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Coconut fiber and enriched hydrogel as promising alternatives for the production of *Passiflora edulis* Sims seedlings

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Abstract: Successful production of yellow passion fruit (*Passiflora edulis* Sims) begins with obtaining high-quality seedlings. This study evaluated the quality of *P. edulis* seedlings using coconut fiber, vermiculite and hydrogel enriched with different concentrations (10, 20, 30, 40 and 50 g L^{-1}) of foliar fertilizer as substrates. As a control treatment, seedlings were produced in the absence of foliar fertilizer. The experiment was conducted in a greenhouse with a completely randomized design, distributed in four replicates of four plants, to evaluate 18 formulations based on soil and coarse charcoal with the addition of vermiculite, hydrogel or coconut fiber enriched with five doses of foliar fertilizer. At 70 days after the start of the research, growth, dry mass accumulation, chlorophyll content and substrate pH were evaluated. The data were subjected to analysis of variance, regression and multivariate analysis. The seedlings produced in the substrate with coconut fiber and enriched hydrogel showed greater vigor compared to vermiculite, with increases of 37% in height, 29% in leaf area, 31% in root volume and 30% in shoot dry mass. The use of coconut fiber with 10 and 20 g of foliar fertilizer formed seedlings with greater vigor. In the multivariate analysis, the seedlings produced with coconut fiber and enriched with hydrogel in the composition (G4 and G5) stood out. Therefore, we recommend using a substrate formulated with soil (40%), coarse charcoal (40%) plus coconut fiber enriched with 10 or 20 g of foliar fertilizer, or hydrogel enriched with between 20 and 50 g of foliar fertilizer.

Keywords: enriched vermiculite; leaf fertilizer; seedling vigor; substrate; yellow passion fruit
Abbreviations: DAS_days after sowing, PH_plant height, LN_leaf number, SD_stem diameter; LA_leaf area ChT_total chlorophyll content, RL_root length, RV_root volume, SDW_shoot dry mass, RDM_root dry mass, S_substrate, S-CG-H_soil, coarse charcoal and hydrogel, S-CG-V_soil, coarse charcoal and vermiculite, S-CG-FC_soil, coarse charcoal and coconut fiber, PCA_principal component analysis, PC1_principal component one, PC2_principal component two, G1_group 1, G2_group 2, G3_ group 2, G4_ group 4, G5_ group 5.

Introduction

The yellow passion fruit tree (*Passiflora edulis* Sims) is a species of the genus Passiflora that is well adapted to the edaphoclimatic conditions of Brazil, which is the leading producer and consumer of the crop worldwide, with production of 697,859 tons and average productivity of 15.3 t ha^{-1} from more of 45 thousand ha^{-1} in 2022 (IBGE, 2023). Currently, the states of Bahia and Ceará are the main producers of yellow passion fruit in Brazil, accounting for approximately 54% of national output (IBGE, 2023).
The prominent position of passion fruit in national fruit production is mainly due to the rapid crop cycle, resulting in rapid economic returns in comparison with other fruit trees (Faleiro et al., 2019; Moura et al., 2020; Cleves-Leguízamo, 2021, Santos et al., 2021). The species is predominantly cultivated in small and medium-sized orchards (Rodrigues et al., 2017, Pinheiro et al., 2022), mainly by small family farmers. However, one of the main problems faced by these producers is the difficulty of acquiring high-quality seedlings (Silva et al., 2023; Stafne et al., 2023).
Seedlings that present low quality and vigor characteristics can cause significant production losses, due to the high mortality after planting, or slow growth in the initial establishment phase in the field (Reed et al., 2022). Furthermore, seedlings of unknown origin can be infected with pests and diseases, resulting in low productivity (Farias et al., 2019; Silva et al., 2021;

Pagán, 2022; Reis et al., 2022). These problems result in an increase in production costs for the acquisition of inputs and labor (Silva et al., 2021).

