo and GreenTest analyses were subjected to a comparison of means using the Scott-Knott method with a significance level of 5% to evaluate the performance of both methods.

At the end of the experiment, to evaluate the correlation between the results obtained with the apps, the values of Normalized Difference Vegetation Index (NDVI) were collected, which is an established method for assessing green coverage in plantations (Prabhakara et al, 2015), and the destructive tests related to green coverage: fresh mass, dry mass and moisture.

Samples collected for destructive testing followed the structure illustrated in Figure 4, considering 40 cells. The variables were plotted in scatter plots, performing linear regression linear regression and obtaining the R² to verify significant relationships.

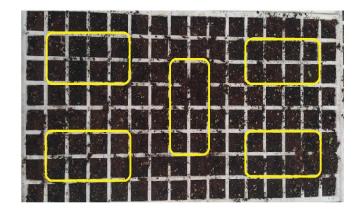


Figure 4 - Structure of cells used for destructive tests of fresh mass, dry mass, and humidity.

3. Results and Discussion

By determining the green coverage using Canopeo and GreenTest apps with a frequency of 3 days, we monitored the behavior of the lettuce in each of the treatments.

When submitting the results obtained from the Canopeo app to the mean comparison test using the Scott Knott method with a 5% significance level, we noticed that at the beginning of the experiment there were statistical differences between the treatments. However, after 12 days from the germination of the lettuce seedlings, we found that the Test treatment and the Humus

treatment presented similar leaf coverage values, which were statistically lower than the Control and Compost treatments, which were equal to each other. This condition was maintained until the end of the experiment, as shown in Table 1.

Table 1. Leaf coverage in lettuce seedlings: Mean comparison using Scott Knott method at 5% singnificance among

 Witness, Humus, Compost and Control treatments over time, measured through the Canopeo App

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	18/05	21/05	24/05	27/05	30/05	02/06	05/06	
Witness	9.886c	11.833b	20.030b	28.411a	28.741a	37.650a	42.420a	
Humus	7.383a	9.033a	16.240a	22.541a	34.263a	31.993a	38.241a	
Compost	8.554b	11.876b	22.541c	37.350b	46.231b	50.575b	65.960b	
Control	9.561c	14.288c	25.078c	39.220b	50.288b	59.491b	58.043b	

Regarding the values of green cover obtained with the GreenTest application, it was found that there was statistical equality between the treatments throughout the experiment, as can be seen in Table 2.

Table 2. Leaf coverage in lettuce seedlings: Mean comparison using Scott Knott method at 5% singnificance among Witness, Humus, Compost and Control treatments over time, measured through the GreenTest App

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	18/05	21/05	24/05	27/05	30/05	02/06	05/06
Witness	11.720a	18.690a	30.138a	40.553a	49.731a	53.726a	59.428a
Humus	10.411a	17.383a	29.251a	39.158a	51.983a	48.420a	56.500a
Compost	10.534a	15.071a	32.260a	44.873a	56.276a	63.930b	65.856a
Control	12.273a	18.790a	30.690a	45.870a	58.581a	60.411b	70.950a

It is observed that the Canopeo app is more sensitive in measurements, while the GreenTest app tends to standardize them. This difference is related to the procedure for obtaining values, as in Canopeo the measurement is directly performed, while in GreenTest an adjustment is required by the operator. Apps that do not require operator intervention bring more reliable results (Heinonen and Mattila, 2021; Qu et al, 2021).

When employing Normalized Difference Vegetation Index (NDVI) values dated 05/06 and applying the Scott Knott test with a significance level of 5%, it is observed that the Humus and Witness treatments exhibit statistical equality, with mean values of 0.295 and 0.314, respectively. However, the Control and Compost treatments demonstrate differences among themselves and in relation to the other treatments, with mean values of 0.400 and 0.475, respectively. These results resemble the behavior observed in the Canopeo application.

After obtaining the values of fresh mass, dry mass, and humidity for each treatment, scatter plots were generated with reflectance values and leaf coverages obtained through the Canopeo and GreenTest applications. Subsequently, a linear regression line was plotted, and the coefficient of determination (R^2) was calculated, as illustrated in Figures 5, 6 and 7.

In conducting linear regression and calculating the coefficient of determination (R^2) for the relationship between Fresh Mass and leaf coverage through the Canopeo APP, a percentage value of 88,67% was obtained (Figure 5a), close to that calculated for NDVI, which was 87,65% (Figure 5c). On the other hand, the leaf coverage values obtained through the Green Test app showed a slight difference, registering 82,83% (Figure 5b).

