



Article

Best Morpho-Physiological Parameters to Characterize Seed-Potato Plant Growth under Aeroponics: A Pilot Study

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Abstract: Although plant characterization under the International Potato Center's (CIP's) aeroponic system requires many morpho-physiological parameters to evaluate a cultivar, there is no method to evaluate the best parameters or the most suitable cultivation time. Thus, several morpho-physiological parameters were compared under a modified aeroponic system, using different statistical tools, to determine the best parameters and most efficient time to characterize seed-potato plants. We evaluated 21 parameters for cv. Agata under a randomized complete block design with weekly harvests for 9 weeks. The best parameters for growth characterization were selected based on multivariate statistical approaches involving correlation plots, similarity clusters (dendrograms), and principal component analysis. The best parameters for seed potato characterization were as follows, in order of importance: main stem diameter, leaf number, the length of the fourth leaf, leaf area, number of mini-tubers, mini-tuber fresh weight, root dry weight, and total dry weight. The days after transplanting (DAT) significantly affected the morpho-physiological parameters, with 45 DAT being the best cultivation time to estimate mini-tuber yield, and the data for bi-weekly harvests were as reliable as for weekly harvests. Our results, applied to either the CIP or to our modified aeroponics method, will be valuable in streamlining the characterization of other seed potato cultivars used by certified producers.

Keywords: biometric parameters; CIP-modified aeroponics; growth rates; mini-tubers; *Solanum tuberosum* L.



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1. Introduction

Aeroponic systems are gaining importance as an alternative to other traditional methods of producing seed-potato tubers, such as in pots and raised beds [1,2]. Aeroponic systems do not require a solid substrate, unlike traditional propagation systems. In this system, plants are grown with bare roots in a misty and dark enclosed environment where nutrient solutions are sprayed directly onto the plant roots, suspended in the air, at specific intervals. This growing technique facilitates enhanced oxygenation and nutrient uptake, stimulating potato plants' expedited and effective development [3,4]. Potatoes are a type of starchy root vegetable commonly grown in soil. However, due to virus transmission in the field, it is crucial to grow certified seed potatoes under greenhouse conditions using aeroponics. In aeroponics, plants are grown in a closed system where their roots are periodically suspended in the air and sprayed with a nutrient-rich mist, as previously

elucidated [3]. This method has several potential advantages over traditional soil-based agriculture, including faster growth, improved water and nutrient efficiency, a nematode-, virus-, and bacteria-free environment, and a more precise growing-environment control [5].

Aiming at the development of seed-potato production and dissemination of virus-free genetic material, the “Centro Internacional de la Papa” (CIP), or International Potato Center, in Lima, Peru, was established in 1971 with its main goal focused on reducing poverty and maintaining food security in developing countries through sound scientific research for the sustainable production of potatoes and other tubers. The CIP proposed using aeroponics to produce seed-potato tubers in 2007 [6]. The system consisted of a wooden box, lined with StyrofoamTM sheets and black plastic, containing misting nozzles fed by irrigation pipes. The nozzles deliver the nutrient solution to the roots and include a return electric pump connected to the nutrient solution source [1]. However, no published data were found that evaluate the potato morpho-physiological parameters proposed by CIP for aeroponic systems. Recently, we reported that the type of spray nozzle and the spray direction can play significant roles in several mini-potato morphological parameters and that the fogger sprayer with a downward spray at 12 L h⁻¹ provided the highest yield of mini-tubers [7]. This modified system was used in the current study to test the morpho-physiologic parameters.

Plant growth-rate analysis requires the quantification of morphological and physiological traits. Morpho-physiological parameter analysis can reveal how efficiently plants synthesize and allocate photoassimilates to different storage organs [8,9]. Measuring plant growth is the most direct way to determine sunlight conversion efficiency into plant biomass [10,11].

Factors such as cultural practices, nutrient availability, electrical conductivity, the pH of the nutrient solution, spray frequency, cultivar, and plant density can affect the development and productivity of potato plants grown in aeroponics [2,12,13]. High plant densities can cause self-shading and hormonal interactions, resulting in tall and spindly stems and reducing the productivity of mini-tuber seed potatoes [14–16].

Specialized analytical tools and methods are essential in understanding the variations in growth patterns, tissue accumulation, and overall plant productivity [17]. Analyzing and interpreting the nuances of potato plant development requires a thorough understanding of the intricate interplay between environmental factors, genetic makeup, and physiological responses [18,19]. During growth, specialized tools, techniques, and statistical analyses can provide valuable insights into the underlying mechanisms of potato mini-tuber productivity. Data on the morpho-physiological aspects of potato plants are essential for comparing the effectiveness of potato seed tuber production strategies [20].

Aeroponics produces virus-free mini-tubers under controlled greenhouse conditions and plant morpho-physiological parameters can be correlated with plant tuber yield and fresh weight [21,22]. If these indices are also used to predict regular potato tuber production, they can help growers and researchers estimate edible potato-tuber yield under aeroponics [23].

We conducted a pilot study to evaluate and improve the efficiency of the current methodology for evaluating potato cultivars under aeroponics. The pilot study will assess the practicability of the current experimental framework under multiparametric statistics and suggest ways to make the method more effective. This strategy may improve the dependability and accuracy of future research efforts.

The key differences between our modified aeroponic system and the CIP aeroponic system are that our aeroponic system includes a compact box size, which is particularly suitable for small-scale research purposes such as studying nutrients, nutrient solutions, salinity, trace elements, genotype analysis, and the production of bite-sized potatoes in urban agriculture for food security. Our system can be expanded and adapted for the commercial production of seed potatoes and small potatoes for human consumption. Additionally, each wooden box is connected to a reservoir with a water pump, providing the advantage of true replication for each experimental unit. Individual wooden boxes

allow for easy control and prevent diseases from spreading to other plants. The wooden box in our proposed aeroponic system is elevated approximately 50 cm above ground level. This allows for the easy return of the nutrient solution to the reservoir, eliminating the need to position the reservoir underground. Furthermore, our aeroponic system offers enhanced maneuverability for the boxes, reservoirs, and water pumps. Although several plant morpho-physiological parameters can be used to characterize potato genotypes under this International Potato Center's (CIP's) modified aeroponic system, the main goal of our work was to use the 'Agata' genotype as a model system to establish which growth and physiological parameters are most effective in characterizing potato genotypes for growth and seed-potato yield under aeraponics.

2. Materials and Methods

2.1. Location and Conditions

An experiment was conducted at the Federal University of Viçosa in the state of Minas Gerais, Brazil (20°45'27.05" S, 42°52'11.63" W, 649 m (2129 feet above sea level). The experiment took place in a greenhouse with an evaporative cooling system and was conducted from August to October of 2013. The average temperatures, daily photoperiod, and relative humidity were 25.2 °C (77.4 °F), 12.1 hrs., and 54.7%, respectively (Figure 1). Night and day temperatures were within the 18.3 to 26.7 °C (day, 65.0 to 80.0 °F) and 12.8 to 18.3 °C (night, 55.0 to 65.0 °F) ranges recommended for optimal potato growth [24,25]. The increases (peaks) in relative humidity (RH), corresponding to decreases (dips) in temperature, are a reflection of rainfall events outdoors. Photoperiod and RH slowly increased as the season changed from winter to spring.

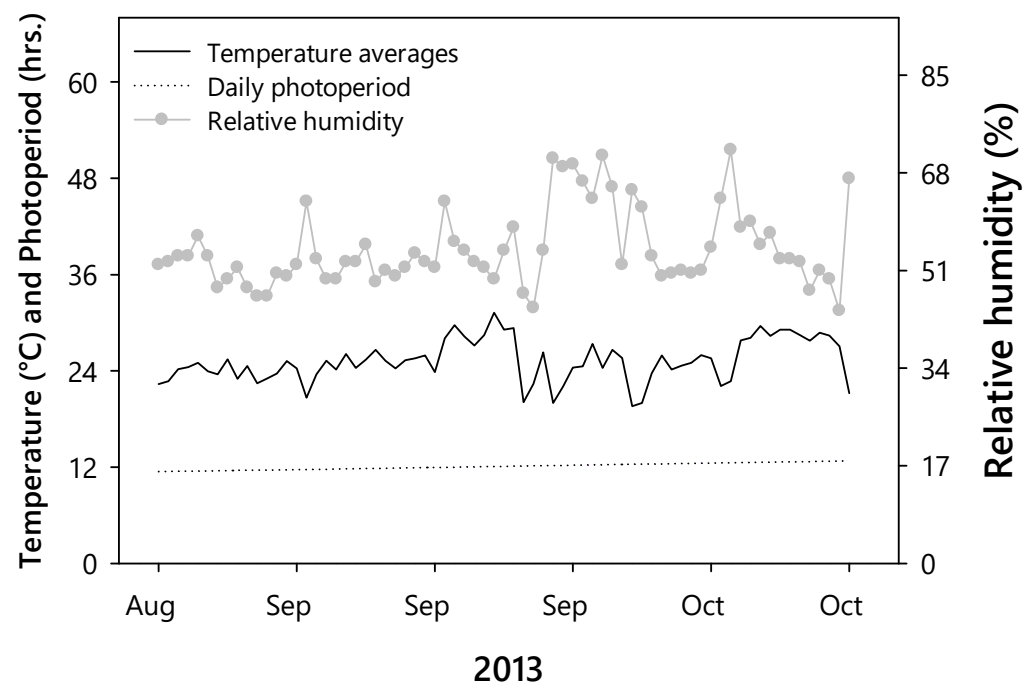


Figure 1. Temperature averages, daily photoperiod, and relative humidity averages during the pilot study under greenhouse conditions. Source: temperature and relative humidity (personal data logger); photoperiod https://www.sunrise-and-sunset.com/pt/sun/brasil/vicosa__15 (accessed on 9 February 2024).

2.2. Aeroponic System

The propagation material used sprouted from pre-nuclear seed tubers of the cv. Agata. Pre-nuclear seed tubers, or pre-nuclear seed plantlets, refer to microtubers or other tissue-cultured materials created in a laboratory and are free of virus [26]. The sprouts were submerged for 2 min in a 2% sodium hypochlorite solution before being thoroughly rinsed for 5 min with tap water [27]. Subsequently, the sterilized sprouts were placed into 72-cell polystyrene seed trays filled with Tropstrato Vegetables®. The present substrate did not undergo sterilization. The composition consisted of peat and expanded vermiculite. Peat enhances soil fertility by supplying organic material and retaining moisture, whereas expanded vermiculite improves soil structure by promoting aeration and facilitating drainage. This combination provides an optimal environment for the early development of potato sprouts; typically, there are no concerns about contamination. The sprouts were cultivated in seed trays using the Tropstrato Vegetables® substrate for 15 days. This phase is critical for the formation of roots and the growth of a robust root system. Subsequently, seedlings measuring 17 cm were thoroughly rinsed with tap water to eliminate their substrate. After cleaning rooted plants, seedlings were transferred to our aeroponic system. Seedlings were placed in the aeroponic system at a density of 12 plants m⁻² (Figure 2). This controlled planting arrangement optimizes space utilization.



Figure 2. Potato plant growth cv. Agata after transplanting under our aeroponic system at a density of 12 plants per m².

