# **Substrate combinations in potato planting to increase the production of minitubers**

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#### **Abstract**

The profitability and initial development of potatoes depend on the type of substrate used in planting. Therefore, this study aimed to evaluate combinations of substrates for the production of minitubers of potatoes. An experiment was carried out with the combinations of substrates based on sphagnum peat, coconut fiber, rice husks, and expanded vermiculite [TFCV]; and sphagnum peat and expanded vermiculite [TV], in different proportions (100%; 75%; 50%; 25%), with five replications. The treatments were: (i) TFCV<sub>100%</sub>; (ii) TFCV<sub>75%</sub>+TV<sub>25%</sub>; (iii) TFCV<sub>50%</sub>+TV<sub>50%</sub>; (iv) TFCV<sub>25%</sub>+TV<sub>75%</sub>; and (v) TV<sub>100%</sub>. At 40 and 70 days after planting (DAP), the development of shoots, tubers, and roots was monitored. Results showed that the isolated application of TV showed the highest production of fresh mass of tubers (93.0 g), and fresh (0.28 g) and dry mass of roots (0.17 g). At 40 DAP, tuber production was highly correlated with the root part (r: 0.77). However, after 70 DAP, tuber production was associated with shoot development (r: 0.66). If is necessary to mix substrates recommend the mixing of TFCV $_{50\%}$ + TV<sub>50%</sub>. The addition of coconut fiber and rice husk increases the availability of nutrients in the substrate with a positive effect on the aerial development of the plants.

**Keywords:** protected cultivation, seed potato, *Solanum tuberosum* L., Sphagnum peat

#### **Introduction**

In Brazil, potato (*Solanum tuberosum* L.) is one of the most important vegetables in agriculture. In 2021, potato production was approximately 116,428 hectares, with a production of 3.8 million tons and productivity of 33,099 kg ha-1. Brazilian production is distributed in the Northeast, Southeast, South, and Midwest regions. The states of Minas Gerais with São Paulo, Paraná, and Rio Grande do Sul produce more than 80% of the total national production (IBGE, 2021).

Potato is an asexually propagated vegetable crop and once infected, the seed tubers favor the spread of diseases leading to early degeneration of the crop with direct influences on the quality and productivity of tubers. Therefore, one of the permanent challenges of potato production is the quality of propagating material with satisfactory phytosanitary (Merenda, 2020). In this context, the use of seeds with high genetic and phytosanitary quality is fundamental for the commercial exploitation of potatoes. Among other reasons, the seed is also one of the highest components in the cost of production (Deleo, 2022). The main costs in potato production are defensives and fertilizers, seeds, and commercialization/processing, representing 26.0%; 13.3%; 19.4% of total costs (Hortifruti Brasil, 2022).

The use of seedlings originating from vitro culture allows the production of minitubers under protected conditions, regardless of the season or demand. Minitubers can be planted in the field, with appropriate technology and conditions to obtain seed tuber. Planting in containers containing a suitable substrate can be an alternative to the production of mini tubers, but there is a limitation in space for root expansion. The volume limitation requires that the substrate be able to maintain the availability of available water, without compromising the oxygen concentration. Studies that demonstrate

alternative substrates to reduce the cost of production and increase the seed tuber quality are necessary, and therefore, justify the performance of this study.

The substrate must offer an adequate distribution between macro and micropores and adequate nutrient conditions for the initial development of the plants. These characteristics favor the physiological activity of the roots and, consequently, the development of the plants (Silva et al., 2021). The chemical and physical properties of the substrates are necessary for the proper initial management of the plants (Gomes and Freire, 2019).

With the hypothesis that the mixture of substrates can interfere in the production of minitubers. This study tested the combinations of substrates for the production of minitubers from potato plants derived from in vitro cultivation.

# **Material and Methods**

## *Study characterization*

The experiment was carried out in a protected cultivation area of approximately 1.5 hectares, in the Cristalina region, Goiás (Latitude: 16° 46' 4'' South, Longitude: 47° 36' 47'' West), in 2019. In Brazil, the Cristalina region has an important production of potatoes with an area of approximately 4,000 hectares, concentrated in large areas with high productivity and technology.