The production of vigorous seedlings, formed with seeds having high physiological potential, free from pests and diseases, can optimize yields (Reis et al., 2022). The formulation of nutritionally balanced substrates also has a strong influence on the quality of the seedlings (Oliveira et al., 2020; Gabira et al., 2021; Silva et al., 2021, 2023). In this regard, the substrate aims to provide suitable conditions for the germination and initial development of the seedling (Melo et al., 2019; Ma et al., 2020). The substrate considered ideal must have slow decomposition and good water retention and aeration, to assure adequate aggregation of the root system, along with low cost and easy accessibility to farmers and nursery operators (Sierra et al., 2015; Andraus, 2017).

In the literature, several studies have evaluated the production of seedlings by species of the genus *Passiflora* using NPK (Miyake et al., 2017; Silva et al., 2021), organic substrates (Siqueira et al., 2020; Antunes et al., 2022; Silva et al., 2023) and controlled-release fertilizer (Guilherme et al., 2024). However, the results differ in relation to the formulation and the time required to obtain seedlings. Furthermore, the corresponding inputs are often expensive or difficult to acquire by small farmers.

In a study conducted by our research group, remarkable quality of *P. edulis* seedlings produced by using hydrogel enriched with leaf fertilizer in the substrate composition was observed (Silva et al., 2021). However, the hydrogel used was expensive, which limits its viability, especially for farmers who produce seedlings on a small scale. Therefore, it is important to explore variations in foliar fertilizer concentrations to establish a more precise and well-founded recommendation, in addition to evaluating the use of other inputs, such as coconut fiber and vermiculite, to replace hydrogel in the substrate formulation due to the low cost and high moisture retention capacity of these materials.

Thus, in this study we evaluated the quality of yellow passion fruit seedlings (*P. edulis*) using substrates based on coconut fiber, vermiculite and hydrogel enriched with different concentrations of leaf fertilizer for the production of yellow passion fruit seedlings for conventional production systems. Extensive analysis considering 18 substrate formulations was carried out to select and recommend the substrate with the greatest practical potential for yellow passion fruit production by small producers and nursery operators. The results of this study can provide important information to attain increased passion fruit production from high-quality seedlings.

Results and Discussion

Comparison of the three substrate groups (hydrogel, vermiculite and coconut fiber)

The biometric characteristics of the *P. edulis* seedlings showed wide variation when comparing the three substrate groups enriched with leaf fertilizer. The mixtures composed of soil and coarse charcoal with the addition of hydrogel or coconut fiber enriched with foliar fertilizer provided better performance compared to the substrate with vermiculite enriched in the formulation. There were increases in height (37%), stem diameter (15%), number of leaves (13%), leaf area (29%), root volume (31%) and shoot dry mass (30%) when using hydrogel and coconut fiber compared to vermiculite. For root length and substrate pH, there were no differences between the three groups of substrates evaluated (Figure 1A-J).

The worse performance of seedlings when using vermiculite in the substrate formulation may have been associated with the strong adsorption capacity of cations such as Ca^{2+}, Mg^{2+}, K^{+} and $NH4^{+}$ due to clay's negative surface charges and its lamellar and accordion-shaped structure, allowing molecules of varying sizes to bind to its structure (Teodoro et al., 2020), thus reducing the availability of nutrients contained in the liquid fraction of the substrate, reducing the growth of passion fruit seedlings. Several studies have recommended the use of vermiculite to improve the physical characteristics of soil, such as water retention and aeration, which can improve root development (Li et al., 2017; Wisdom et al., 2017) and vegetative production (Bebayehu et al., 2023). On the other hand, other studies have shown a reduction in nutrient content in leaves when using vermiculite (Chatzistathis et al., 2021), and low seedling vigor using vermiculite enriched with NPK (Silva et al., 2021). Therefore, its effectiveness may vary depending on specific conditions, such as the type of plant, quantity used in the substrate formulation and the growing environment.

Regression analysis

In the regression analysis carried out for the three mixtures, considering the six doses of foliar fertilizer (Figure 2A-J), the substrates formulated with soil, coarse charcoal and coconut fiber fitted the quadratic regression model in six of the ten characteristics evaluated. In substrates formulated with hydrogel, four variables fitted the quadratic model and another four the linear model (Figure 2A-J). Interestingly, the different doses of leaf fertilizer associated with vermiculite did not result in variation in the characteristics evaluated in *P. edulis* seedlings, with the exception of the chlorophyll content, which presented adjustment to the quadratic regression model with a total chlorophyll peak (51 μm g^{-1}) when using 34 g of foliar fertilizer (Figure 2E).