The aeroponic system consisted of a rectangular wooden box 1.0 m long, 0.6 m wide, and 0.7 m high. The nutrient solution was applied using two downward misting nozzles placed 0.3 m apart inside each box. Box walls were constructed of 50 mm (2 inches)-thick Styrofoam™ sheets with removable windows to facilitate the harvesting of the mini-tubers. The inside of the box was lined with the black side of a Panda film 5.5-mil thick (0.14 mm) black-and-white poly-film to prevent light penetration (<https://www.homedepot.com/p/Hydro-Crunch-Panda-Film-100-ft-x-10-ft-Black-and-White-Poly-Film-5-5-Mil-D940011100/302968943>, accessed on 7 February 2024). The irrigation system consisted of a 60-L bucket, a ½-horsepower electric water pump, 25-mm-diameter PVC tubing, and a MA-30 nozzle. The MA-30 nozzle had a flow rate of 34 gallons per hour at a pressure of 3 atm without drip protection. A digital timer, which alternated between 25 s of pumping and 3 min of rest, controlled the pumps of the experiment. Perforations were made in the box lids to facilitate the installation and cultivation of transplanted seedlings 15 days after rooting. Figure 3 shows a schematic representation of the whole propagation process and the acquisition of results.

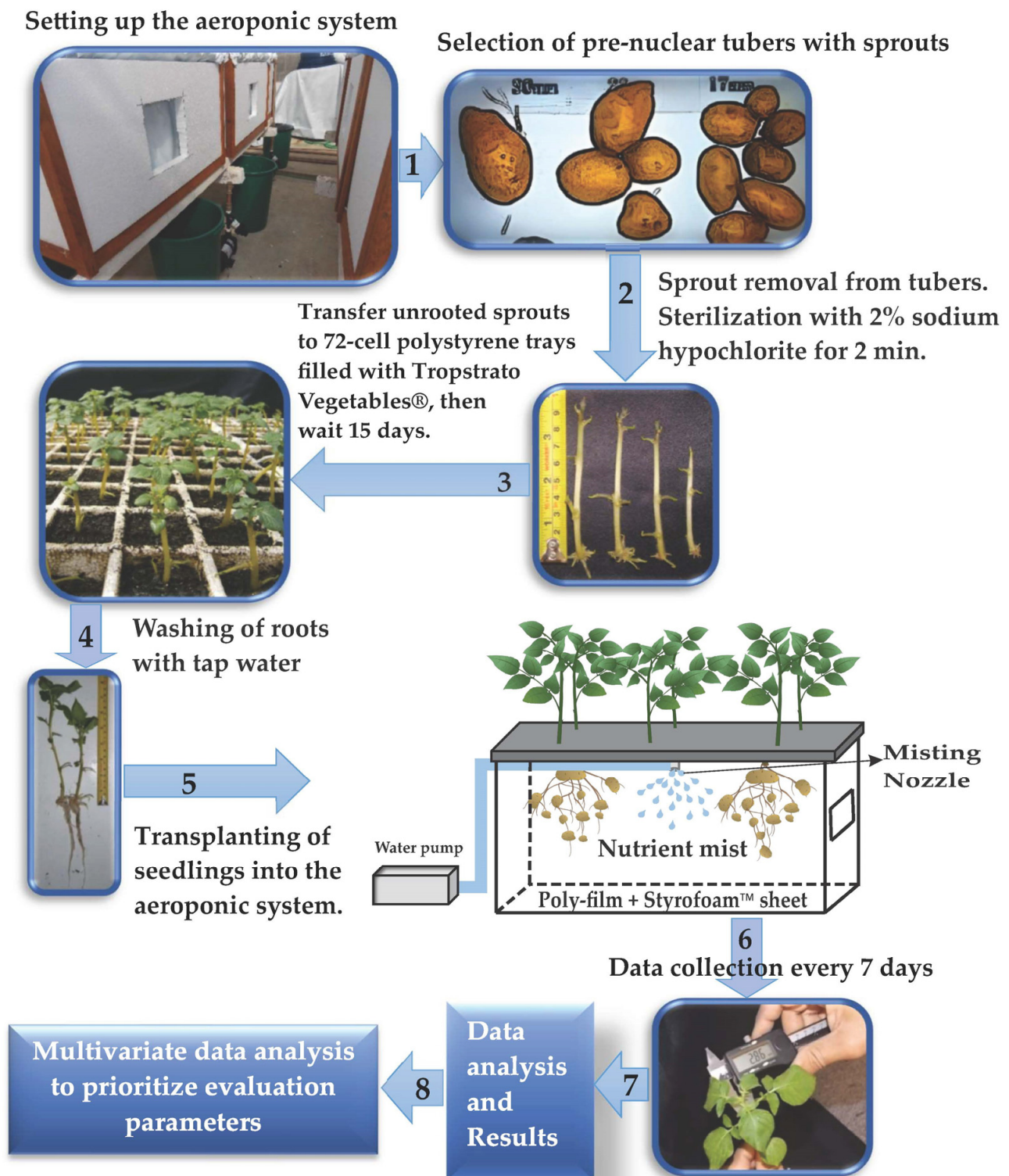


Figure 3. Flux diagram, from system setup to the acquisition of results, of the aeroponic propagation process and system used to characterize the seed-potato cv. Agata.

2.3. Design

The experiment used a randomized complete block design (RCBD) with eight replications ($n = 8$) for each parameter evaluated in each of the nine harvest periods, totaling 72 plants evaluated during the experimental period, which started 7 days after transplanting (DAT). Each harvest period had a 7-day interval. Plant growth characteristics and physiological aspects were evaluated from 7 to 63 DAT in each of the nine harvest periods.

2.4. Nutrient Solution

We used the nutrient solution proposed by Otazú et al. [1], as recommended in the manual for quality seed-potato production under aeroponics recommended by the International Potato Center (CIP). The salts in this solution were magnesium sulfate heptahydrate ($\text{MgSO}_4 \cdot 7\text{H}_2\text{O}$), ammonium nitrate (NH_4NO_3), potassium nitrate (KNO_3), calcium dihydrogen phosphate monohydrate ($\text{Ca}(\text{H}_2\text{PO}_4)_2 \cdot \text{H}_2\text{O}$), copper sulfate (CuSO_4), zinc sulfate heptahydrate ($\text{ZnSO}_4 \cdot 7\text{H}_2\text{O}$), manganese sulfate monohydrate ($\text{MnSO}_4 \cdot \text{H}_2\text{O}$), boric acid (H_3BO_3), ammonium molybdate tetrahydrate ($(\text{NH}_4)_6\text{Mo}_7\text{O}_{24} \cdot 4\text{H}_2\text{O}$), ferric chloride hexahydrate ($\text{FeCl}_3 \cdot 6\text{H}_2\text{O}$), and ethylenediaminetetraacetic acid (EDTA). Furthermore, as stated by Otazú [1], the NH_4NO_3 content in the nutrient solution decreased by 50% 36 days after transplanting (Table 1). Research by Silva Filho et al. [23] showed that this nutrient solution consistently kept the salt level below the crucial threshold for potato plants.

Table 1. The concentration of nutrient ions in Otazú's nutrient solution at 100% of the crop recommendation rate used during the evaluation of the aeroponic system.

Nutrient	Concentration	
	Up to 35 DAT	36 to 63 DAT
	mmol _c L ^{−1}	
Nitrate	9.80	7.60
Ammonium	4.40	2.20
Phosphorus	2.60	2.60
Potassium	5.40	5.40
Calcium	1.30	1.30
Magnesium	1.00	1.00
Sulfur	1.00	1.00
	μmol L ^{−1}	
Iron (Chelated)	151.97	151.97
Manganese	8.74	8.74
Boron	98.15	98.15
Zinc	2.75	2.75
Copper	2.83	2.83
Molybdenum	0.13	0.13
Electrical Conductivity (dS m ^{−1})	1.60	1.35

DAT: days after transplantation.

The nutrient solution used in this study was calculated using the principle of cation–anion balance [28]. The initial electrical conductivity (EC) value was 1.60 deciSiemens per meter (dS m^{−1}), and the pH value was 5.5. The pH was adjusted to a value of 5.5 before measuring the EC. The reservoirs were replenished with deionized water every two days to maintain the level of the nutrient solution, returning it to its original level at the time of initial production [29]. When the EC of the nutrient solution decreased by 30% from its initial concentration, it was replaced by a fresh nutrient-balanced solution.

2.5. Data Collection

We provided a flowchart to summarize the data collection from our study with the cultivar Agata under our aeroponic system (Figure 4).

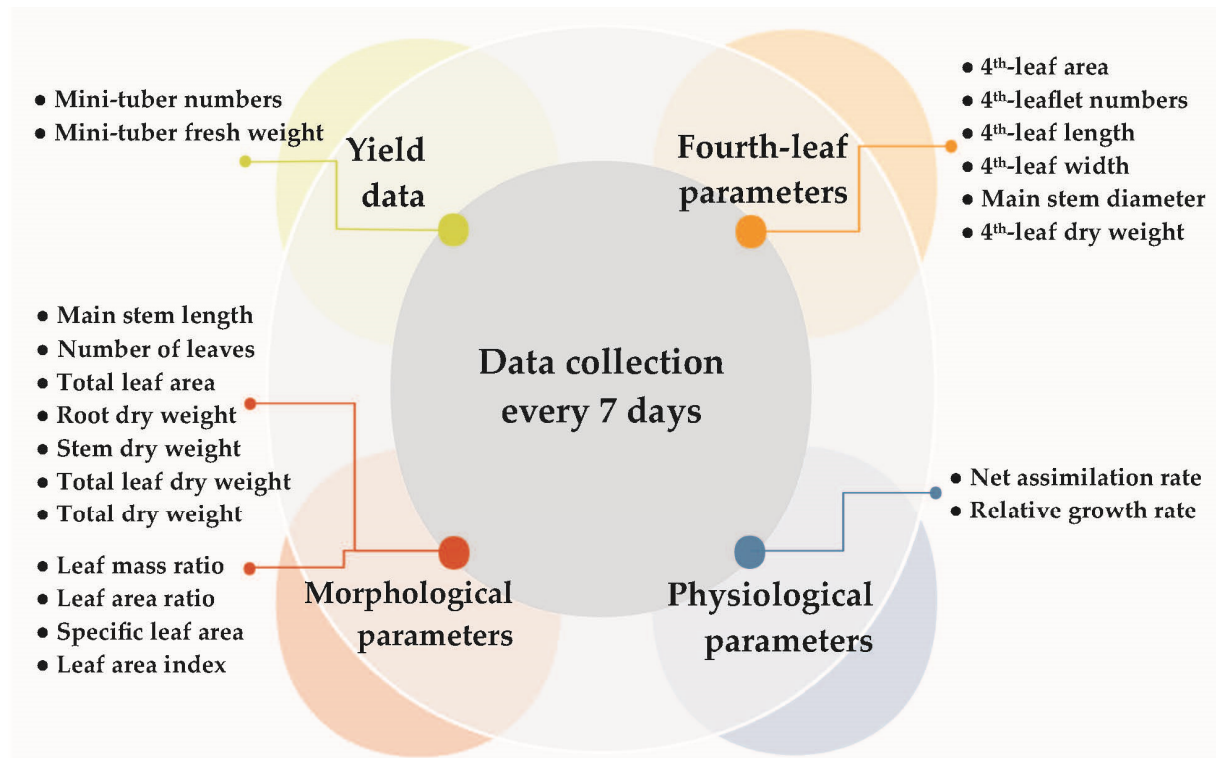


Figure 4. Flowchart of the variables evaluated for the cv. Agata under our aeroponic system.

2.5.1. Morphological Aspects

The parameters evaluated were main stem length (SL), number of leaves (LN), total leaf area (TLA), and biomass production, the latter including the dry weights of roots and stolons (RDW), stems (SDW), leaves (LDW), and the total plant dry weight without mini-tubers (TDW).

The leaf mass ratio (LMR) to dry weight was one of the morphological parameters considered. The LMR was calculated by dividing the mass of all leaves on a single plant (LW) by the total plant dry weight (W). The LMR represents the proportion of plant biomass allocated to leaves as a percentage of the total plant dry weight. The total leaf area (LA) per unit of total plant dry weight (W) was used to calculate the leaf area ratio (LAR). The ratio of leaf area (LA) per unit leaf dry weight (LW) is represented by specific leaf area (SLA) [30,31].

$$SLA = \frac{LA}{LW},$$

where LA = leaf area and LW = leaf dry weight.