The experimental design was established in randomized blocks with combinations of substrates based on sphagnum peat, coconut fiber, rice husks, and expanded vermiculite [TFCV]; and sphagnum peat and expanded vermiculite [TV], in different proportions (100%; 75%; 50%; 25%), with five replications. The treatments were: (i) TFCV<sub>100%</sub>; (ii) TFCV<sub>75%</sub> + TV<sub>25%</sub>; (iii) TFCV<sub>50%</sub> + TV<sub>50%</sub>; (iv) TFCV<sub>25%</sub> + TV<sub>75%</sub>; and (v) TV<sub>100%</sub>. TFCV and TV represent 100% of the concentration, respectively. Each subplot was 60 cm wide, spacing 10 cm between plants and 10 cm between rows, with 36 plants, totaling a plot area of 0.7 m2 (**Figure 1**).

Before mixing, the substrates were characterized according to the IN 17 and IN 31 regulations, in the IAC laboratory (Agronomic Institute of Campinas) to determine the chemical and physical conditions (**Table 1**). Characterized contents of macro (nitrate: NO<sub>3</sub>; ammonium: NH<sub>4</sub><sup>+</sup>; phosphorus: P; potassium: K; magnesium: Mg; calcium: Ca; sulfur: S) and micronutrients (iron: Fe; zinc: Zn; manganese: Mn; boron: B; copper: Cu), and electrical conductivity, wet and dry density, and total porosity of the substrate.

# *Crop management*

The study was carried out in greenhouses with



Figure 1. Spatial distribution of treatments and their repetitions (Replication 1, R-I; Replication 2, RII; Replication 3, RIII; Replication 4, RIV; Replication 5, RV).

a protective structure in the arch model, with width, length, and height of the lateral pillars and central span, respectively, of 7.0; 30.0 m; 3.0m; and 3.8 m. The structure consists of an upper arch covered with a 0.15 mm thick diffuser low-density polyethylene film, with the front and side parts closed with a 50-mesh anti-aphid screen.

Micropropagated seedlings (Agate variety), from 2 to 3 cm long, were sown on a surface of 1.2 m wide and 38 m long, with a mixture of substrates. The seedlings were healthy, free of viruses, and were produced by the local Tissue Culture Laboratory.

Plants were cultivated with the application of nutrient solution with concentrations of 134 mg L<sup>-1</sup> (nitrate,  $NO<sub>3</sub>$ ), 24 mg L<sup>-1</sup> (ammonium, NH $<sub>4</sub>$ <sup>+</sup>), 40 mg L<sup>-1</sup> (phosphorus),</sub> 295 mg L-1 (potassium), 162 mg L-1 (magnesium), 40 mg  $L^{-1}$  (calcium), 64 mg  $L^{-1}$  (sulfur), 1.0 mg  $L^{-1}$  (iron), 0.3 mg  $L^{-1}$  ( zinc), 1.0 mg  $L^{-1}$  (manganese), 0.13 mg  $L^{-1}$  (boron),  $0.04$  mg L<sup>-1</sup> (copper), and  $0.04$  mg L<sup>-1</sup> (molybdenum), and conductivity electricity of approximately 2.0 dS m-1. Disease and insect control was carried out preventively using fungicides and insecticides registered for the potato crop, in periodic applications.

# *Analyzed variables and statistical analysis*

After 40 and 70 days of planting, monitored the development of shoots (number and length of stems, fresh mass, and dry mass), tubers (number of tubers, fresh mass, and dry mass), and roots (fresh mass and mass dry). In each plot, four plants were used per plot to determine these variables. To quantify the dry mass, plant samples were dried in an oven with forced air circulation at the temperature of 75º C for 24 hours.

After 80 days of planting, the final harvest was also carried out, manually, collecting 20 central plants of each treatment to monitor the weight and number of tubers. Tubers were classified according to size into 3



**Table 1.** Substrates based on sphagnum peat, coconut fiber, rice husks, and expanded vermiculite [TFCV]; and sphagnum peat and expanded vermiculite [TV]

nitrate: NO<sub>3</sub>; ammonium: NH<sub>4</sub>+; phosphorus: P; potassium: K; magnesium: Mg; calcium: Ca; sulfur: S; iron: Fe; zinc: Zn; manganese: Mn; boron: B; copper: Cu; electrical conductivity: Ec; wet density: DsW; dry density: dsD; Total porosity: TP.

types: Type I (30 – 40 mm), Type II (23 – 30 mm), and Type  $III$  (< 23 mm).

The results were submitted to statistical analysis, using the R statistical program. The data were submitted to analysis of variance (ANOVA), when the F test was significant, the averages were compared using Fisher's test (P < 0.05). Correlations between variables were performed using Pearson's test (P < 0.05).