The cations adsorbed by vermiculite can be gradually released when conditions favorable to ion exchange occur. This release occurs when other cations in the solution compete with the cations adsorbed by the surface charges of the vermiculite (Batista et al., 2018). Thus, due to the high number of negative charges present in vermiculite, there was adsorption of the cations present in the leaf fertilizer, limiting their availability to the plants, resulting in lower growth and mass accumulation, even in treatments with a higher dose of fertilizer. These nutrients can later be made available to plants in field conditions, when the substrate adhered to the root system interacts with the chemical and physical components of the soil, which can result in a medium-term release of these nutrients.

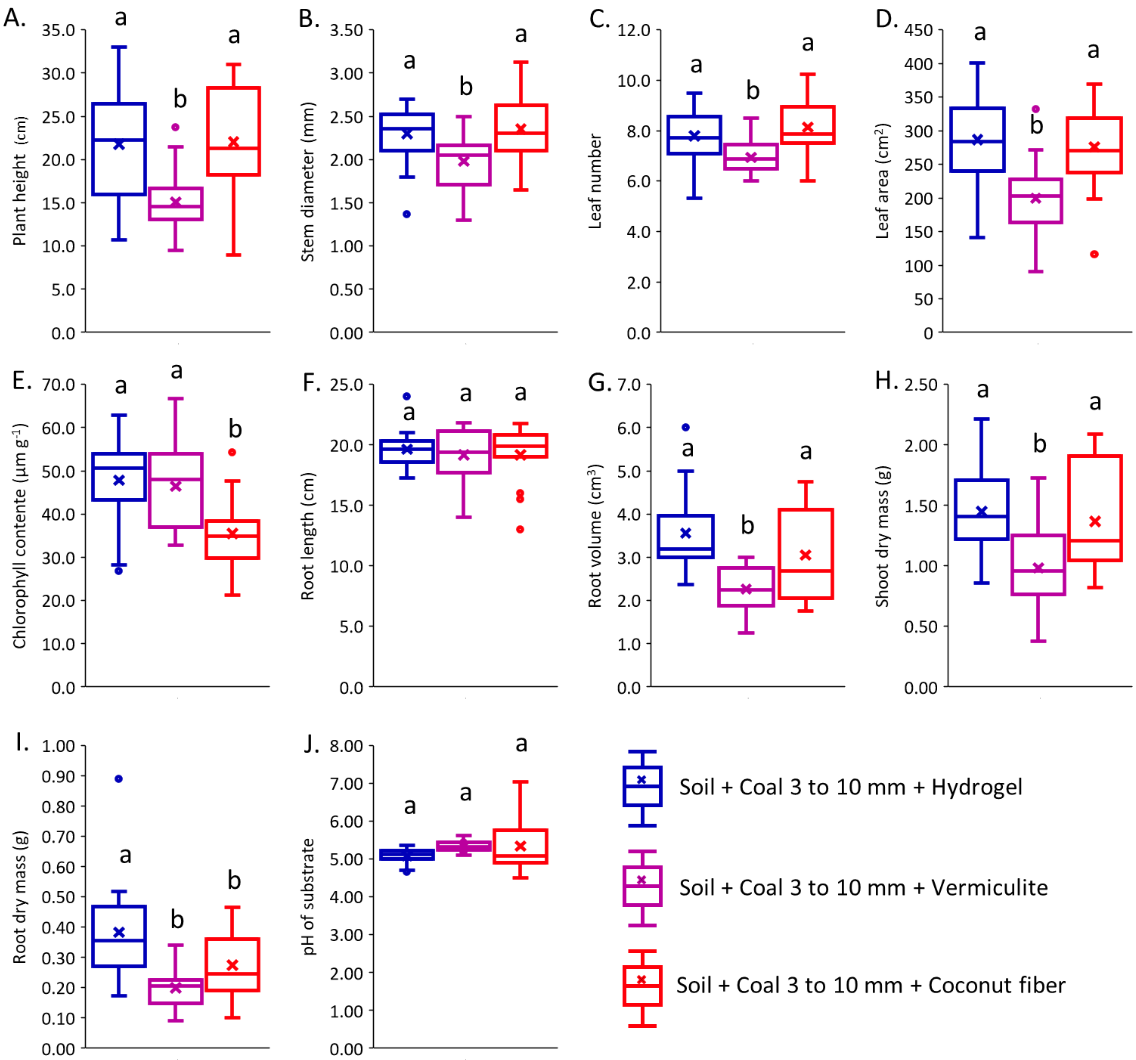

Figure 1. Boxplot of the biometric attributes of *P. edulis* seedlings produced in a substrate formulated with soil and coarse charcoal, plus 20% hydrogel, vermiculite or coconut fiber enriched with leaf fertilizer in different concentrations. A. Plant height; B. Stem diameter; C: Leaf number; D. Leaf area; E. Total chlorophyll; F. Root length; G. Root volume; H. Shoot dry; I. Root dry mass; J. Substrate pH. Lowercase letters compare the means between the three substrate groups using the Tukey test ($p \leq 0.05$).

The cation exchange capacity of vermiculite can vary depending on the cation concentration in the solution, ion exchange, solution pH, solution flow rate, and specific ionic interactions, but generally ranges from 80 to 150 meq/100 g (Ugarte et al., 2005). A study conducted by Teodoro et al. (2020) reported 122 meq/100 grams of expanded vermiculite, corresponding to 97.2% cation exchange. These results indicate a strong adsorption capacity of vermiculite, which can impact the availability of nutrients to plants, as found in our study. Further corroborating this finding, Silva et al. (2021) using vermiculite enriched as NPK observed less efficient supply of nutrients to *P. edulis* seedlings when compared to hydrogel. However, it is necessary to evaluate the performance of these plants in the field to quantify the impact on growth and yield. The regression curves indicated that the use of coconut fiber caused saturation, that is, an inflection point of the curve from 20 g of foliar fertilizer, since above this quantity reductions occurred in plant height, number of leaves, leaf area and substrate pH (Figures 2A, C, D, J). On the other hand, the enriched hydrogel negatively affected seedlings from 30 g of foliar fertilizer (Figures 2A, E, H). Reinforcing this observation, characteristics such as stem diameter, leaf area, root volume and root dry mass demonstrated increasing linear behavior when using the hydrogel enriched with leaf fertilizer (Figures 2B, D, G, I). This result indicates that the use of coconut fiber resulted in greater efficiency in the supply of nutrients in the short term and greater savings in the use of foliar fertilizer compared to hydrogel and vermiculite.

Coconut fiber has been widely used in the formation of seedlings of fruit species, in horticulture and gardening (Freire et al., 2019; Cardozo et al., 2021). This use is mainly due to its accessibility and low cost in relation to various other materials, such as hydrogels (Silva et al., 2023). Coconut fiber has the advantages of high water retention capacity and soil aeration, neutral pH, biodegradability and presence of nutrients that can stimulate plant growth. However, like all biological materials, its properties can vary depending on the source and processing, since when poorly processed it can accumulate high levels of salts (Sá et al., 2015; Silva et al., 2023).