The leaf area index (LAI) [31] was calculated using digital image analysis (DIA) as green leaf area per unit of soil area, considered a value [32,33], using the following formula:

$$LAI = \frac{LA}{SA},$$

where LA = leaf area and SA = surface area.

2.5.2. Parameters of the fourth leaf

The fourth leaf, counted from the plant's apex, was used as the reference leaf. This leaf is fully expanded and is considered the standard for morphometric measurements [34]. The area of the fourth leaf (4LA) was calculated using digital image analysis (DIA) and the WinDias 3 Delta-T program (<https://delta-t.co.uk/product/wd3/>, accessed on 5 February 2024). The leaflet numbers (LLN), length (4LL), and width (4LW) of the reference leaf were measured using a millimeter ruler. A digital caliper was used to measure the diameter of the main stem (SD) at the base of the fourth leaf. Fourth leaf dry weight (4LDW) was also determined.

2.5.3. Physiological Aspects

The net assimilation rate (NAR) is the ratio of leaf mass to leaf area produced during each harvest [23,24]. The NAR was calculated using the following formula [35]:

$$NAR = \left(\frac{W_2 - W_1}{t_2 - t_1} \right) \times \left(\frac{\ln L_{A2} - \ln L_{A1}}{L_{A2} - L_{A1}} \right),$$

where W_2 and W_1 = plant dry weights, which were measured at time 2 and 1, respectively; $\ln$ = natural logarithm; L_{A2} and L_{A1} = leaf areas, which were calculated at time 2 and 1, respectively; and t_2 and t_1 = time 2 and 1, respectively, which are the times between two subsequent harvests.

As an estimate of biomass production over time, the relative growth rate (RGR) was calculated as the amount of biomass (dry weight) produced relative to existing biomass in each time interval [32,33]. The RGR was calculated using the following formula [35]:

$$RGR = \frac{\ln W_2 - \ln W_1}{t_2 - t_1},$$

where $\ln$ = natural logarithm, W_2 and W_1 = plant dry weight, collected at time 2 and 1, respectively, and t_2 and t_1 = time 2 and 1, respectively.

2.5.4. Yield

The mini-tubers were measured for their number (TUN) and fresh weight (TUF). The mini-tubers with a diameter of 8 mm or greater were collected at each harvest period.

2.6. Data Analysis

The relationship between the two sets of data was analyzed using regression analysis. Different models were selected for each measurement based on previously recognized patterns, such as the sigmoidal curve of plant biomass over time. The model was chosen based on biological logic, the t -test results for statistical significance of regression coefficients ($p < 0.10$), and a coefficient of determination (R^2) value greater than 0.75. A multivariate approach was then used to optimize the number of variables and times evaluated. For this purpose, a Pearson correlation was carried out to evaluate the data, and a cluster analysis (dendrogram) using the Euclidean distance for data independence and principal components analysis (PCA) was used to understand the contribution of the variables in the characterization of morphological and physiological parameters. All analyses were performed using R software version 4.3.2 (R Core Team 2023, Vienna, Austria) [36], SAEG version 9.1 [37], and SigmaPlot/SigmaStat (Version 15.0, Systat Software, San José, CA, USA [www.systatsoftware.com], accessed on 7 February 2024) for conducting statistical analysis.

3. Results

3.1. Morphological Aspects

As expected, both leaf number and stem length increased throughout the study until plants entered the senescence stage at 63 days after transplanting (DAT), with a maximum final average of 216 leaves per plant and 93 cm for stem length (Figure 5A). Thus, 63 DAT was the final data harvest time. A similar trend was observed for the total leaf area, which increased at each sampling date, reaching a maximum average of 4673.96 cm² plant⁻¹ at 63 DAT (Figure 5B).

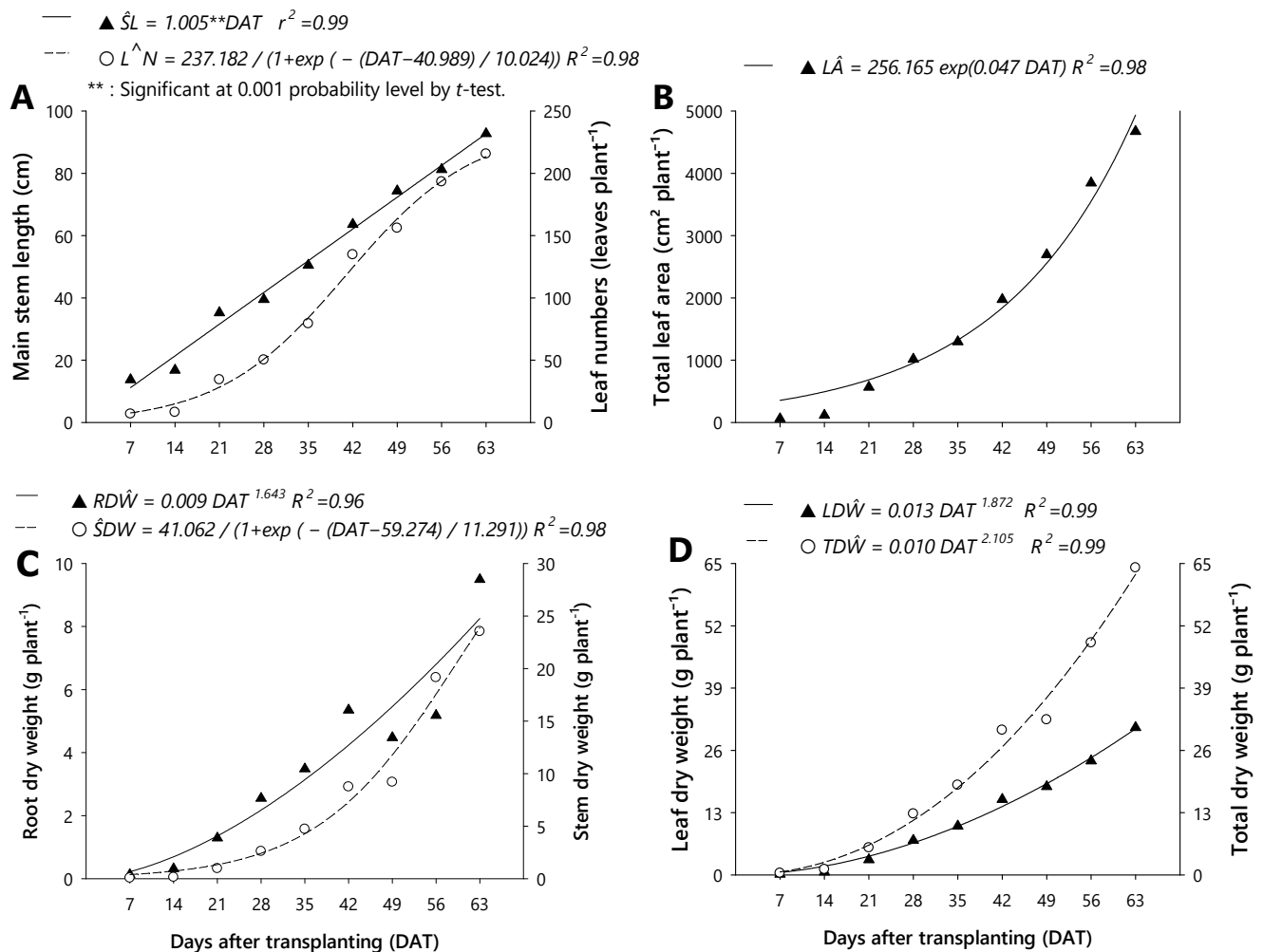


Figure 5. Plant growth responses based on main stem length and leaf number (A), total leaf area (B), root and stem dry weight (C), total leaf and total dry weight (D) of cv. Agata under our aeroponic system, with estimated coefficients of determination (r^2 , R^2). DAT = days after transplanting.

At 63 DAT, the average dry weight of roots, stems, leaves, and the total plant dry weight increased in a pattern similar to the number of leaves, reaching 9.49, 23.52, 30.76, and 64.09 g plant⁻¹, respectively (Figure 5C,D).

Throughout the study, the measurements of the reference leaf (fourth-leaf area, fourth-leaf dry weight, fourth-leaf length, fourth-leaf width, and main stem diameter) followed a sigmoidal pattern, stabilizing at 35 DAT. In that order, the maximum average values were 76.5 cm², 0.33 g, 18.3, 14.1 cm, and 6.3 mm (Figure 6A–C).

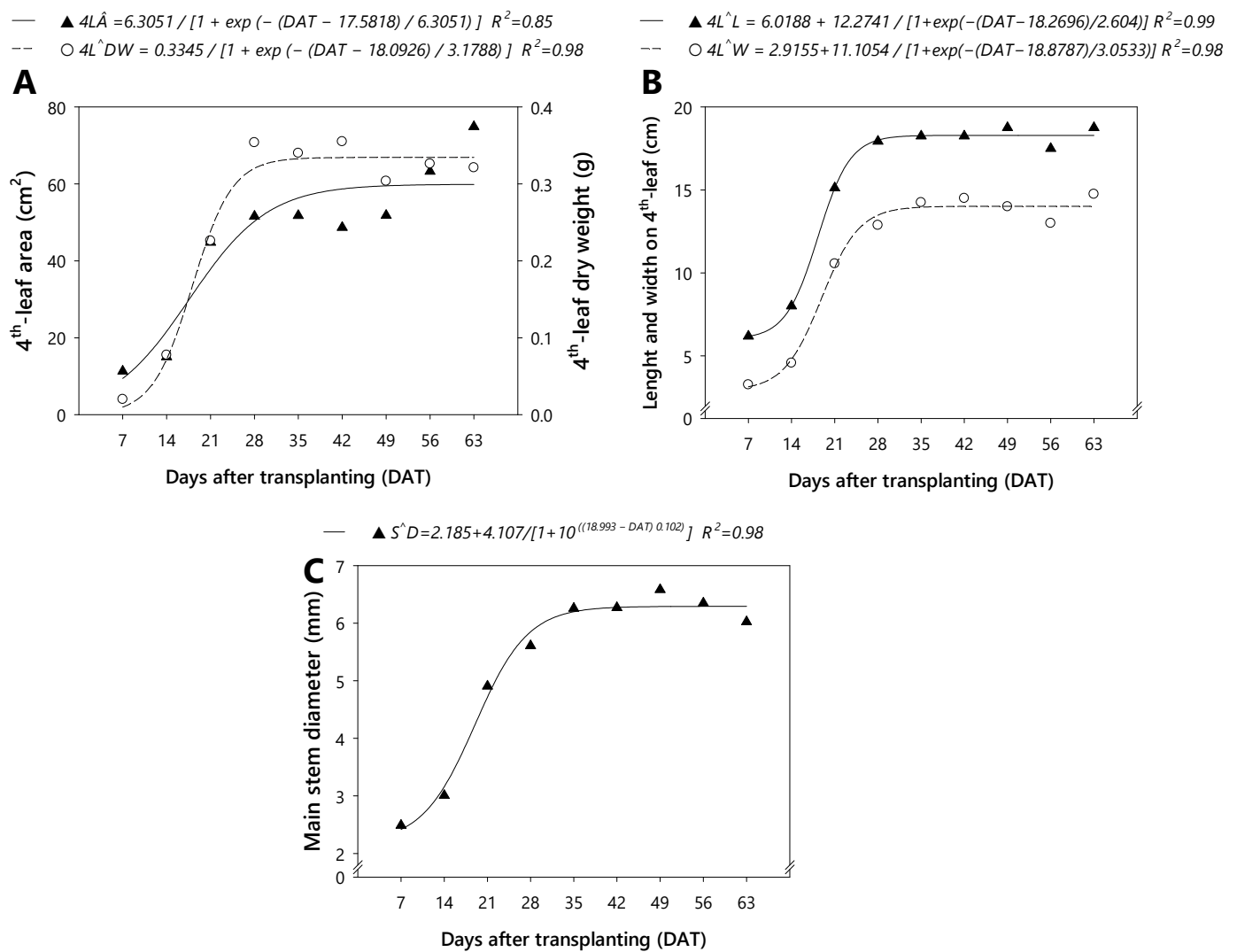


Figure 6. Plant growth response expressed through the 4th-leaf area, 4th-leaf dry weight (A), 4th-leaf length and width (B), and stem diameter (C) of cv. Agata under our aeroponic system throughout the study, with estimated coefficients of determination (R^2). DAT = days after transplanting.

