







# Effect of potassium fertilization of robusta coffee mother plants on clonal seedlings growth

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## ABSTRACT

This study aims to evaluate the effects of different doses of potassium (K) on the growth of the clonal seedlings of *Coffea canephora* in a clonal garden. The mother plants of *C. canephora* coffee of the Robusta botanical variety in the clonal garden system from the Plant Breeding Program of Embrapa-RO in Brazil were used in the experiment. The potassium fertilizer was applied to these plants at different rates, including 0 (control), 40, 80, 120, 160, 200, and 240 kg ha<sup>-1</sup>, divided into 4 applications, during 150 days of the growth of the orthotropic stems (shoots). Cuttings were collected from the mother plants, and their dry matter and nutrient concentrations were determined. In addition, these cuttings were used for the production of seedlings, whose growth was then evaluated at 0, 48, 61, 80, 101, 122, 143, and 164 days after sticking the unrooted cutting into the growth media (DAS), and their vegetative traits were also analyzed at 122 DAS. The accumulation of nutrients in the clonal cutting showed the following order: K > N > Ca > P > Mg > S, irrespective of the dose of K applied to the mother plant, indicating that potassium was the nutrient accumulated in the greatest amount. Applying K doses up to 240 kg K<sub>2</sub>O ha<sup>-1</sup> during the cuttings growth period enhances vegetative traits and raises the Dickson Quality Index (DQI), thereby producing *C. canephora* clonal seedlings with superior physiological quality.

**Key words:** *Coffea canephora*; clonal garden; cutting nutrition; physiological quality; clonal seedlings.

## 1 INTRODUCTION

Potassium (K) is the second most accumulated nutrient in coffee plants. The nutrients most required by *Coffea canephora* coffee plants and accumulated in tissues are nitrogen, potassium, calcium, phosphorus, magnesium, and sulfur in descending order (Covre et al., 2016; Santos et al., 2021; Schmidt et al., 2022).

The accumulation of K is variable in different plant organs. Plant tissues showed the following order for K concentration: leaves > fruit > branches > stem > roots (Prezotti & Bragança, 2013). In addition, the nutrient requirements of plants vary according to the stage of the plant life cycle, with a high demand during both the vegetative growth and fruit maturation (Oliosi et al., 2021). Therefore, depending on not only the essential physiological processes, such as photosynthesis, stomatal opening, pollination, and fruit formation, but also the environmental and genetic factors, plants have different nutrient demands (Santos et al., 2021; Schmidt et al., 2022; Silva et al., 2021).

Potassium is the second-highest accumulated nutrient in the plant fruit after nitrogen, demonstrating the order of N > K > Ca > P > S > Mg (Covre et al., 2016, 2018). In the husk, however, potassium occupies the first place among other nutrients (Covre et al., 2016; Schmidt et al., 2022; Torres et al., 2022).

In leaves, K holds the third place, preceded by N and Ca, in the sequence of nutrients (Dubberstein et al., 2016). Besides, it ranks second in roots (Prezotti & Bragança, 2013), flowers (Santos et al., 2021; Schmidt et al., 2022), and secondary orthotropic stems (shoots) (Bazoni et al., 2020). The cuttings or the vegetative propagules, however, have reportedly shown the greatest accumulation of K (Bazoni et al., 2024; Kolln et al., 2022, 2024).

The potassium demand of all vegetative structures of the coffee plant indicates the necessity of its supply in adequate amounts according to the requirements of the plant for both its development and the production of fruits or cuttings, with the latter being produced in clonal gardens.

In this context, potassium plays an essential role in the production of clonal seedlings, where new plants are produced from the segments of young orthotropic stems (cuttings) (Verdin Filho et al., 2018). The functions that K plays in the seedling formation process include the maintenance of cell turgidity and the reduction of the dehydration rate of the cutting due to the decreased osmotic pressure, as well as the production of new plants from the vegetative structures of the original plant through cell expansion. Overall, K is involved in protein synthesis, carbohydrate metabolism, respiration, etc., which together lead to the greater root and leaf expansion (Cunha et al., 2009; Oosterhuis et al., 2014).