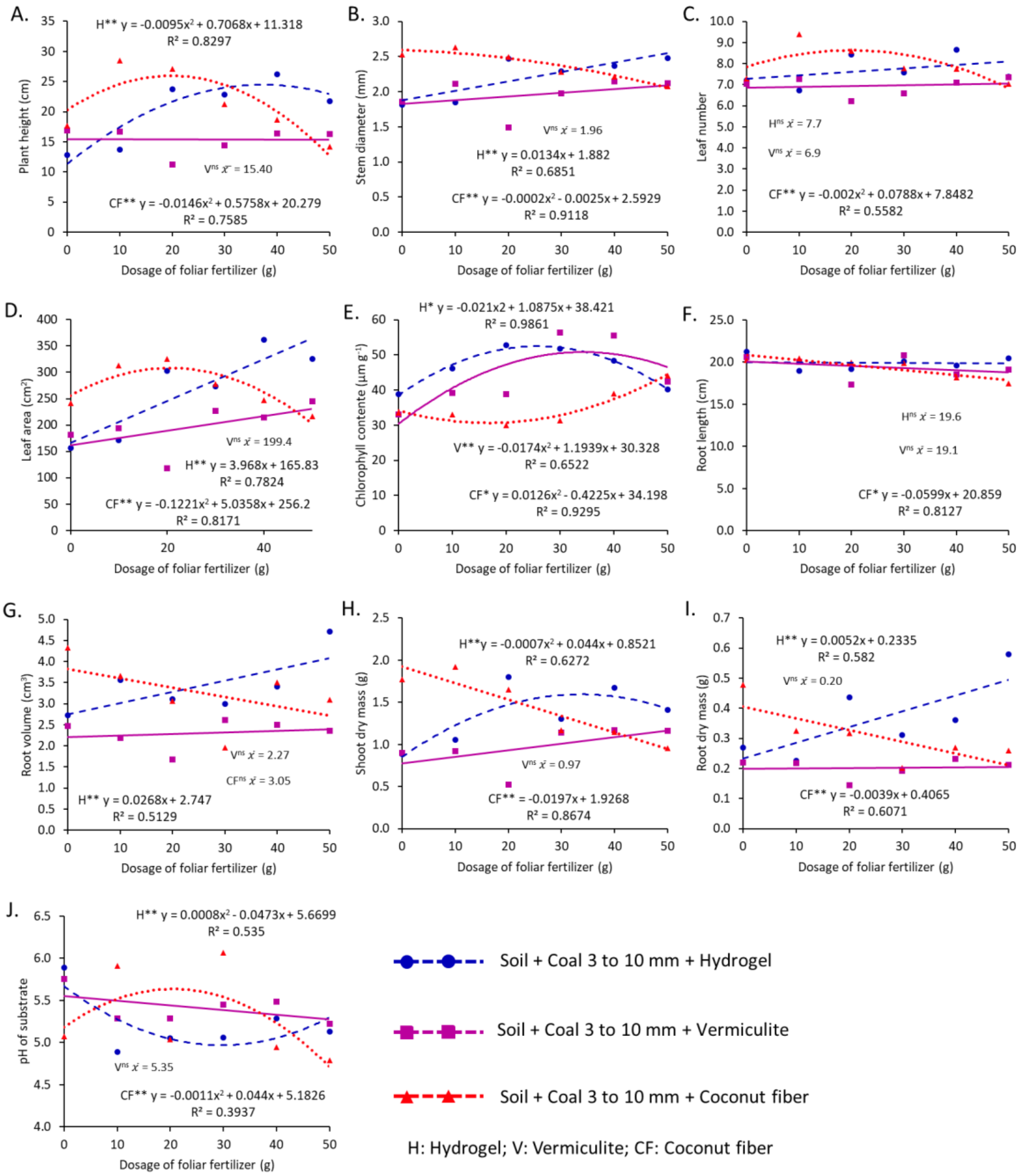

Figure 2. Regression of the biometric attributes of *P. edulis* seedlings produced in a substrate formulated with soil and coarse charcoal, plus 20% hydrogel, vermiculite or coconut fiber enriched with different concentrations of foliar fertilizer. A. Plant height; B. Stem diameter; C. Leaf number; D. Leaf area; E. Total chlorophyll; F. Root length; G. Root volume; H. Shoot dry; I. Root dry mass; J. Substrate pH. S-CG-H: soil, coarse charcoal and hydrogel; S-CG-V: soil, coarse charcoal and vermiculite; S-CG-FC: soil, coarse charcoal and coconut fiber. ns, not significant, * and ** significant at 5 and 1%, respectively, by the F-test.