Under the CIP system, leaf mass ratio (LMR) measurements followed a square root linear pattern over time, reflecting the balance of photosynthetic tissue to total plant biomass (Figure 7A).

The leaf area ratio (LAR) and specific leaf area (SLA) decreased exponentially. They stabilized at 28 DAT with mean values of 77 and 145 cm² g^{−1}, respectively, according to the equations describing the curves (Figure 7A,B).

The leaf area index (LAI) increased exponentially while the net assimilation rate (NAR) decreased, possibly due to self-shading (Figures 7B and 8). At 7 and 63 DAT, the initial and final average LAI values were 0.10 and 7.48 cm² cm^{−2}, respectively.

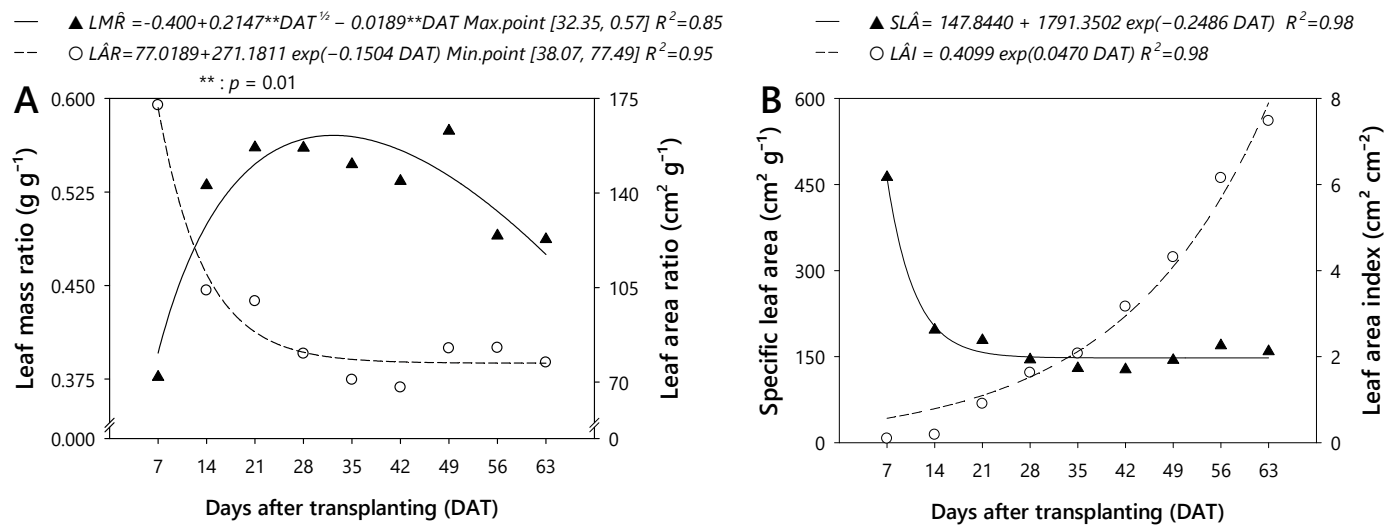


Figure 7. Plant growth responses based on leaf mass ratio, leaf area ratio (A), specific leaf area, and leaf area index (B) of cv. Agata under our aeroponic system throughout the study, with estimated coefficients of determination (R^2). DAT = days after transplanting.

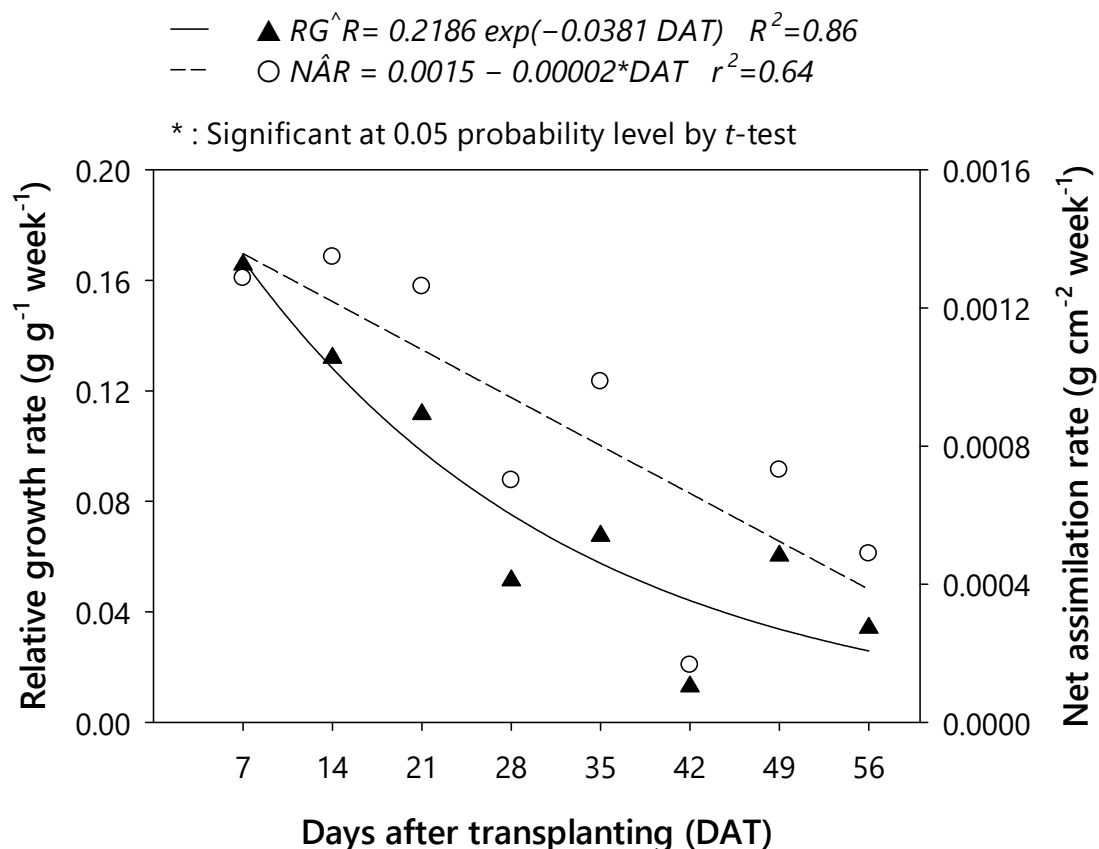


Figure 8. Plant growth responses based on relative growth rate (RGR) and net assimilation rate (NAR) of cv. Agata under our aeroponic system throughout the study, with estimated coefficients of determination (r^2 , R^2). DAT = days after transplanting.

3.2. Physiological Aspects

The study's relative growth rate (RGR) decreased over time, following a negative exponential curve. The initial value at 7 DAT was $0.1656\ g\ g^{-1}\ week^{-1}$, which decreased to $0.03417\ g\ g^{-1}\ week^{-1}$ at 56 DAT (Figure 8). From 7 to 21 DAT, the highest RGR and

NAR values were observed at $0.17 \text{ g g}^{-1} \text{ week}^{-1}$ and $0.0014 \text{ g cm}^{-2} \text{ week}^{-1}$, respectively (Figure 8).

As the RGR decreased, the leaf area ratio and specific leaf area remained constant (Figures 7A and 8). This indicates that plants were not producing above-ground tissues at that time, most likely due to resource allocation to tuber production.

3.3. Yield

Tuber yield started at 18 DAT and increased throughout the study. Still, the highest increase was from 18 to 45 DAT (Figure 9). Tuber number and fresh weight increased in a sigmoidal and exponential pattern, respectively. Figure 9 shows that productivity peaked at $74.6 \text{ tubers plant}^{-1}$ at 45 DAT. The average fresh weight of the mini-tubers at 42 DAT and 63 DAT was 82 g/plant and 439 g/plant , respectively. Mini-tubers with a diameter of 8 mm or more were harvested as previously indicated. The average mini-tuber yield per plant was 415.88 mini-tubers per plant and 960.63 g per plant across all harvest events, resulting in an average productivity of 6654 mini-tubers per square meter and 15,370 g per square meter.

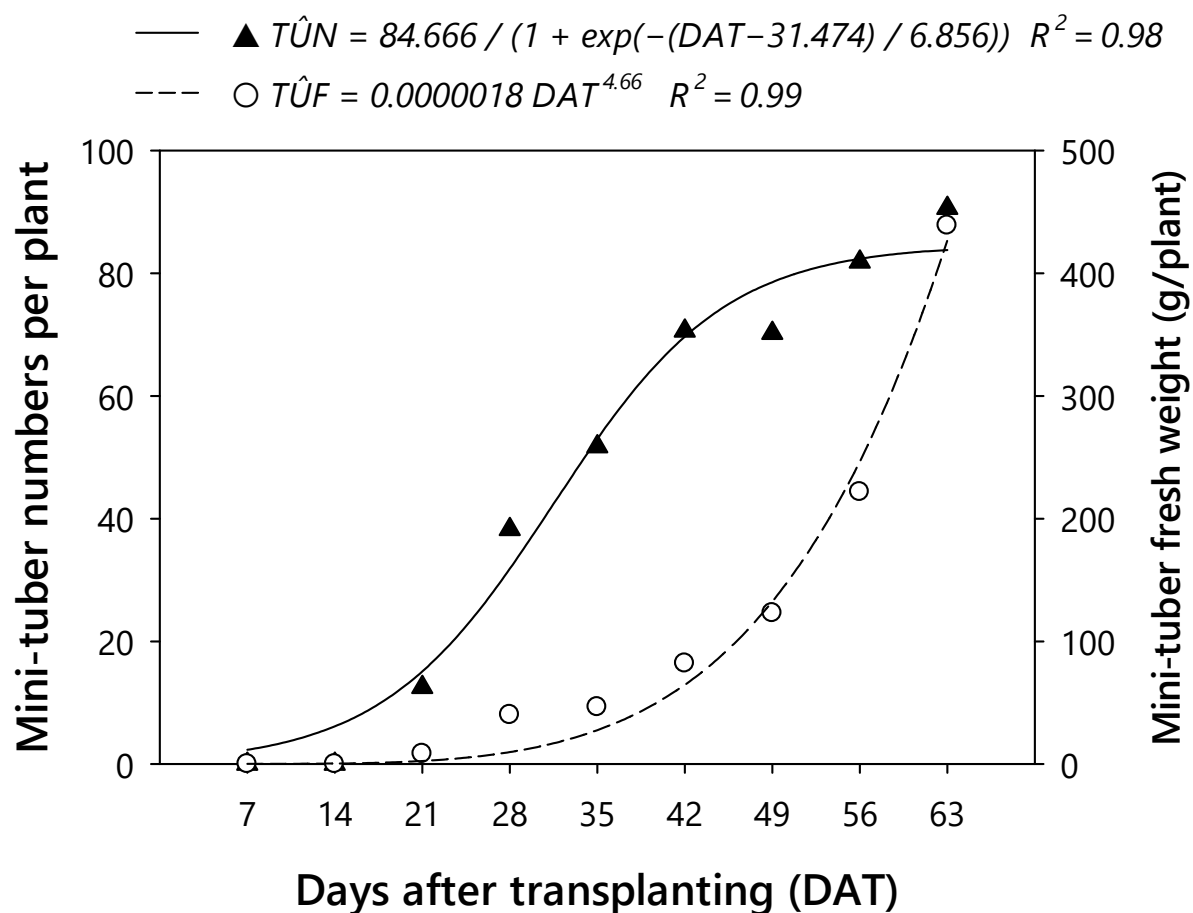


Figure 9. Plant growth responses through mini-tuber numbers (TUN) and mini-tuber fresh weights (TUF) of cv. Agata under our aeroponic system throughout the study, with estimated coefficients of determination (R^2). DAT = days after transplanting.