In addition to overall nutrient demand, potassium sources have distinct effects on soil and plant physiology. Potassium chloride (KCl), the most commonly used source, supplies both K<sup>+</sup> and Cl<sup>-</sup> ions, whereas potassium sulfate (K<sub>2</sub>SO<sub>4</sub>) also contributes sulfur (S) to the system. When KCl is used, careful management is essential to mitigate potential salinity effects (Faisal et al., 2024). Strategies to improve potassium use efficiency include splitting fertilizer doses and using controlled-release formulations, which ensure a gradual nutrient supply and enhance nutrient uptake (Damalas & Koutroubas, 2024).

Since mother plants are grown for different purposes, the management practices for potassium fertilization in clonal gardens are also expected to be different than those adopted for fruit production. However, in fruit production, K demand varies with seasons and peaks during the coffee bean filling (Dubberstein et al., 2016; Partelli et al., 2014), and for the production of cuttings, this demand may be determined by the maximum growth of the new shoots, which is variable, being especially dependent on both the climate characteristics of the growing region and the plant management practices implemented in the garden.

Since the exclusive use of clonal gardens for clonal production by cuttings is not yet a widespread technique adopted by Brazilian coffee growers, knowledge on the nutrition management of plants in this crop system is limited. Thus, this study was carried out with the aim of evaluating the influences of the application of different doses of K on the growth of clonal seedlings of *C. canephora* in a clonal garden.

## 2 MATERIAL AND METHODS

The experiment was conducted in the experimental field of Embrapa (10°43'55" S and 62°15'19" W; at an altitude

of 300 m) in the county/ municipality of Ouro Preto do Oeste, state of Rondônia (RO), Brazil, from August 20, 2018, to July 1, 2019. The region is dominated by the tropical monsoon climate (Am) according to the Köppen classification system (Alvares et al., 2013), with a mean annual temperature of 25 °C and a mean annual rainfall of 2,000 mm year<sup>-1</sup> and the rainy season from October and November to April and May.

The study is conducted in two distinct experiments. Experiment 1 consists of collecting cuttings produced in the field during the propagation material production period to analyze their dry matter accumulation and macronutrient concentrations. Experiment 2 involves seedling production in a nursery from these cuttings and evaluating their dry matter accumulation, growth, and vegetative characteristics 122 days after sticking (DAS).

The clonal cuttings were obtained from the *C. canephora* mother plants of the Robusta botanical variety from the plant breeding program of Embrapa, RO. A vertical clonal garden was set up with a soil classified as an *Argissolo Vermelho-Amarelo* according to the Brazilian Soil Classification System in November 2016 (Santos et al., 2018). The spacings between rows and between plants were 2.0 and 0.5 m, respectively, with a total of 10,000 plants ha<sup>-1</sup>. The plants, which were trained in the clonal garden with only one orthotropic stem and prevented bending, were managed according to the technical recommendations of the model of the vertical clonal garden proposed for the production of Robusta coffee seedlings (Espindula et al., 2022).

The initial chemical properties of the soil of the experimental area were determined in the 0-20, 20-40, and 40-60 cm soil layers before establishing the treatments in August 2018 (Table 1). Thereafter, 50 grams of dolomitic limestone (Effective Calcium Carbonate Equivalent; ECCE) was applied

**Table 1:** Chemical properties of the *Argissolo Vermelho-Amarelo* soil in the clonal garden of the experimental field of Embrapa in Ouro Preto do Oeste, Rondônia, Brazil, in 2018.