Multivariate analysis

In the cluster analysis considering the 16 substrate formulations and the ten characteristics evaluated, five groups were formed. The most vigorous seedlings were obtained when produced in substrates from groups 4 and 5 (G4, G5), with G4 substrates being formed by hydrogel enriched with leaf fertilizer in doses of 20 to 50 g L^{-1} and G5 being formed by fiber coconut enriched with 10 and 20 g L^{-1} of fertilizer. The seedlings with lowest quality were those produced in the substrate with vermiculite and 20 g L^{-1} of leaf fertilizer (G2).

The same grouping trend was confirmed in the principal component analysis (PCA), with seedlings formed in substrates from groups G1 and G3 having intermediate quality. The correlation circle identified that characteristics such as root length (2.7%) and total chlorophyll content (0.38%) had low contribution to PC1 to explain the variation observed in treatments. However, characteristics such as stem diameter (16.68%), shoot dry mass (16.41) and leaf area (15.48) presented high contributions to PC1. On the other hand, when considering the sum of the first two components (PC1 and PC2), all characteristics presented a contribution above 15% (Figure 3A-C).

The seedlings formed in the G4 and G5 substrates stood out in all the evaluated characteristics, as depicted by the light and dark blue color, which indicates high vigor seedlings according to heatmap analysis. Although the use of hydrogel involved a higher cost compared to coconut fiber, acclimatization after planting can be more efficient when using this component,