#### **Results and Discussion**

### *Production of shoots, tubers, and roots*

In shoot development, the combinations of substrates with TFCV and TV did not influence the length and number of stems, and fresh and dry mass of shoots with respective means of 77.9 and 2.4 cm; 82.5 and 4.5 g in the 40<sup>th</sup> day after the transplant (Table 2).

Favorreto (2005) and Léon (2007) showed that the dry mass of the Atlantic cultivar can vary between

5.11 and 8.65 g in the  $49<sup>th</sup>$  day after planting. In this study, the dry mass varied between 3.9 and 5.0 g in the  $40<sup>th</sup>$ day, close to the values found by Favorreto (2005) and Léon (2007). Possibly, all substrates provided adequate conditions for the development of the shoot and root system, guaranteeing the supply of water and nutrients necessary for the development of the seedlings.

 Fresh mass of tubers, and fresh and dry mass of roots were influenced by substrate combinations in the 40<sup>th</sup> day after the transplant. The isolated application of TV showed the highest fresh mass of tubers (93.0 g), and fresh (0.28 g) and dry mass of roots (0.17 g), considering production of 34; 35; and 24%, greater than the  $TFCV_{75\%}$ +TV<sub>25%</sub> that presented the lowest performance (Table 2). Possibly, the positive effect on the root system and tuber development with planting in the TV must be associated with the better physical conditions of the substrate, which presented a wet and dry density,

**Table 2.** Number of stems [NS], length of stems [LS], number of tubers [NT], and fresh [FM] and dry [DM] mass of shoot, tubers, and roots of potatoes in substrates based on sphagnum peat, coconut fiber, and rice husk [TFC], and sphagnum peat and expanded vermiculite [TV] in different proportions (25%; 50%; 75%; and 100%) in the 40<sup>th</sup> and 70<sup>th</sup> day after the transplant

	------------- Shoot parts--------			-------- Tubers --------			--------Roots --------		
	NS	$LS$ (cm)	FM(g)	DM(g)	NT	FM(g)	DM(g)	FM(g)	DM(g)
				40 <sup>th</sup> day after the transplant					
<b>TFCV</b>	2.2	75.5	77.3	3.9	7.8	81.1 ab	11.5	0.22 <sub>b</sub>	0.14 <sub>b</sub>
TV	2.6	80.6	94.0	5.0	6.4	93.0 <sub>a</sub>	13.1	0.28 <sub>a</sub>	0.17 <sub>a</sub>
$TFCV_{75\%}$ + $TV_{25\%}$	2.4	77.9	73.9	3.9	8.5	60.7 c	8.9	0.18 <sub>b</sub>	0.13 <sub>b</sub>
$TFCV_{50\%} + TV_{50\%}$	2.4	75.6	80.7	4.7	7.2	76.6 abc	10.7	0.22 <sub>b</sub>	0.14 <sub>b</sub>
$TFCV_{25\%} + TV_{75\%}$	2.5	80.0	86.6	4.9	6.7	62.8 bc	10.7	0.22 <sub>b</sub>	0.15 <sub>b</sub>
P (ANOVA)									
$P_{value}$	0.56	0.70	0.18	0.15	0.28	< 0.05	0.15	< 0.05	< 0.05
<b>CV</b>	15.98	9.15	16.55	18.94	21.7	24,2	22.2	16.60	15.37
				70 <sup>th</sup> day after the transplant					
<b>TFCV</b>	2.1	79.7	38.4 C	4.5 <sub>c</sub>	7.8	177.1B	24.6 B	0.14	0.13
TV	2.0	82.3	53.1 bc	$6.0$ ab	6.9	200.7AB	26.1 AB	0.19	0.16
$TFCV_{75\%}$ + $TV_{25\%}$	2.4	85.9	63.8 <sub>q</sub>	6.4 <sub>q</sub>	8.9	231.5A	30.8 A	0.18	0.16
$TFCV_{50\%} + TV_{50\%}$	2.2	83.0	47.1 bc	4.8 bc	8.2	168.1B	24.2 B	0.17	0.15
$TFCV_{25\%} + TV_{75\%}$	1.9	88.1	55.5 ab	5.1 abc	8.3	175.3B	23.8 B	0.16	0.15
				P (ANOVA)					
$P_{value}$	0.68	0.29	< 0.05	< 0.05	0.14	< 0.05	< 0.05	0.43	0.42
<b>CV</b>	26.79	7.44	21.57	20.35	14.6	14.8	16.1	24.84	19.83

CV: coefficient of variation. Averages were compared using the LSD test (P < 0.05), when presented with uppercase letter there is a significant difference. TFCV and TV represent 100% of the concentration, respectively.

respectively, of 527.8 and 230.8 kg  $m<sup>3</sup>$ , considered 20 and 5% greater than TCFV (Table 1).