Sample cm	pH water	P mg dm <sup>-3</sup>	K	Ca	Mg	H+Al	Al	CEC
				----- cmol <sub>c</sub> dm <sup>-3</sup> -----				
0 - 20	4.36	53.13	0.35	1.11	0.76	8.01	0.88	10.24
20 - 40	4.63	13.75	0.36	1.02	0.45	4.81	0.54	6.64
40 - 60	4.98	5.50	0.45	1.59	0.53	3.37	0.05	5.94
Sample cm	OM g kg <sup>-1</sup>	m	V	Cu	Fe	Mn	Zn	
		----- % -----		-----mg/ dm <sup>-3</sup> -----				
0 - 20	15.20	31.00	21.13	42.61	230.16	119.27	49.98	
20 - 40	7.70	25.50	28.00	19.38	247.83	120.34	46.24	
40 - 60	4.95	2.50	43.00	15.75	159.01	119.74	40.98	

Soil pH was measured using water at a ratio of 1:2.5. OM: organic matter (wet digestion); P: phosphorus; K: potassium; Ca: calcium; Mg: magnesium; Al: aluminum; Cu: copper; Fe: iron; Mn: manganese; Zn: zinc. P and K contents were determined using the Mehlich 1 method. Exchangeable Ca, Mg, and Al were extracted with 1 mol dm<sup>-3</sup> KCl. H+Al: potential acidity; CEC: cation exchange capacity; V: base saturation; m: Al<sup>3+</sup> saturation.

per plant, covering a total area of 0.5 m<sup>2</sup>, involving the spacings of 0.5 m within the row and 1.0 m between the rows and a plant spacing of 0.5 m, in September.

The experimental treatments consisted of seven potassium doses, applied as potassium chloride (KCl), including 0, 40, 80, 120, 160, 200, and 240 kg K<sub>2</sub>O ha<sup>-1</sup> during the growth cycle, and clonal cuttings were grown from August 2018 to January 2019. The treatments were arranged in a completely randomized design (CRD) with ten replications.

The seven doses of potassium chloride fertilizer were applied four times at 30-day intervals, with the first application beginning on August 20, 2018, 30 days after the initial standardization. In addition, plants received 150 kg ha<sup>-1</sup> of nitrogen fertilizer during the growth cycle in the form of urea (45 % N) in four split applications, together with the potassium. Furthermore, in October 2018, to meet the needs of plants for micronutrients, boron was applied as boric acid at the rate of 30 kg ha<sup>-1</sup>, while the application rate of zinc in the form of zinc sulfate was 30 kg ha<sup>-1</sup>.

The cuttings were collected from the orthotropic stems (shoots) of mother plants that grew for 150 days for the production of seedlings, and their weight and nutrient concentrations were evaluated. The cuttings were detached from the orthotropic stems by a straight cut 0.5 cm above the joint with the plagiotropic branches and 6 cm below the node or the leaf joint, with the omission of the basal and apical segments of the stems and leaves (Bazoni et al., 2020). Finally, after the removal of plagiotropic branches, the length of the leaf blade was reduced to about two-thirds to avoid water loss and thus dehydration (Dias et al., 2012).

## 2.1 Experiment 1 - Dry matter accumulation and macronutrient concentrations of the clonal cuttings

The dry matter and nutrient concentrations were evaluated in 20 cuttings collected from the stems of mother plants from each treatment. After sampling, these cuttings went through homogenization and then were divided into four equal replications.

First, the cuttings were allowed to dry in a laboratory drying oven with forced air circulation at 65 °C until a constant weight was achieved, and then, their weight was determined using an analytical balance. After grinding in a Wiley mill, they were transferred to the Soil and Plant Nutrition Laboratory of Embrapa Rondônia.

The determination of the concentrations of nutrients such as nitrogen (N), phosphorus (P), potassium (K), calcium (Ca), magnesium (Mg), and sulfur (S) was carried out in the laboratory. N content was determined using the semi-micro Kjeldahl method by distillation with sulfuric acid as a digestion medium. To determine P content, molecular spectrophotometry

was used, while K content was measured by flame photometry. Moreover, the measurement of the contents of nutrients such as Ca, Mg, and S was done by plasma spectrophotometry using the nitric acid-perchloric acid digestion method (Silva, 2009). The results of nutrient concentrations were expressed as grams of dry weight per cutting (g cutting<sup>-1</sup>).