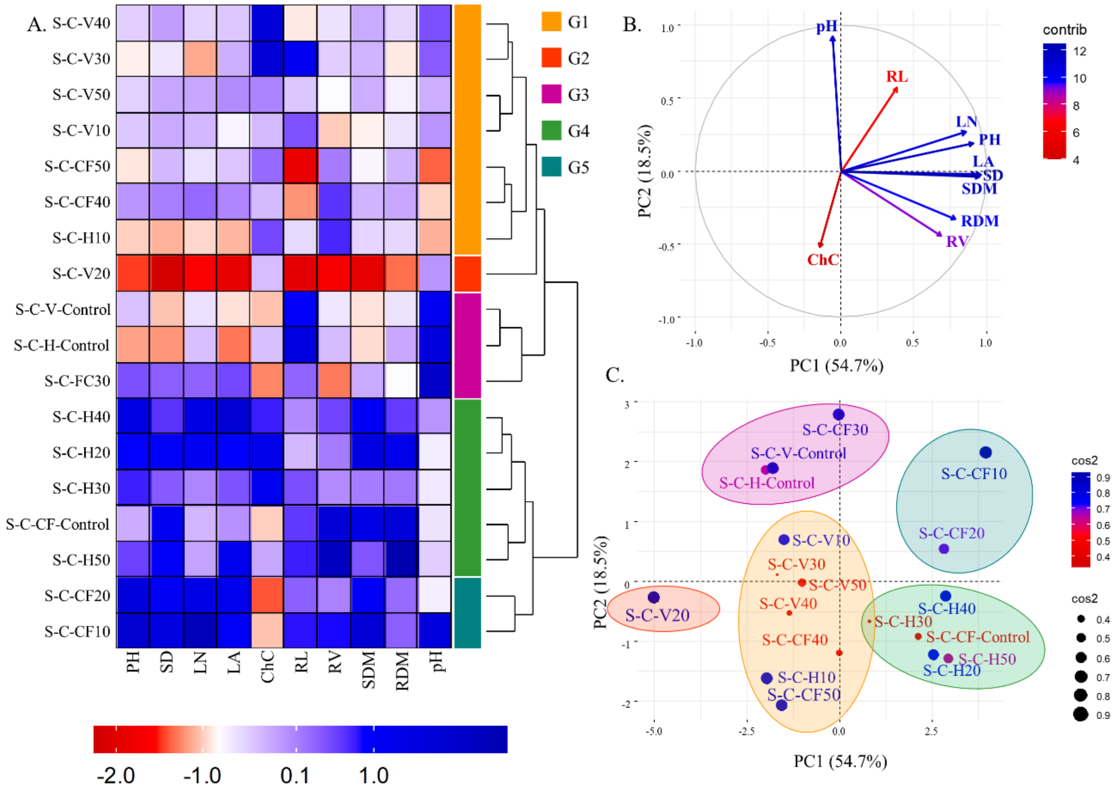

Figure 3. Multivariate analysis considering 18 substrates and 10 characteristics evaluated in *P. edulis* seedlings produced in a substrate formulated with soil and coarse charcoal, plus 20% hydrogel, vermiculite or coconut fiber, enriched with different concentrations of foliar fertilizer. A. Cluster analysis associated with the heatmap of the substrates and characteristics evaluated, with the formation of five groups (G1-G5). Each square represents the contribution of each characteristic to the performance of seedlings produced in the different substrates tested. The scale varies from red to blue, corresponding to low and high contribution, respectively. B. Circle of correlation and contribution of characteristics depending on the first two components. The arrows represent the direction of the characteristic and their colors represent the contribution – red (low) to blue (high) – of each characteristic to the two components; C. Dispersion of treatments depending on the first two components. The color and size of the circle represent the contribution of the first two components to explaining the treatment.

since the hydrogel is formed by superabsorbent polymers that have the ability to absorb and retain large amounts of water in their internal structures. When mixed with soil, it increases the water retention capacity of the substrate, reducing the amount and frequency of irrigation (Neves et al., 2021).

The results presented in the multivariate analysis allowed us to obtain a direct relationship between all substrates and the characteristics evaluated. Thus, the use of vermiculite under the conditions of this study is not recommended, which is confirmed by the red color in the heatmap and the positioning in the PCA analysis. The use of multivariate analysis has been shown to be efficient in separating groups and recommending substrates for the production of seedlings of various species (Gomes Júnior et al., 2019; Silva et al., 2021, 2023). The similarity in quality obtained in groups 4 and 5 makes it possible to use any of the substrates present in these groups, enabling farmers and nurseryman to select the most suitable substrate for their conditions.

Materials and Methods

Experiment location and plant material