Howerver, the porosity of the TV was similar to the porosity of the TFCV (both ranging from 87.9 to 88.7%), considered adequate for the root development of the plants. Porosity is related to the water and oxygen available for plant growth in containers, with a value of 85% as a reference. The pores are responsible for gas exchange, and determine the movement of water in the container and the drainage pattern (Zorzeto, 2014). Martins et al. (2015) evaluating the porosity and organic carbon in an Oxisol with different uses and management in the Cerrado Mineiro verified that the distribution of pores presents a direct relation with the availability of water in the soil. Macropores (>0.25 mm) are related to water infiltration into the soil, while micropores are related to water retention (Ferraz-Almeida, 2022). Milner (2001) points out that the physical conditions of the substrate are considered the main factor for acquiring the substrate, since the conditions of pH and availability of nutrients can be modified with the use of fertigation.

After 70 days of transplanting, planting with  $TFCV_{75\%}$ +TV<sub>25%</sub> showed the highest dry and wet mass of shoots and tubers with respective means of 63.8; 6.4; 231.5; and 30.8 g (Table 2). This is the mixture that showed the least development in the first evaluation. TFCV showed the highest availability of all macro and micronutrients, therefore, it proved to be a better source of nutrients for the plant (Table 1). However, when applied alone, it showed the lowest mean root and aerial development.

TFCV is predominantly composed of organic matter derived from sphagnum fibers, rice husks, and expanded vermiculite. The availability of nutrients is influenced by the dynamics of organic matter, positively influencing the ability to exchange cations in the substrate/soil, and acting as a regulator of nutrient availability that can affect root development (Mikhael et al., 2019). Generally, the cation exchange capacity in minerals tends to be lower than that of organic matter (Almeida et al., 2019). This factor may explain the lower development of potatoes in the TV that is composed of organic material (sphagnum peat) and mineral (expanded vermiculite). In general, the higher the cationexchange capacity of a substrate/soil, promote the lower the frequency of fertigation (Favoretto, 2005). In this experiment, the frequency of fertilization was equivalent, explaining the better performance in substrates with higher input of organic material. For future studies, a possible effect of fertigation frequency and nutrient dynamics is recommended.

 The ratio of tuber growth was associated with shoot and root dry mass in the 40<sup>th</sup> day after transplanting. This correlation of tuber growth was higher with the root part (r: 0.77), indicating a greater association with tuber development (**Figure 2**A-B). However, after 70 days of transplanting, the period related to active tuber production, there was an intense relationship with aerial development (r: 0.66), and no correlation with root development (Figure 2C-D). This behavior was demonstrated by Yorinori (2003) in potato crops (cv. Atlantic), with positive tuber development rates with the root part of the plant in the period from planting to flowering, and an inversion of values after flowering, making a greater relationship with the aerial part of the plant. This result is explained by the high rates of photoassimilates and minerals that are exported from the shoot to the roots during flowering and tuber growth.



Figure 2. Relationship between dry mass of tuber, shoot and root of potatoes produced in mixtures of substrates based on sphagnum peat, coconut fiber, rice husks and vermiculite expanded vermiculite [TFCV] and sphagnum peat, and expanded vermiculite [TV] in different proportions (25%; 50%; 75%; and 100%) in the 40<sup>th</sup> and 70<sup>th</sup> day after transplanting. Correlations were significant and tested by Pearson's test (P<0.05). ns: no significant difference between treatments.

The relationship between the production of tuber and shoot was influenced by substrate mixtures (p<0.05). The isolated TFCV and TV promoted the highest tuber/shoot ratios with averages of 6.0 and 5.3 g of g tuber g shoot<sup>1</sup>, in the 40<sup>th</sup> day after transplanting (Table **3**). Plantings in TFCV and TV showed a more vigorous area development, consequently, with a greater solar

**Table 3.** Ratios of tuber [T], shoot [S] and root [R] of potato produced in mixtures of substrates based on sphagnum peat, coconut fiber, rice husks, and expanded vermiculite [TFCV] and sphagnum peat and expanded vermiculite [TV] in different proportions (25%; 50%; 75%; and 100%), in the 40<sup>th</sup> and 70<sup>th</sup> day after transplantation



of variation; averages were compared using the LSD test (P<0.05), when presented with uppercase letter there is a significant difference. ns: no significant difference between treatments.

interception, higher photosynthetic rate, and production of photoassimilates that were translocated to tuber growth. Corroborating the results found by Pereira et al. (2020), who obtained a high positive phenotypic correlation (r = 0.73) between tuber mass and leaf area index.