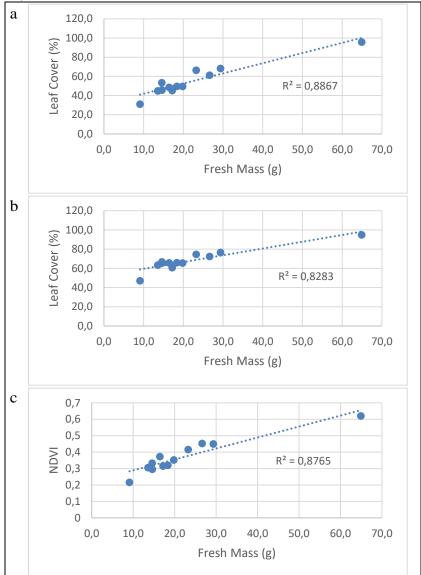


Figure 5 – Scatter Analysis, Linear Regression and Coefficient of Determination (R^2) for fresch mass and leaf coverage data obtained via applications Canopeo (a), GreenSeeker (b), and NDVI (c).

In the teste using Dry Mass, the leaf coverage estimated by the Canopeo app demonstrated a value of 88,51% (Figure 6a), while that obtained by GreenTest was 82,65% (Figure 6b), and NDVI recorded 87,63% (Figure 6c). It is observable that the values obtained by Canopeo approach those If NDVI and differ from the values of GreenTest.

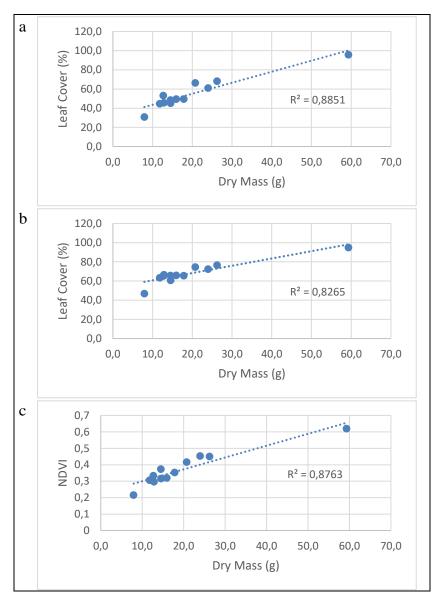


Figure 6 - Scatter Analysis, Linear Regression and Coefficient of Determination (R^2) for dry mass and leaf coverage data obtained via applications Canopeo (a), GreenSeeker (b), and NDVI (c).

When analyzing humidity, a correlation of 86,19% was observed with leaf through the Canopeo app (Figure 7a), 80,61% with the GreenTest app (Figure 7b), and 83,61% with NDVI.

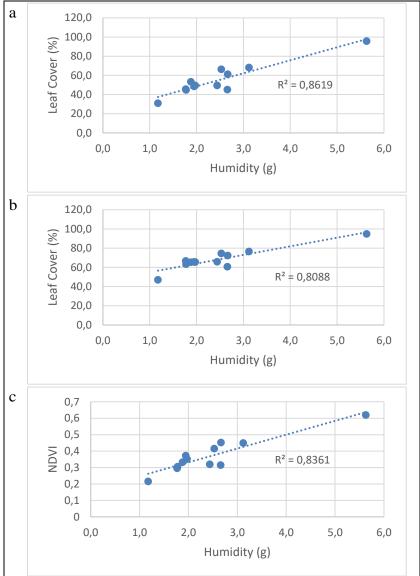


Figure 7 - Scatter Analysis, Linear Regression and Coefficient of Determination (R²) for humidity and leaf coverage data obtained via applications Canopeo (a), GreenSeeker (b), and NDVI (c).

The values provided by the Canopeo Application closely approximate the NDVI obtained via GreenSeeker, offering a non-invasive alternative to assess plant development. This finding is consistent with the finding of Hale et al. (2023), who highlight the Canopeo App as a low-cost and user-friendly tool for monitoring plant growth throughout their life cycle. On the other hand, a lower correlation is observed between NDVI values and the GreenTeste App.

4. Conclusions

Based on the result obtained in this study, there is a tendency for both the Canopeo App and the NDVI to demonstrate a strong correlation with destructive tests compared to the GreenTest App. Another finding made through the Scott Knott test over time is that the Canopeo App shows sensitivity in evaluating leaf coverage in lettuce seedlings similar to that of the GreenSeeker (NDVI), while the GreenTest App did not have sufficient sensitivity to identify statistical differences between the treatments under study. These findings suggest that the Canopeo App appears particularly promising as a viable alternative to destructive assays in assessing substrates for vegetable seedling development.

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Conflicts of Interest

The authors declare no conflict of interest.

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