## 2.2 Experiment 2 - Dry matter accumulation and growth rates and vegetative characteristics of seedlings at 122 days after sticking the cuttings (DAS)

The cuttings taken from the mother plants were transferred to a nursery on the same day of collection for growth analysis. 50 % shading and a mist irrigation system were provided in a nursery to induce the growth and root formation of seedlings.

The cuttings with four replications from each treatment were combined into a composite sample, and thereafter, to evaluate the performance of the produced seedlings, a new experiment was carried out in a nursery. The experimental design consisted of a 7 × 8 factorial arrangement of treatments including seven doses of potassium fertilizer, as previously mentioned, applied to the mother plants in a field and eight evaluation periods, namely 0 (initial weight of the cutting), 48, 61, 80, 101, 122, 143, and 164 DAS. The experiment was laid out in a completely randomized design (CRD) with treatments replicated four times and six cuttings/seedlings in each experimental plot.

The individual cuttings prepared for planting were submerged in the 25 % fungicide azoxystrobin (Amistar® WG). First, the 280-cm<sup>3</sup> returnable polyethylene plug pots were filled with the commercial substrate (Tropstrato HT-Vida Verde®) containing a mixture of pine bark, expanded vermiculite, single superphosphate, and potassium nitrate. This product contains a moisture content of 60 %, water holding capacity (WHC) of 130 % (w/w), and density of 200 kg m<sup>-3</sup> (on a dry basis) and 500 kg m<sup>-3</sup> (on a wet basis), as well as a pH value of 5.8 and an electrical conductivity (EC) of 0.5 mS cm<sup>-1</sup>, both at the ratio of 5: 1 (water: substrate). Every 100 kg of substrate was then supplemented with 0.5 kg of the fertilizer Basacot® Plus 6M containing 16 % N, 8 % P, 12 % K, 2 % Mg, 5 % S, 0.4 % Fe, 0.02 % B, 0.02 % Zn, 0.05 % Cu, 0.06 % Mn, and 0.015 % Mo and 0.2 kg triple superphosphate.

Next, each cutting was placed in plug pots, which were then placed on trays on the raised benches in a nursery and were constantly irrigated using a mist irrigation system with a timer to ensure an adequate water supply. The programmed irrigation system was activated every 5 minutes for 10 seconds during the first 30 days, every 20 minutes for 30 seconds from 30 to 90 days, and every 30 minutes for 60 seconds from 90 to 120 days. From 120 to 164 days, however, the seedlings were irrigated manually three times a day, e.g., 8:00 a.m., 12:00 noon,

and 4:00 p.m. The seedlings underwent no acclimatization period because they were damaged at the evaluation times due to the changes in the growth environment.

The seedlings were first collected from the plug pots, washed to remove the substrate, and then dried in a forced-air drying oven at 65 °C until a constant weight was reached. Thereafter, their weight was evaluated using an analytical balance at 0 (initial weight of the cutting), 48, 61, 80, 101, 122, 143, and 164 DAS. Based on the weights, the curves of dry matter accumulation (DMA), absolute growth rate (AGR; mg day<sup>-1</sup>), and relative growth rate (RGR; mg g<sup>-1</sup> day<sup>-1</sup>) were drawn. At 122 DAS, 50 cm<sup>3</sup> of the calcium nitrate solution was given to seedlings, each plug pot receiving 5 cm<sup>3</sup> dm<sup>-3</sup>.

The dry matter accumulation was determined by directly determining the total dry matter of the complete plants (including the cutting). To calculate the AGR, the following equation was used:  $AGR = (M2 - M1) / (T2 - T1)$ , where M1 and M2 indicate the mass of dry matter of the two consecutive samples taken at the times T1 and T2, respectively. The RGR was calculated as follows:  $RGR = (\ln M2 - \ln M1) / (T2 - T1)$ , where ln denotes the Napierian logarithm and M1 and M2 represent the dry matter at the times T1 and T2, respectively (Peixoto, Cruz, & Peixoto, 2011).

At 122 DAS, in addition to dry matter accumulation and growth rates, the vegetative traits, average time for the formation of clonal seedlings of *C. canephora* coffee plants grown in the State of Rondônia were evaluated (Espindula et al., 2015).