3.4. Multivariate Analyses

Multivariate analysis graphs representing correlations between the parameters (COR-RPLOT), Principal Component Analysis (PCA), and a dendrogram of the hierarchical clustering based on the relative values of DAT and morpho-physiological parameters of mini-tuber seed potatoes are presented in Figure 10, Figure 11, and Figure 12, respectively.

For the parameters evaluated in the aerial part (SL, SD, LN, LLN, 4LL, 4LW, 4LA, LA, LMR), the root system parameters (TUN, TUFW, and RDW) and the dry mass analyses (RDW, SDW, 4LDW, LDW, and TDW) showed positive and significant correlations. On the other hand, LAI values suggest low importance, i.e., poorly correlated with LMR (0.17) or negatively correlated (not important) for the parameters LAR (−0.49) and SLA (−0.42) (Figure 10).

Physiological parameters such as RGR and NAR show a certain antagonism to the area variables, being significantly negatively correlated, except for leaf area ratio (0.61 and 0.51) and SLA (0.4 and 0.29), where the correlations were not significant (Figure 10).

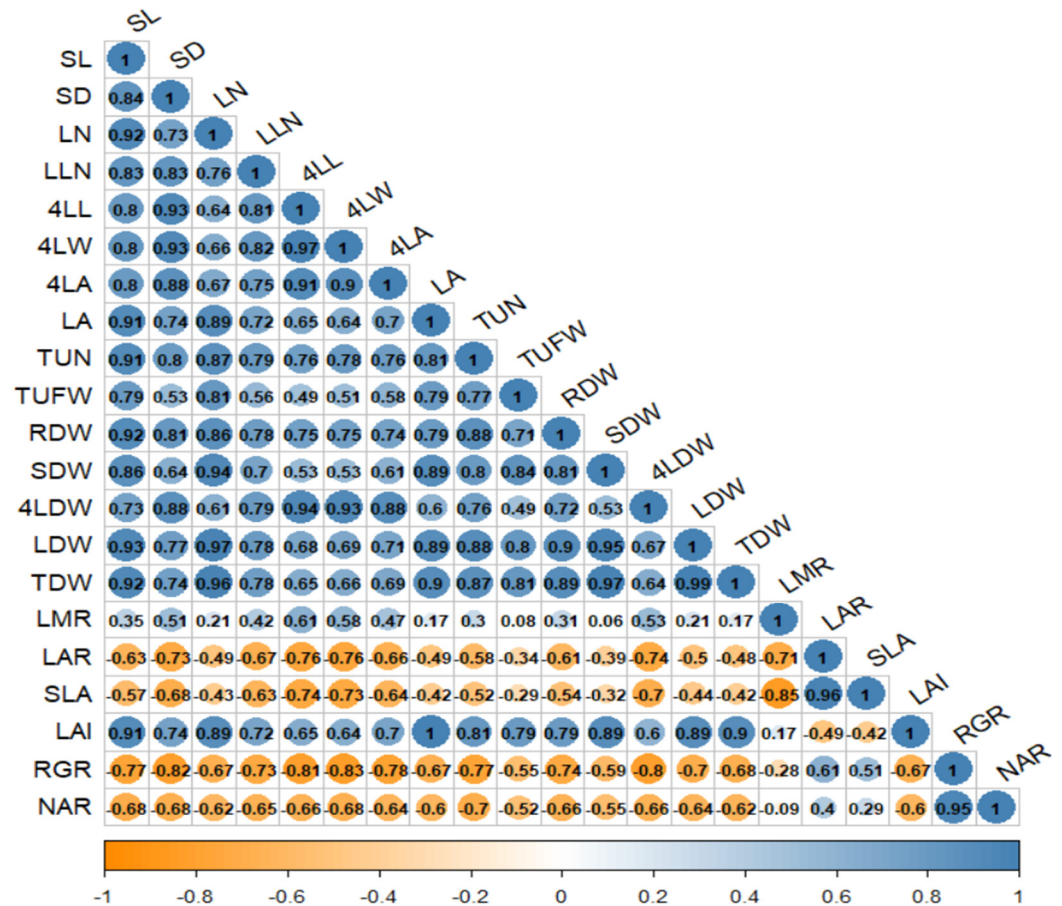


Figure 10. Correlations between the evaluated parameters, shades of blue (positive) and orange (negative), their intensity, and circle size determine the magnitude of the correlation on the CIP aeroponic system. Caption: SL—main stem length, SD—stem diameter, LN—leaf numbers, LLN—leaflet numbers, 4LL—4th leaf length, 4LW—4th leaf width, 4LA—4th leaf area, LA—leaf area, TUN—mini-tuber numbers, TUFW—mini-tuber fresh weight, RDW—root dry weight, SDW—stem dry weight, 4LDW—4th leaf dry weight, LDW—leaf dry weight, TDW—total dry weight, LMR—leaf mass ratio, LAR—leaf area ratio, SLA—specific leaf area, LAI—leaf area index, RGR—relative growth rate and NAR—net assimilation rate.

Using two dimensions, it was possible to perform a Principal Component Analysis (PCA), which explained that approximately 94% of all the variation obtained in the experiment was contained in two dimensions (PC 1: 79.8% and PC 2: 13.6%). In this sense, it was possible to identify the parameters that varied little, such as the physiological parameters (RGR and NAR), which contributed little to the results obtained in the first dimension (X). The parameters LAR and SLA stood out on the X-axis (first dimension, PC 1), accounting for 40% of the total variation (Figure 11).

It was also evident that the harvest event at 7 days after transplanting (DAT) had the highest relative growth rate (RGR), leaf area ratio (LAR), and specific leaf area (SLA). This supports the results shown in Figure 7A,B and Figure 8. At 14 DAT, the harvest event had the highest net assimilation rate.

Assessments at 49 and 56 (DAT) provided a good capture of the variability observed in the experiment; despite being far from the variables, harvesting at 56 (DAT) resulted in the maximum production of mini-tubers, main stem length, leaf area index, biomass, and total leaf area. In the period from 28 DAT to 35 DAT, there was an increase in the leaf mass ratio (Figure 11).

Furthermore, the parameters 4th-leaf dry weight, 4th-leaf length, 4th-leaf width, main stem diameter, 4th-leaf area, and 4th-leaflet numbers are inserted in the same quadrant, which includes 28, 35, and 42 days after transplanting (DAT). In this sense, the parameter evaluations can be represented by only one evaluation at 35 (DAT), as this managed to capture the greatest variation obtained in the experiment (Figure 11).

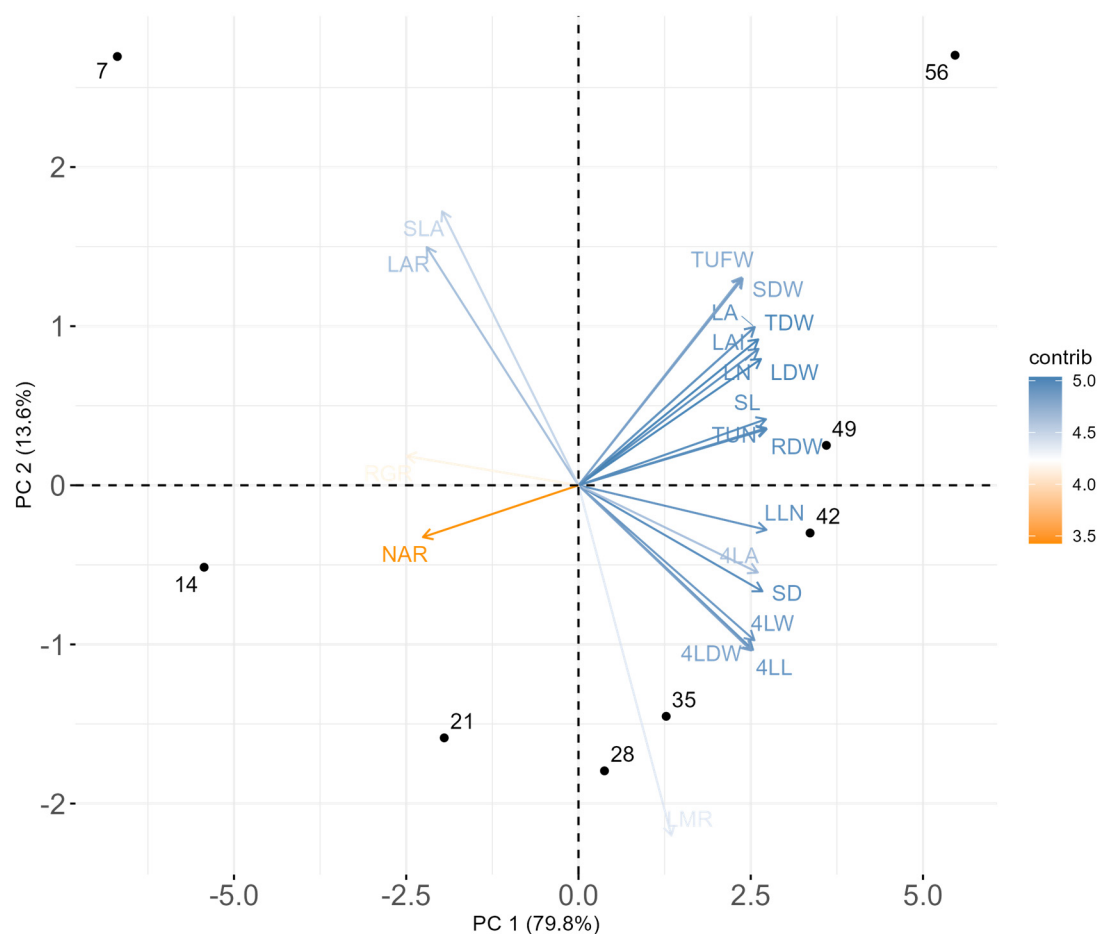


Figure 11. Principal component (PC) analysis, shades of blue (positive) and orange (negative), their intensity, and line size determine the magnitude of the contribution of the parameters to the experiment's overall variation under our modified-CIP aeroponic system. Project lines: SL—main stem length, SD—stem diameter, LN—leaf numbers, LLN—leaflet numbers, 4LL—4th-leaf length, 4LW—4th-leaf width, 4LA—4th-leaf area, LA—leaf area, TUN—mini-tuber numbers, TUFW—mini-tuber fresh weight, RDW—root dry weight, SDW—stem dry weight, 4LDW—4th-leaf dry weight, LDW—leaf dry weight, TDW—total dry weight, LMR—leaf mass ratio, LAR—leaf area ratio, SLA—specific leaf area, LAI—leaf area index, RGR—relative growth rate, and NAR—net assimilation rate. Black circles: 7, 14, 21, 28, 35, 42, 49, and 56-days after transplanting.