The experiment was conducted under an anti-aphid screen at the experimental farm of the Embrapa Cassava and Fruits research unit (Embrapa Mandioca e Fruticultura), located in the municipality of Cruz das Almas, Bahia (12° 39' 25" S, 39° 07' 27" W, 222 m). Seeds of *Passiflora edulis* Sims (BGP418) from the Passion Fruit Germplasm Bank of Embrapa Mandioca e Fruticultura were used. The seeds were extracted from ripe fruits, with completely yellow skin, obtained from controlled pollination. They were washed in running water and dried at room temperature for one week until their use in the tests.

Experimental design and treatments

The experiment was conducted in a completely randomized design, consisting of 18 treatments (Table 1) distributed in four replicates of four plants in each plot. The different formulations were prepared in addition to those analyzed in the previous study conducted by our research group, aiming to reduce the cost of seedling production and determine the best concentration of foliar fertilizer present in the formulation (Silva et al., 2021).

Table 1. Percentage of materials used in the formulation of substrates for the production of *Passiflora edulis* Sims seedlings.

Substrate	Soil (%)	Coal 3 to 10mm (%)	Vermiculite (%)	Coconut fiber (%)	Hydrogel (%)	Leaf fertilizer (g L^{-1})
S1	40	40	0	0	20	10
S2	40	40	0	0	20	20
S3	40	40	0	0	20	30
S4	40	40	0	0	20	40
S5	40	40	0	0	20	50
S6	40	40	0	0	20	0
S7	40	40	20	0	0	10
S8	40	40	20	0	0	20
S9	40	40	20	0	0	30
S10	40	40	20	0	0	40
S11	40	40	20	0	0	50
S12	40	40	20	0	0	0
S13	40	40	0	20	0	10
S14	40	40	0	20	0	20
S15	40	40	0	20	0	30
S16	40	40	0	20	0	40
S17	40	40	0	20	0	50
S18	40	40	0	20	0	0

The substrate used as a base was formulated with 40% soil (clay texture, Alic Dystrophic Red Oxisol) and 40% coarse charcoal (3 to 10 mm particle size), while the remaining 20% was composed of hydrogel variations (Forth, Cerquilho Velho, Cerquilho-SP), vermiculite (Brasil Minérios, Santa Genoveva, Goiânia, GO) or coconut fiber (Golden Mix, Amafibra, Artur Nogueira, SP). These additional components were enriched with different concentrations of soluble foliar fertilizer for fruits (10, 20, 30, 40 and 50 g of soluble foliar fertilizer in 1 L of water), depending on the treatment (Forth®, Cerquilho Velho, Cerquilho-SP). As a control treatment, seedlings were produced in different mixtures without foliar fertilizer (Table 1).