Evaluations included 1) stem length (SL), which was directly measured from the joint of the shoot to the apical meristem on the cutting; 2) stem diameter (SD), which was determined at the base of the branch, 3 cm above the joint of the shoot; 3) number of roots (NR) which was evaluated by direct counting; 4) root volume (RV), which was determined by immersing the roots in a graduated cylinder using a water volume displacement technique; 5) shoot dry matter (SDM), determined with the shoot cutting; 6) root dry matter (RDM), determined using an analytical balance after drying in an air forced laboratory oven at 105 °C until achieving a constant weight; 7) total dry matter (TDM), estimated by the sum of SDM and RDM; 8) leaf area (LA), determined using the Determinador Digital de Áreas (Digital Determination of Areas (DDA) software (Ferreira, Rossi, & Andrighetto, 2008); and 9) Dickson quality index (DQI), calculated using the following formula:  $DQI = [TDM / [(SL/SD) + (SDM/RDM)]]$  (Dickson, Leaf, & Hosner, 1960).

### 2.3 Statistical Analysis

The data were analyzed using analysis of variance (ANOVA) and regression analysis. The selection of mathematical models in this study was carried out based on the criterion of maximum parsimony (Ferreira, 2018) and involved two characteristics, namely the highest estimates of the coefficients of determination (R<sup>2</sup>) and the greatest

simplicity of the model according to the response of the biological phenomenon. To evaluate the significance of the regression coefficients ( $\beta_i$ ), analysis of variance was used at a 5 % level of significance ( $p \leq 0.05$ ). The scatterplots of the means of all characteristics, including those that were not significantly affected by treatments, were created based on the following Equations 1, 2 and 3:

$$\hat{Y} = \beta_0 + \beta_1 x \quad (1)$$

$$\hat{Y} = \beta_0 + \beta_1 x + \beta_2 x^2 \quad (2)$$

$$\hat{Y} = \beta_0 + \beta_1^x \quad (3)$$

Where  $\hat{Y}$  indicates the response variable (dry matter and nutrient concentrations), while x denotes the explanatory variable (potassium doses); and  $\beta_0$ ,  $\beta_1$ ,  $\beta_2$  and are regression coefficients that quantify the values of changes in the response variable with changes in the explanatory variable.

## 3 RESULTS

According to the ANOVA results (Table 2), potassium fertilization significantly affected K concentration in cuttings ( $p < 0.05$ ), as well as key vegetative traits, including stem length ( $p < 0.01$ ), root number ( $p < 0.05$ ), and leaf area ( $p < 0.05$ ). The coefficients of variation (CV%) were below 20% for most traits, which demonstrates the experimental precision and reliability of the results.

### 3.1 Experiment 1 - Dry matter accumulation and macronutrient concentrations in clonal seedlings

The dry matter production of the cuttings was not affected by the increase in the dose of K<sub>2</sub>O, with a mean value of 1.28 g cutting<sup>-1</sup> (Figure 1).

Contrarily, different doses of potassium affected the concentrations of K, Ca, and Mg in the cuttings from *C. canephora* (Figure 2). A linear increase was observed for K concentration, while Ca and Mg exhibited a decreasing linear trend. In contrast, other nutrients, e.g., N, P, and S, were not affected as the K<sub>2</sub>O doses increased.

### 3.2 Experiment 2 - Dry matter accumulation growth rates and vegetative characteristics of seedlings at 122 days after sticking the cuttings (DAS)

Dry matter (DM) increased exponentially during the formation of seedlings, regardless of K<sub>2</sub>O dose. The cuttings of similar weight were used for seedling formation, but over

time, the seedlings produced from the cuttings subjected to the higher doses of  $K_2O$  exhibited the greatest dry matter accumulation. This trend was particularly observed at 200 and 240 kg  $K_2O$  ha<sup>-1</sup>, with the highest cumulative weight of seedlings (4.47 g seedling<sup>-1</sup> and 4.84 g seedling<sup>-1</sup>, respectively), whereas, at 0 or 40 kg  $K_2O$  ha<sup>-1</sup>, the seedlings gave the lowest cumulative weight, e.g., 3.54 g seedling<sup>-1</sup> and 3.42 g seedling<sup>-1</sup>, respectively, at the end of the growth cycle (Figure 3).