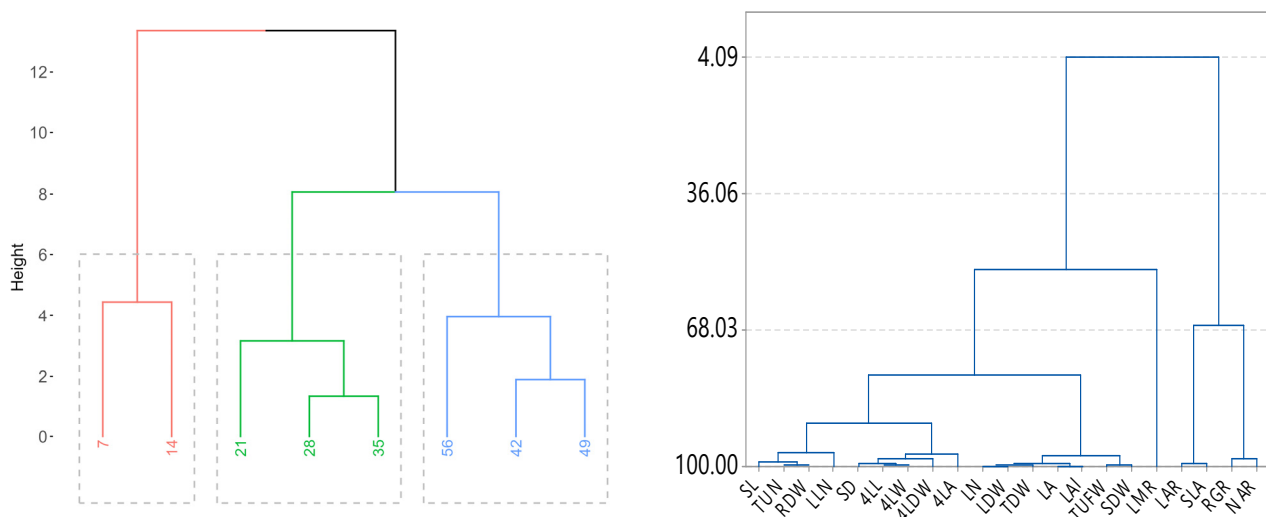


Figure 12. Dendrogram of the hierarchical clustering based on the relative values of DAT and morpho-physiological parameters of mini-tuber seed potatoes. Caption: SL—main stem length, SD—stem diameter, LN—leaf numbers, LLN—leaflet numbers, 4LL—4th leaf length, 4LW—4th leaf width, 4LA—4th leaf area, LA—leaf area, TUN—mini-tuber numbers, TUFW—mini-tuber fresh weight, RDW—root dry weight, SDW—stem dry weight, 4LDW—4th leaf dry weight, LDW—leaf dry weight, TDW—total dry weight, LMR—leaf mass ratio, LAR—leaf area ratio, SLA—specific leaf area, LAI—leaf area index, RGR—relative growth rate, and NAR—net assimilation rate. Left-side figure, X axis: 7, 14, 21, 28, 35, 56, 42, and 49-days after transplanting.

From the dendrogram, it is possible to verify the formation of three clusters when the days after transplantation (DAT) are prolonged, the first one including the first two weeks and the other two groups consisting of three weeks. If the parameters are observed, it is possible to deduce them in a more restricted group, since the objective is to be able to better explain the variations obtained in the characterization over the days after planting (Figure 12). The physiological variables (RGR and NAR), although correlated (Figure 10), were not deemed important in the cultivar characterization because they showed little variation, as seen in the PCA (Figure 11).

4. Discussion

4.1. Morphological Aspects

The results of our study are consistent with previous research on hydroponically grown potatoes, conducted by Mobini et al. [38]. Mobini's study analyzed two types of potato varieties, Agria and Sante, and their ability to produce mini-tubers under four different levels of aeration. In the control group, where there was no aeration in the nutrient solution, the length of the mini-tubers reached 92.1 cm and 74.1 cm for the two varieties, respectively. In a study by Ritter et al. [39], two production methods, a hydroponic system in greenhouse beds and an aeroponic system using the Nagore cultivar, were evaluated. The results of that study showed that the plants in the hydroponic system reached final heights between 90 and 110 cm in the greenhouse bed system. Additionally, the authors reported that the aeroponic system produced plants that grew up to 150 and 180 cm in height. In our study, we observed that several variables can affect the height of potato plants under aeroponics, as seen through their correlations (Figure 9). The leaf area ratio (LAR) and specific leaf area (SLA), despite having a low correlation, were significantly different ($p < 0.05$) and directly affected the growth of plants, in addition to the intrinsic characteristics of the cultivars and the processes of photosynthesis and respiration [29,40]. In addition, short days (long nights) can lead to early tuber development in potato plants by limiting the amount of light plants receive [41]. This, in turn, stimulates stem elongation in potato plants and affects gibberellin levels [42].

Singh et al. [43] evaluated the additional interactive effects of UV-B and mineral nutrients (NPK) on potato (var. Kufri Badashah) plant development in a dry tropical climate under field conditions in Varanasi, Uttar Pradesh, India. They found an average leaf number of 185.7 leaves per plant. On the other hand, Sumarni et al. [44] conducted a study in Purwokerto, Indonesia to determine the effect of electrical conductivity (EC) on the growth and yield of potato seeds grown in aeroponics with root-zone cooling. They reported a maximum plant height of 41.3 cm and 84.8 leaves with an EC of 1.0 mS cm^{-1} (for the first three weeks) and 2.0 mS cm^{-1} (for the rest of the study) under greenhouse conditions.

Rapid leaf-area development is advantageous because it facilitates the early capture of solar radiation and increases photosynthetic activity, leading to improved final mini-tuber production [45]. Despite being interesting as evaluation parameters, leaf characteristics such as leaf mass ratio (LMR) show little correlation, or even negative correlation (leaf area ratio and specific leaf area), with parameters of interest such as mini-tuber number (TUN).

According to Wohleb et al. [45], leaves need to develop quickly after emergence to establish their leaf area, maximize their solar radiation absorption and dry matter, and achieve the highest production of assimilates that will be translated into mini-tuber biomass. Leaf area values decrease due to leaf senescence, reducing the plant's production of photosynthates. Plant biomass is produced by net photosynthetic activity, which influences vegetative and reproductive growth. The photosynthetic efficiency of a plant depends on the efficiency of the leaves in intercepting available sunlight (plant architecture) by CO_2 uptake and fixation, and temperature, among other factors [46]. These traits are dictated by plant growth factors, genotype, and environmental conditions during growth [35].

In this study, it was possible to confirm previous observations [35,45], in which the correlations between the plant aerial part variables (length of the main stem, stem diameter, number of leaves, number of leaflets, and leaf area) and the number of mini-tubers were positively and significantly correlated ($p < 0.05$). This illustrates that good development of plant shoots results in good production of mini-tubers.

The highest accumulation of leaf dry weight occurred at 14, 21, and 28 DAT, accounting for 61%, 60%, and 59% of the total dry weight, respectively. At 63 DAT, leaf dry weight accounted for 48% of the total dry weight (Figure 5D). In the early stages of potato growth, leaves and roots are the main sinks for photoassimilates. In contrast, in the final stages of the plant's growth cycle, tuber production becomes the main sink, using resources from the leaves [47].

Nonlinear models have been used to describe processes that evaluate plant growth. In addition, nonlinear models best represent real-world observations in a study that evaluated nonlinear regression models to describe dry weight accumulation in potato plants over time [48]. Models were selected based on three criteria: (1) biological logic, (2) significance of the regression coefficients ($p < 0.10$), and (3) significance of the coefficient of determination ($R^2 > 0.75$). The construction of these models was based on previous research that thoroughly analyzed potato crops' growth and development patterns [49–51].

Leaf mass ratio (LMR) is a morphological parameter that measures the amount of a plant's biomass allocated to leaves as a percentage of its total biomass. This ratio helps to understand a plant's resource allocation strategy, particularly concerning the allocation of resources to photosynthetic tissues [52]. The LMR was stable between 14 and 42 DAT and then decreased as more resources were allocated to tuber production. This LMR reduction was probably responsible for the low contribution of variation observed in the PCA (Figure 11), in which the parameter contributes little to explain the results obtained. Ascione et al. [53] found that LMR values decreased over the plant life cycle in the field with nine potato cultivars in southern Italy and that the length of the plant life cycle affected the maximum LMR values, with the long-cycle cv. Casanova having the highest values (0.75 g g^{-1}) and the short-cycle cv. Rz-91-450 showing the lowest values (0.45 g g^{-1}).

While previous research suggested that the decrease in LAR was due to the allocation of assimilates to developing tubers [54], LAI varied with leaf shape and distribution in

the canopy. An ideal LAI promotes rapid dry weight accumulation by allowing better interception of photosynthetically active radiation [55,56]. This supports our research results, which show that the values found in LAI were positively related to all dry matter parameters (Figure 9), including stem dry weight (SDW), fourth-leaf dry weight (4LDW), leaf dry weight (LDW), and root dry weight (RDW). Total dry weight (TDW) significantly correlated with LAI (0.9).

The results of our study showed that LAI followed a similar trend to plant biomass. The highest LAI occurred when plants accumulated more biomass capable of intercepting photosynthetically active radiation (PAR) [57,58]. In a study by Wang et al. [59], researchers investigated the effects of calcium nitrate and gibberellic acid on two cultivars, Favorita and Mira. The researchers discovered that the increased production of tubers resulted from the gibberellic acid treatment. These characteristics included increased plant height, increased stem strength, increased leaf area index (LAI), and increased stolon branching. As a result, plant expansion led to increased photosynthate production, which, in turn, led to increased tuber production. LAI showed a progressive increase from 28 to 84 days after transplanting, except for some treatments of the cultivar Favorita.

4.2. Physiological Aspects

Changes in RGR, according to Singh et al. [43], can be attributed to differences in leaf photosynthetic efficiency (net assimilation rate, NAR) or reduced exposure of photosynthetically active tissues to PAR, often due to shading (LAR). The correlation values obtained support this statement, as the specific leaf area (SLA, -0.49) and leaf area index (LAI, -0.42) correlation values showed an antagonistic relation to RGR.

The highest values of net assimilation rate (NAR) were obtained at the beginning of the growth of potato plants under this modified aeroponic system when the young leaves had no competitive shading and the specific leaf area (SLA) was high. At the end of the growing cycle, the photosynthetic capacity of potato plants began to decrease, with an increase in leaf area index (LAI) and, consequently, lower NAR values. The RGR and NAR values of this study confirm the work of Tekalign and Hammes [60] on four cultivars (CIP-388453-3(A), CIP-388453-3(B), AI-624, and AI-436) at the research farm of Alemaya University, Ethiopia. Due to the continuous accumulation of dry weight, both RGR and NAR values decreased as the plants approached maturity, indicating that their newly produced biomass was a smaller proportion of the total plant mass. In Botucatu, São Paulo State, Brazil, Ferreira et al. [61] found the same pattern, a sigmoidal path, in the potato crop cycle of the Mondial cultivar grown in 17-L (4.5 gallons) pots filled with dirt from a structured red Nitosol soil under greenhouse conditions.