The ratios between tuber/root and shoot/root did not differ between substrate mixtures in the 40<sup>th</sup> day after transplanting. Also, there was no effect of substrate mixtures on any of the ratios in the 70<sup>th</sup> day after transplantation (Table 3).

Understanding these ratios is important for the balance between tuber production and the photosynthetic part of the plant. The balance between the parts is necessary to control water loss, maintaining the balance between the transpiring surface associated with a greater number of roots to supply the loss through transpiration, and thus having a greater capacity for absorbing water.

# *Classification of tuber*

The tuber classification showed a higher frequency of size for types II (mean: 79.44) > III (mean:  $50.24$ ) > I (mean: 19.24), respectively, with mean size between 23- 30mm; <23 mm; and 30-40 mm (**Table 4**). There was a predominance in tubers of medium and small sizes, which can be explained by a greater competition for nutrients and water, leading to the formation of smaller tubers.

The substrate mixtures showed a significant difference in the total weight of tuber. The TV promoted the highest total weight with the average of 3,100 g, followed by TFCV<sub>50%</sub> + TV<sub>50%</sub> with 3,000 g, considered an increase of 600 g in relation to use with the TFCV (**Figure 3**).

In Brazil, minitubers from tissue cultures are sold by the unit, unlike potatoes, seeds of older generations that are sold by the net mass of 30 kg boxes. Therefore, a box with smaller tubers is capable of having higher planting yields per area, as described by Filgueira (2008). Therefore, even without significant difference, TFCV<sub>50%</sub> + TV<sub>50%</sub> is presented as more attractive for seed potato production. Silva et al. (2006) evaluating different combinations of substrate in the production of minitubers from shoots of

**Table 4.** Size classification of minitubers into Type I, II, and III produced in mixtures of substrates based on sphagnum peat, coconut fiber, rice husks and expanded vermiculite [TFCV] and sphagnum peat and expanded vermiculite [TV] in different proportions (25%; 50%; 75%; and 100%)

Substrates	Tipo I <sup>ns</sup>	Tipo II <sup>ns</sup>	Tipo III <sup>ns</sup>						
	30-40 mm	23-30 mm	$<$ 23 mm						
<b>TFCV</b>	16.8	74.6	56.4						
TV	24.6	76.6	49.4						
$TFCV_{75\%}$ + $TV_{25\%}$	22.6	78.0	50.6						
$TFCV_{50\%} + TV_{50\%}$	17.4	83.6	52.2						
$TFCV_{25\%}$ + $TV_{75\%}$	14.8	84.4	42.6						
P (ANOVA)									
$P_{value}$	0.32	0.43	0.61						
$\overline{C}V$	42.95	12.27	27.14						

CV: coefficient of variation; averages were compared using the LSD test (P<0.05), when presented with uppercase letter there is a significant difference. ns: no significant difference between treatments.



**Figure 3.** Total weight of tubers in mixtures of substrates based on sphagnum peat, coconut fiber, rice husk and expanded vermiculite (TFCV) and sphagnum peat and expanded vermiculite (TV) in different proportions (25%; 50 %; 75%; and 100%). Means were compared using the LSD test (P<0.05), when presented with uppercase letter there is a significant difference.

three cultivars (Asterix, Monalisa and Ágata), did not observe significant differences in the number of seed potatoes. However, the authors observed that the Ágata cultivar had a higher production of number of tubers per plant.

## **Conclusion**

Substrate combinations influence the production of minitubers from potato plants derived from vitro cultivation. Substrate based on sphagnum peat and expanded vermiculite is a great alternative to increase the production of tubers due to the physical conditions of the substrate that promote better root development and a greater proportion of shoots and tuber mass. Higher tuber production showed a high correlation with plant root development. If it is necessary to mix substrates, it is recommended to mix (i) 50% substrate based on sphagnum peat, coconut fiber, rice husks and expanded vermiculite (ii) with 50% substrate based on sphagnum peat and expanded vermiculite. The addition of coconut fiber and rice husk increases the availability of nutrients in the substrate with a positive effect on the shoot development of the plants.

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**Conflict of Interest Statement:** The authors declare that the research was conducted in the absence of any commercial or financial relationships that could be construed as a potential conflict of interest.

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