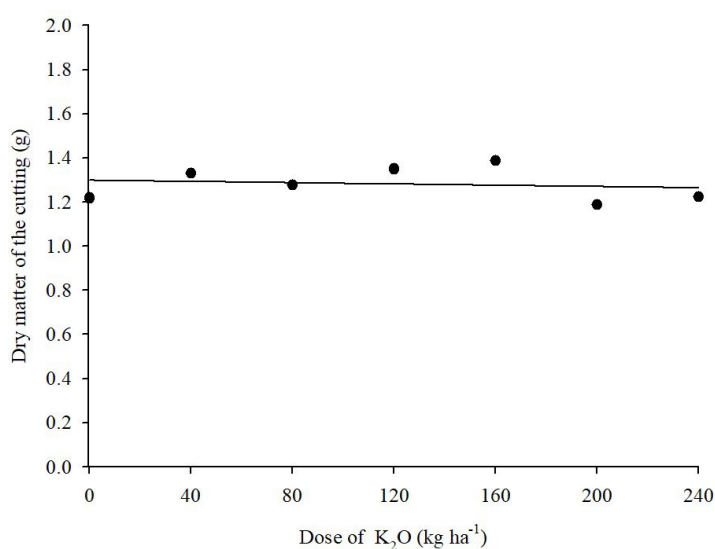
Concerning the absolute and relative growth rates, no interaction was found between the evaluation periods and  $K_2O$  doses, and therefore, these two parameters had moderate values over time (Figure 4). The mean values of

both the absolute and relative growth rates experienced an initial slow growth up to 48 DAS, which was, however, followed by a rapid growth up to 61 DAS, and they continued increasing linearly up to 122 DAS. From 122 DAS, on which calcium nitrate fertilizer was applied, up until 143 DAS, plants grew steadily. At 143 DAS, plants underwent substantial growth up to 164 DAS. The growth rates were found not to fit any mathematical model; they, however, achieved two peaks, e.g., AGR of 17.65 mg day<sup>-1</sup> and RGR of 10.03 mg mg<sup>-1</sup> day<sup>-1</sup> on 61 DAS and AGR and RGR of 38.64 mg day<sup>-1</sup> and 10.43 mg mg<sup>-1</sup> day<sup>-1</sup>, respectively, on 164 DAS (Figure 4).