The amount of new plant material produced relative to the total plant biomass over a given time interval is one of the most appropriate measures to evaluate plant growth [62]. The net assimilation rate (NAR) is most strongly correlated with the relative growth rate (RGR), according to Shipley [63], which corroborates the data obtained for the correlation between NAR and RGR in this study (0.95). Accumulated biomass is directly related to the net assimilation rate (NAR) and relative growth rate (RGR). At 56 days after transplanting (DAT), lower NAR and RGR were observed. These values decline as plants approach maturity, partly due to the continuous accumulation of dry matter, which may be related to low biomass production efficiency (Figure 8). Throughout the vegetative and reproductive cycles, the net assimilation rate (NAR) decreased linearly. The average weekly decrease was $-0.00002 \text{ g cm}^{-2}$. NAR reached a minimum of $0.000388 \text{ g cm}^{-2} \text{ week}^{-1}$ at 56 DAT (Figure 8); the actual minimum estimate value was calculated from the linear regression equation. The net assimilation rate (NAR) is a physiological parameter related to the photosynthetic efficiency of leaves [64,65].

4.3. Yield

The initial growth stage of potato plants, which occurred between 7 and 34 DAT, determines the final production potential of seed potato mini-tubers. Several factors can

affect the final production of seed potatoes, such as the nutritional status of the plant, pathogens, photoperiod (hours of daylight), temperature, quality of light reaching the leaves, phytochrome levels, gibberellic acid levels, and factors inherent to the genetic potential of each cultivar [45,66,67]. Therefore, it is possible to link morphological and physiological parameters that will best predict the production of seed-potato mini-tubers at this early stage of the crop. In the last third of the potato plant cycle, the accumulated nutrients in plant shoots are transferred to produce stolons and mini-tubers, a process known as the source-sink effect. While the photosynthetic machinery is still functioning as a source, there is already a strong sink-effect exerted by mini-tuber development [45,68]. In this case, it is essential to consider the limits of each cultivar's genetic capacity and the biotic and abiotic factors mentioned above.

4.4. Multivariate Analyses

Tabachnick and Fidell [69] defined principal component analysis (PCA) as a statistical technique used to identify coherent subsets of variables in a data set that are independent of each other. Hence, PCA may condense the multitude of observed variables by capturing the underlying patterns of correlations, resulting in a reduced set of variables. Additionally, PCA produces several linear combinations of tested variables, where each linear combination represents a factor.

A study with organic potatoes evaluated six plant variables and 13 soil variables, for a total of 19 variables, for evaluating crop cycle rotations (4-years) in a field experiment in Nova Scotia, Canada. They used principal component analysis to evaluate the characteristics studied. The PCA results allowed them to choose two variables to characterize the cropping systems: plant nutrient uptake and total tuber weight. Other variables were discarded because they were not strongly correlated with soil variables. Further, the authors concluded that PCA was effective in correlating complex cropping systems with high site heterogeneity [70].

5. Conclusions

This is the first time that several morphological and physiological parameters have been evaluated to optimize seed potato characterization under a modified aeroponic system. Mini-tubers of 'Agata' potato plants were produced as early as 18 days after transplanting (DAT). However, our results suggest that the optimal harvest time to estimate tuber productivity for 'Agata' was at 45 DAT, when the number of tubers reached a plateau.

Based on the multivariate analyses of the 21 parameters evaluated in this work, we concluded that the best morphological parameters to characterize potato genotypes under our aeroponic system were main stem diameter, leaf number, fourth-leaf length, leaf area, mini-tuber numbers, mini-tuber fresh weight, root dry weight, and total dry weight. The best physiological parameter was the net assimilation rate.

To optimize future experiments characterizing seed-potato cultivars under aeroponic systems, we suggest that plants be evaluated every 14 days, which will provide information as reliable as obtained through weekly assessments in this study. The parameters that gave the least details on growth variation were 4th-leaf morphological parameters, leaf mass ratio, leaf area ratio, specific leaf area, and leaf area index. These parameters can be omitted from future experiments without harm to genotype characterization. The best parameters to predict seed potato mini-tuber yield were main-stem length, main-stem diameter, and root dry weight. Future evaluations to predict mini-tuber yield could focus on non-destructive measures like stem length and diameter. We encourage other researchers to further test these optimized parameters and harvest times in other cultivars to validate the strength and applicability of our findings with the cv. Agata. The optimized morpho-physiological parameters, selected through a multivariate statistical analysis of 'Agata,' can be applied to aeroponic methods in general as a valuable optimized method for streamlining the characterization of other seed-potato cultivars of interest to certified producers.

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Data Availability Statement: The new data created here were for the sole purpose of testing our treatments and hypotheses and are not to be entered into any public database or archives. Thus, data sharing does not apply to this article.

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References

- Otaú, V. *Manual on Quality Seed Potato Production Using Aeroponics*; International Potato Center (CIP): Lima, Perú, 2010; pp. 1–42.
- Tunio, M.H.; Gao, J.; Shaikh, S.A.; Lakhiar, I.A.; Qureshi, W.A.; Solangi, K.A.; Chandio, F.A. Potato production in aeroponics: An emerging food growing system in sustainable agriculture for food security. *Chil. J. Agric. Res.* **2020**, *80*, 118–132. [\[CrossRef\]](#)
- Lakhiar, I.A.; Gao, J.; Syed, T.N.; Chandio, F.A.; Buttar, N.A. Modern plant cultivation technologies in agriculture under controlled environment: A review on aeroponics. *J. Plant Interact.* **2018**, *13*, 338–352. [\[CrossRef\]](#)
- Garzón, J.; Montes, L.; Garzón, J.; Lampropoulos, G. Systematic review of technology in aeroponics: Introducing the technology adoption and integration in sustainable agriculture model. *Agronomy* **2023**, *13*, 2517. [\[CrossRef\]](#)
- Edmonds, J.W.; Sackett, J.D.; Lomprey, H.; Hudson, H.L.; Moser, D.P. The aeroponic rhizosphere microbiome: Community dynamics in early succession suggest strong selectional forces. *Antonie Van Leeuwenhoek* **2019**, *113*, 83–99. [\[CrossRef\]](#) [\[PubMed\]](#)
- Otaú, V.C. Producción de semilla de papa de calidad por aeroponía. In *Alternativas al Uso de Bromuro de Metilo en la Producción de Semilla de Papa de Calidad*; Centro Internacional de la Papa: Lima, Peru, 2007.
- Silva Filho, J.B.; Fontes, P.C.R.; Cecon, P.R.; Ferreira, J.F.S.; McGiffen, M.E., Jr.; Montgomery, J.F. Yield of potato minitubers under aeroponics, optimized for nozzle type and spray direction. *HortScience* **2020**, *55*, 14–22. [\[CrossRef\]](#)
- Lemoine, R.; Camera, S.L.; Atanassova, R.; Dédaldéchamp, F.; Allario, T.; Pourtau, N.; Bonnemain, J.-L.; Laloi, M.; Coutos-Thévenot, P.; Maurousset, L.; et al. Source-to-sink transport of sugar and regulation by environmental factors. *Front. Plant Sci.* **2013**, *4*, 272. [\[CrossRef\]](#) [\[PubMed\]](#)
- Chen, H.; Hu, Y.; Li, P.; Feng, X.; Jiang, M.; Sui, Z. Single-cell transcriptome sequencing revealing the difference in photosynthesis and carbohydrate metabolism between epidermal cells and non-epidermal cells of *Gracilaria lemaneiformis* (Rhodophyta). *Front. Plant Sci.* **2022**, *13*, 968158. [\[CrossRef\]](#) [\[PubMed\]](#)
- Evans, J.R. Improving photosynthesis—A review. *Plant Physiol.* **2013**, *162*, 1780–1793. [\[CrossRef\]](#)
- Buckseth, T.; Sharma, A.K.; Pandey, K.K.; Singh, B.P.; Muthuraj, R. Methods of pre-basic seed potato production with special reference to aeroponics—A review. *Sci. Hortic.* **2016**, *204*, 79–87. [\[CrossRef\]](#)
- Bročić, Z.; Momčilović, I.; Poštić, D.; Oljača, J.; Veljković, B. Production of high-quality seed potato by aeroponics. In *The Potato Crop: Management, Production, and Food Security*; Villa, P.M., Ed.; Nova Science Publishers: New York, NY, USA, 2021; pp. 25–59.
- Slattery, R.A.; Ort, D.R. Photosynthetic energy conversion efficiency: Setting a baseline for gauging future improvements in important food and biofuel crops—A review. *Plant Physiol.* **2015**, *168*, 383–392. [\[CrossRef\]](#)
- Farran, I.; Mingo-Castel, A.M. Potato minituber production using aeroponics: Effect of plant density and harvesting intervals. *Am. J. Potato Res.* **2006**, *83*, 47–53. [\[CrossRef\]](#)
- Arora, K.; Kaur, P.; Kumar, P.; Singh, A.; Patel, S.K.S.; Li, X.; Yang, Y.H.; Bhatia, S.K.; Kulshrestha, S. Valorization of wastewater resources into biofuel and value-added products using microalgal system. *Front. Energy Res.* **2021**, *9*, 646571. [\[CrossRef\]](#)
- Çalışkan, M.E.; Yavuz, C.; Yağız, A.K.; Demirel, U.; Çalışkan, S. Comparison of aeroponics and conventional potato mini tuber production systems at different plant densities. *Potato Res.* **2021**, *64*, 41–53. [\[CrossRef\]](#)