Enriching the substrate with foliar fertilizer

The result of the mixture gave rise to three groups of substrates, which were stored and stirred for 15 days before sowing. The three groups of substrates were enriched with nutrient solution, prepared from solutions containing 10, 20, 30, 40 or 50 g of soluble foliar fertilizer for every 1.0 liter of water. The preparation was carried out using 1.0 L of each concentration of nutrient solution or water (S6) for every 5.0 g of hydrogel.

The first set of substrate (S1 to S6) was initially composed of soil and charcoal, with the enriched hydrogel being added after the storage period, with the exception of S6, used as a control treatment. The second set of substrate was composed of soil, coarse charcoal and vermiculite (S7 to S12), and the third set of substrate contained soil, coarse charcoal and coconut fiber (S13 to S18). In the first, the nutrient solution was mixed with 5.0 g of hydrogel, while in second and third the nutrient solution was incorporated into the substrate. In control treatments (S6, S12 and S18), the nutrient solution was replaced by water. The final volume of each formulation was 5.0 L.

After mixing, the substrates were distributed in polyethylene tubes measuring 12.0 x 1.4 cm (280 cm^3). Sowing using BGP418 was carried out at a depth of 1.0 cm and irrigation was performed daily.

Variables analyzed

The biometric variables evaluated 70 days after sowing (DAS) were plant height (PH) in cm, measured with a ruler; leaf number (LN); stem diameter (SD) in mm measured with a digital caliper at 3.0 cm from the plant neck; leaf area (LA) in cm^2 where the leaves were distributed on a white surface with a millimeter ruler that served as a reference to estimate the leaf area using the ImageJ® software; total chlorophyll content (ChT), determined by a chlorophyll meter (Chlorophyll Content Meter, CCM-200, OptSciences®). In addition to the aerial part characteristics, root length (cm) and root volume (RV) in cm^3 were determined. The shoot dry mass (SDM) and root dry mass (RDM) were obtained after drying in a forced-air oven at 65 °C, until they reached constant weight. Finally, the pH of the substrate after completion of the experiment was determined with a bench pH meter (pH Bench - mPA-210 - MS Tecnopon®).

Data analysis

The data from the three substrate groups considering hydrogel, vermiculite and coconut fiber enriched with foliar fertilizer, having in common the use of soil and coarse charcoal, were represented by box-plots. The means of the three groups were subjected to analysis of variance, and when significant were compared using the Tukey test ($p \leq 0.05$). Then the data on the three mixtures were subjected to regression analysis depending on the doses of foliar fertilizer, with the best model being selected according to significance using the ANAVA F-test and the coefficient of determination (R^2) using the SISVAR program (Ferreira et al., 2019).

To complement the analysis of variance, the data were subjected to joint analysis using multivariate statistics and heatmap graphs. Principal component analysis (PCA) with correlation circles and contribution of characteristics was also performed to verify the groupings of substrates depending on the characteristics evaluated, the formation of six groups (G1 to G6) was recorded after analysis. The ggfortify, nbcluster, factoextra and superheat packages of the R program (R Core Team 2022) were used.

Conclusions

To produce vigorous seedlings of sour passion fruit (*Passiflora edulis*), it is recommended to use substrates from groups 4 and 5, formulated using soil, coarse charcoal plus coconut fiber enriched with 10 or 20 g of foliar fertilizer (S3 and S14) or formulated with soil, coarse charcoal and hydrogel enriched with foliar fertilizer ranging from 20 to 50 g (S2 to S5).
The use of vermiculite enriched with foliar fertilizer is not recommended for the production of *Passiflora edulis* seedlings due to its ability to adsorb the cations provided by the foliar fertilizer and consequently reduce the release of nutrients to the seedlings.

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