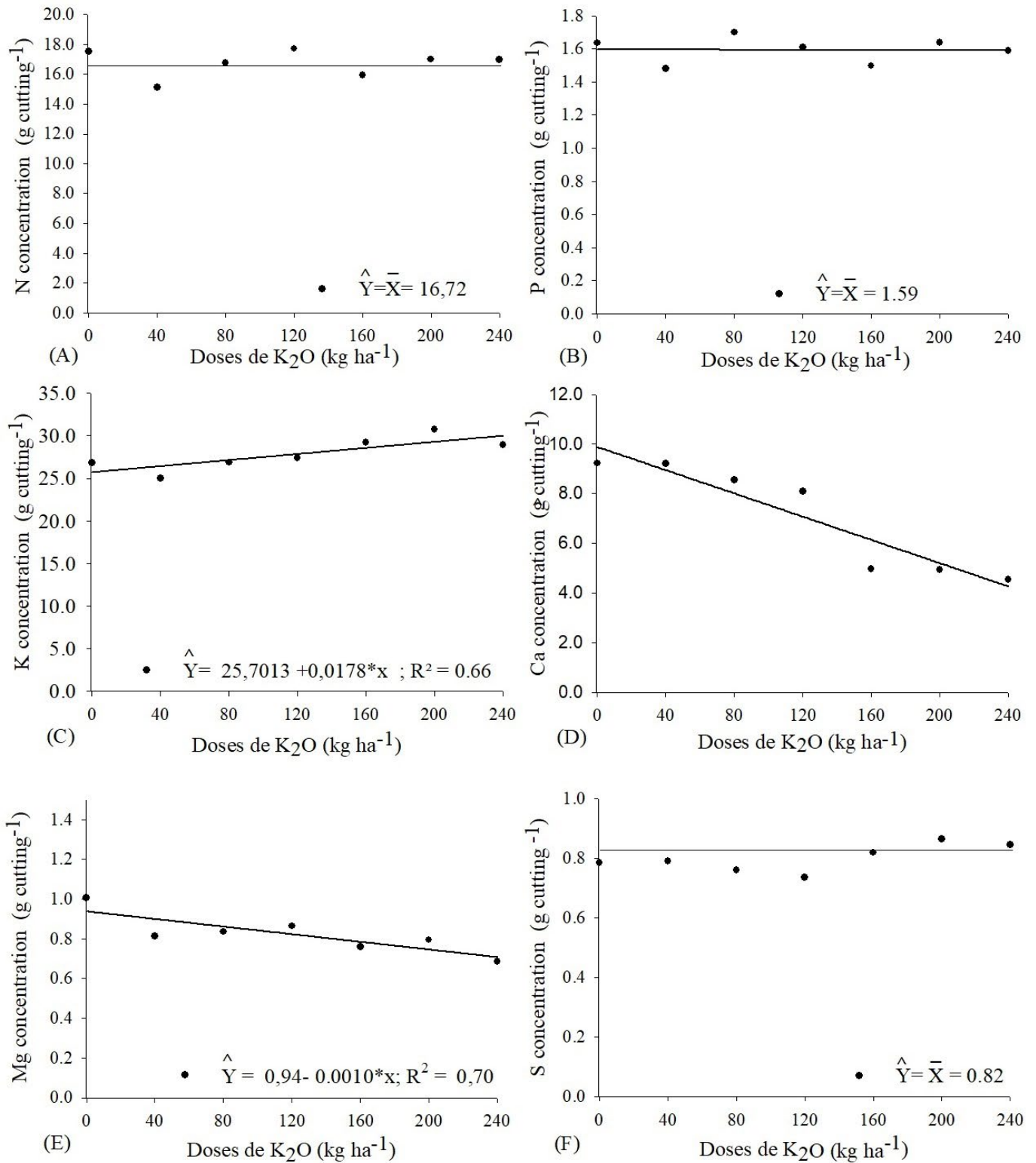
**Table 2:** Summary of analysis of variance (ANOVA) for growth and nutritional traits of *Coffea canephora* managed with different potassium doses.

FV	DF	DM	N	P	K	Ca	Mg	S	SL
Blocks	3	0.04	87.13	0.55	22.31	58.40	0.33	0.03	21.04
Treatments	6	0.02 <sup>NS</sup>	3.19 <sup>NS</sup>	0.02 <sup>NS</sup>	16.85*	18.16 <sup>NS</sup>	0.02 <sup>NS</sup>	0.02 <sup>NS</sup>	7.38**
Residue	18	0.01	2.70	0.03	5.83	7.41	0.02	0.01	1.20
Mean		1.29	16.94	1.61	28.37	7.02	0.82	0.77	8.08
CV(%)		8.6	9.71	10.36	8.51	38.8	18.70	12.73	13.59
FV	DF	SD	NR	RV	LA	DQI	SDM	RDM	TDM
Blocks	3	0.03	4.53	0.7139	11439.79	0.00	0.28	0.00	0.22
Treatments	6	0.04 <sup>NS</sup>	2.54*	0.46 <sup>NS</sup>	2754.91*	0.01 <sup>NS</sup>	0.13 <sup>NS</sup>	0.01 <sup>NS</sup>	0.15 <sup>NS</sup>
Residue	18	0.04	0.82	0.31	735.69	0.01	0.07	0.02	0.08
Mean		3.58	5.98	2.86	140.90	0.40	2.45	0.46	2.91
CV(%)		5.58	15.18	19.49	19.25	22.23	11.12	27.37	9.90