17. Anderson, M.S.; Barta, D.; Douglas, G.; Motil, B.; Massa, G.; Fritsche, R.; Quincy, C.; Romeyn, M.; Hanford, A. Key gaps for enabling plant growth in future missions. In Proceedings of the AIAA SPACE and Astronautics Forum and Exposition, Orlando, FL, USA, 12–14 September 2017. [CrossRef]
18. Golenberg, E.M.; West, N.W. Hormonal interactions and gene regulation can link monoecy and environmental plasticity to the evolution of dioecy in plants. *Am. J. Bot.* **2013**, *100*, 1022–1037. [CrossRef] [PubMed]
19. Aphalo, P.J.; Sadras, V.O. Explaining pre-emptive acclimation by linking information to plant phenotype. *J. Exp. Bot.* **2022**, *73*, 5213–5234. [CrossRef]
20. Oliveira, J.S.; Brown, H.E.; Moot, D.J. Assessing potato canopy growth and development at the individual leaf level to improve the understanding of the plant source–sink relations. *New Zealand J. Crop Hortic. Sci.* **2021**, *49*, 325–346. [CrossRef]
21. Zhang, Z.; Wang, Q.C.; Spetz, C.; Blystad, D.R. In vitro therapies for virus elimination of potato-valuable germplasm in Norway. *Sci. Hortic.* **2019**, *249*, 7–14. [CrossRef]
22. Bettoni, J.C.; Mathew, L.; Pathirana, R.; Wiedow, C.; Hunter, D.A.; McLachlan, A.; Khan, S.; Tang, J.; Nadarajan, J. Eradication of Potato Virus S, Potato Virus A, and Potato Virus M from infected in vitro-grown potato shoots using in vitro therapies. *Front. Plant Sci.* **2022**, *13*, 878733. [CrossRef]
23. Silva Filho, J.B.; Fontes, P.C.R.; Ferreira, J.F.; Cecon, P.R.; Crutchfield, E. Optimal nutrient solution and dose for the yield of nuclear seed potatoes under aeroponics. *Agronomy* **2022**, *12*, 2820. [CrossRef]
24. Wheeler, R.M.; Steffen, K.L.; Tibbitts, T.W.; Palta, J.P. Utilization of potatoes for life support systems II. The effects of temperature under 24-h and 12-h photoperiods. *Am. Potato J.* **1986**, *63*, 639–647. [CrossRef] [PubMed]
25. Wheeler, R.M.; Tibbitts, T.W.; Fitzpatrick, A.H. Potato growth in response to relative humidity. *HortScience* **1989**, *24*, 482–484. [CrossRef]
26. OSCS, Oregon Seed Certification Service. 2023 Oregon Potato Seed Certification Standards; Oregon State University: Corvallis, OR, USA, 2023; pp. 1–36. Available online: https://seedcert.oregonstate.edu/sites/seedcert.oregonstate.edu/files/2023-05/potato_standards23.pdf (accessed on 7 February 2024).
27. Badoni, A.; Chauhan, J.S. In vitro sterilization protocol for micropropagation of *Solanum tuberosum* cv. ‘Kufri Himalini’. *Academ. Arena* **2010**, *2*, 24–27. [CrossRef]
28. Kirkby, E.A.; Knight, A.H. Influence of the level of nitrate nutrition on ion uptake and assimilation, organic acid accumulation, and cation-anion balance in whole tomato plants. *Plant Physiol.* **1977**, *60*, 349–353. [CrossRef]
29. Silva Filho, J.B.; Fontes, P.C.R.; Cecon, P.R.; McGiffen, M.E., Jr. Evaluation of “UFV aeroponic system” to produce basic potato seed minitubers. *Am. J. Potato Res.* **2018**, *95*, 443–450. [CrossRef]
30. Lambers, H.; Chapin, F.S.; Pons, T.L. *Plant Physiological Ecology*, 2nd ed.; Springer Science Business Media, LLC: New York, NY, USA, 2008; pp. 321–374. [CrossRef]
31. Hunt, R. *Basic Growth Analysis: Plant Growth Analysis for Beginners*; Unwin Hyman Ltd.: London, UK, 1990; pp. 35–54. [CrossRef]
32. Radford, P.J. Growth analysis formulae—Their use and abuse. *Crop Sci.* **1967**, *7*, 171–175. [CrossRef]
33. Evans, G.C. *The Quantitative Analysis of Plant Growth*; University of California Press: Berkeley, CA, USA, 1972.
34. Li, L.; Qin, Y.; Liu, Y.; Hu, Y.; Fan, M. Leaf positions of potato suitable for determination of nitrogen content with a SPAD meter. *Plant Prod. Sci.* **2012**, *15*, 317–322. [CrossRef]
35. Gardner, F.P.; Pearce, R.B.; Mitchell, R.L. *Physiology of Crop Plants*, 2nd ed.; Scientific Publishers: Jodhpur, India, 2019; pp. 187–208.
36. R Core Team. *R: A Language and Environment for Statistical Computing*; R Foundation for Statistical Computing: Vienna, Austria, 2023; Available online: <https://www.R-project.org> (accessed on 7 February 2024).
37. Ribeiro Junior, J.I.; Melo, A.L.P. *Guia Prático Para Utilização Do SAEG*; Folha Artes Graficas Ltd.a.: Viçosa, Brazil, 2008.
38. Mobini, S.H.; Ismail, M.R.; Arouiee, H. The impact of aeration on potato (*Solanum tuberosum* L.) minituber production under soilless conditions. *Afr. J. Biotechnol.* **2015**, *14*, 910–921.
39. Ritter, E.; Angulo, B.; Riga, P.; Herrán, C.; Relloso, J.; San Jose, M. Comparison of hydroponic and aeroponic cultivation systems for the production of potato minitubers. *Potato Res.* **2001**, *44*, 127–135. [CrossRef]
40. Taiz, L.; Zeiger, E.; Møller, I.M.; Murphy, A. *Plant Physiology and Development*, 6th ed.; Sinauer Associates Inc.: Sunderland, MA, USA, 2015; p. 761.
41. Malladi, A.; Burns, J.K. Communication by plant growth regulators in roots and shoots of horticultural crops. *HortScience* **2007**, *42*, 1113–1117. [CrossRef]
42. Poštić, D.; Momirović, N.; Bročić, Z.; Dolijanović, Z.; Aleksić, G. The evaluation of biological viability of potato seed tubers grown at different altitudes. *Afr. J. Agric. Res.* **2012**, *7*, 3073–3080. [CrossRef]
43. Singh, S.; Kumari, R.; Agrawal, M.; Agrawal, S.B. Growth, yield and tuber quality of *Solanum tuberosum* L. under supplemental ultraviolet-B radiation at different NPK levels. *Plant Biol.* **2011**, *13*, 508–516. [CrossRef]
44. Sumarni, E.; Farid, N.; Soesanto, L. Effect of electrical conductivity (EC) in the nutrition solution on aeroponic potato seed production with root zone cooling application in tropical lowland, Indonesia. *Agric. Eng. Int. CIGR J.* **2019**, *21*, 70–77.
45. Wohleb, C.H.; Knowles, N.R.; Pavek, M.J. Plant Growth and Development. In *The Potato: Botany, Production, and Uses*, 1st ed.; Navarre, R., Pavek, M.J., Eds.; CABI: Boston, MA, USA, 2014; pp. 64–82.
46. Zhu, X.-G.; Long, S.P.; Ort, D.R. Improving photosynthetic efficiency for greater yield. *Annu. Rev. Plant Biol.* **2010**, *61*, 235–261. [CrossRef]

47. Struik, P.C. Responses of the potato plant to temperature. In *Potato Biology and Biotechnology Advances and Perspectives*, 1st ed.; Vreugdenhil, D., Bradshaw, J., Gebhardt, C., Govers, F., Mackerron, D.K.L., Taylor, M.A., Ross, H.A., Eds.; Elsevier: Oxford, UK, 2007; pp. 367–393.
48. Bojacá, C.R.; García, S.J.; Schrevers, E. Analysis of potato canopy coverage as assessed through digital imagery by nonlinear mixed effects models. *Potato Res.* **2011**, *54*, 237–252. [\[CrossRef\]](#)
49. Chang, D.C.; Cho, I.C.; Suh, J.T.; Kim, S.J.; Lee, Y.B. Growth and yield response of three aeroponically grown potato cultivars (*Solanum tuberosum* L.) to different electrical conductivities of nutrient solution. *Am. J. Potato Res.* **2011**, *88*, 450–458. [\[CrossRef\]](#)
50. Chang, D.C.; Park, C.S.; Kim, S.Y.; Lee, Y.B. Growth and tuberization of hydroponically grown potatoes. *Potato Res.* **2012**, *55*, 69–81. [\[CrossRef\]](#)
51. Roosta, H.R.; Rashidi, M.; Karimi, H.R.; Alaei, H.; Tadayyonnejhad, M. Comparison of vegetative growth and minituber yield in three potato cultivars in aeroponics and classic hydroponics with three different nutrient solutions. *J. Sci. Technol. Greenh. Cult.* **2013**, *4*, 80–87.
52. Lambers, H.; Oliveira, R.S. Growth and Allocation. In *Plant Physiological Ecology*, 3rd ed.; Lambers, H., Oliveira, R.S., Eds.; Springer: Cham, Switzerland, 2019; pp. 385–449. [\[CrossRef\]](#)
53. Ascione, S.; Ruggiero, C.; Vitale, C. Contribution of growth components on relative, plant, crop, and tuber growth rate of nine potato cultivars in Southern Italy. *Int. J. Sci.* **2013**, *2*, 1–11.
54. Poorter, H.; Remkes, C. Leaf area ratio and net assimilation rate of 24 wild species differing in relative growth rate. *Oecologia* **1990**, *83*, 553–559. [\[CrossRef\]](#) [\[PubMed\]](#)
55. Jongschaap, R.E. Run-time calibration of simulation models by integrating remote sensing estimates of leaf area index and canopy nitrogen. *Eur. J. Agron.* **2006**, *24*, 316–324. [\[CrossRef\]](#)
56. Bangemann, L.W.; Sieling, K.; Kage, H. The effect of nitrogen and late blight on crop growth, solar radiation interception and yield of two potato cultivars. *Field Crops Res.* **2014**, *155*, 56–66. [\[CrossRef\]](#)
57. Baret, F.; Guyot, G.; Major, D.J. Crop biomass evaluation using radiometric measurements. *Photogrammetria* **1989**, *43*, 241–256. [\[CrossRef\]](#)
58. Fernandes, G.S.T.; Lopes, P.M.O.; Moura, G.B.A.; Silva, M.V.; Galvencio, J.D.; Santos, A. Balance of photosynthetically active radiation by remote sensing in a seasonally dry tropical forest in Northeastern Brazil. *Rev. Bras. Geogr. Fís.* **2021**, *14*, 2486–2509. [\[CrossRef\]](#)
59. Wang, C.C.; Wang, X.Y.; Wang, K.X.; Hu, J.J.; Tang, M.X.; He, W.; Vander Zaag, P. Manipulating aeroponically grown potatoes with gibberellins and calcium nitrate. *Am. J. Potato Res.* **2018**, *95*, 351–361. [\[CrossRef\]](#)
60. Tekalign, T.; Hammes, P.S. Growth and productivity of potato as influenced by cultivar and reproductive growth: II. Growth analysis, tuber yield and quality. *Sci. Hortic.* **2005**, *105*, 29–44. [\[CrossRef\]](#)
61. Ferreira, L.C.; Scavroni, J.; Silva, J.R.V.; Cataneo, A.C.; Martins, D.; Boaro, C.S.F. Copper oxychloride fungicide and its effect on growth and oxidative stress of potato plants. *Pestic. Biochem. Physiol.* **2014**, *112*, 63–69. [\[CrossRef\]](#) [\[PubMed\]](#)
62. Hilty, J.; Muller, B.; Pantin, F.; Leuzinger, S. Plant growth: The what, the how, and the why. *New Phytol.* **2021**, *232*, 25–41. [\[CrossRef\]](#) [\[PubMed\]](#)
63. Shipley, B. Net assimilation rate, specific leaf area and leaf mass ratio: Which is most closely correlated with relative growth rate? A meta-analysis. *Funct. Ecol.* **2006**, *20*, 565–574. [\[CrossRef\]](#)
64. Tan, G.Y.; Tan, W.K. Net assimilation rate and relative nitrogen assimilation rate in relation to the dry matter production of alfalfa cultivars. *Plant Soil* **1981**, *59*, 185–192. [\[CrossRef\]](#)
65. Daminee, D.V.; Ramteke, L.K. Morpho-physiological attributes of wheat (*Triticum aestivum* L.) as influenced by plant growth regulators and fertilizer application. *Pharm. Innov. J.* **2022**, *11*, 5111–5135.
66. Kloosterman, B.; Bachem, C. Tuber Development. In *The Potato: Botany, Production, and Uses*, 1st ed.; Navarre, R., Pavek, M.J., Eds.; CABI: Boston, MA, USA, 2014; pp. 45–63.
67. Kreuze, J.F.; Souza-Dias, J.A.C.; Jeevalatha, A.; Figueira, A.R.; Valkonen, J.P.T.; Jones, R.A.C. Viral Diseases in Potato. In *The Potato Crop Its Agricultural, Nutritional and Social Contribution to Humankind*, 1st ed.; Campos, H., Ortiz, O., Eds.; Springer Nature: Cham, Switzerland, 2020; pp. 389–430. [\[CrossRef\]](#)
68. Caporale, A.G.; Paradiso, R.; Liuzzi, G.; Palladino, M.; Amitrano, C.; Arena, C.; Arouna, N.; Verrillo, M.; Cozzolino, V.; De Pascale, S.; et al. Green compost amendment improves potato plant performance on Mars regolith simulant as substrate for cultivation in space. *Plant Soil* **2023**, *486*, 217–233. [\[CrossRef\]](#)
69. Tabachnick, B.G.; Fidell, L.S. *Using Multivariate Statistics*, 7th ed.; Pearson India Education Services Pvt. Ltd.: Tamil Nadu, India, 2020; pp. 476–527.
70. Liu, K.; Hammermeister, A.M.; Patriquin, D.G.; Martin, R.C. Assessing organic potato cropping systems at the end of the first cycle of four-year rotations using principal component analysis. *Can. J. Soil Sci.* **2008**, *88*, 543–552. [\[CrossRef\]](#)

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