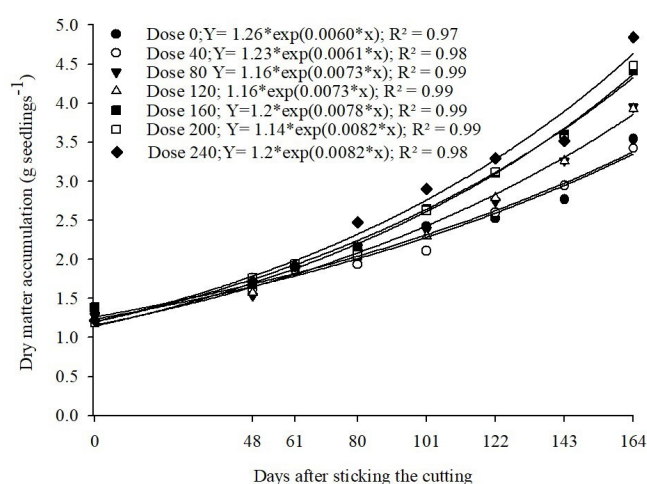
<sup>NS</sup> not significant; \* significant at 5% probability; \*\* significant at 1% probability; SV: source of variation; DF: degrees of freedom; DM: dry matter of cuttings; N: nitrogen; P: phosphorus; K: potassium; Ca: calcium; Mg: magnesium; S: sulfur; SL: Stem length; SD: stem diameter; NR: number of roots; RV: root volume; LA: leaf area; DQI: Dickson Quality Index; SDM: shoot dry matter, RDM: root dry matter; TDM: total dry matter; CV: coefficient of variation.



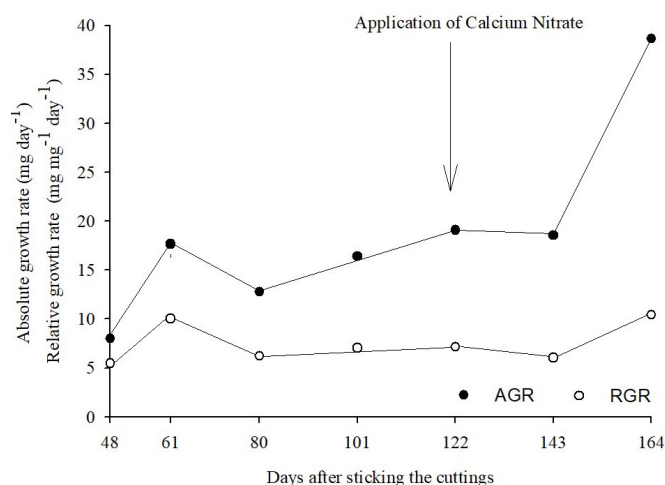
**Figure 1:** Dry matter production in clonal cuttings taken from *Coffea canephora* mother plants grown under different potassium doses in a clonal garden system in Ouro Preto do Oeste, RO, Brazil, in 2019.



**Figure 2:** Concentrations of nitrogen (A), phosphorus (B), potassium (C), calcium (D), magnesium (E), and sulfur (F) in the cuttings taken from *Coffea canephora* coffee plants grown under different doses of potassium.



**Figure 3:** Dry matter accumulation in the seedlings of *Coffea canephora* coffee plants formed from cuttings grown under different doses of potassium.



**Figure 4:** Absolute growth rate (AGR) and relative growth rate (RGR) of the seedlings of *Coffea canephora* coffee plants formed from cuttings exposed to different doses of potassium.

All vegetative characteristics of seedlings, including SL, SD, NR, RV, LA, DQI, SDM, RDM, and TDM, displayed a linear increase in response to the application of different doses of K<sub>2</sub>O to the mother plants (Figure 5).

## 4 DISCUSSION

### 4.1 Experiment 1 - Dry matter production and macronutrient concentrations in clonal cuttings

The dry matter of cuttings remained constant irrespective of potassium doses, which may have occurred through the distribution of photo-assimilates within the vegetative shoots of mother plants due to the production of a larger number of

secondary stems and cuttings, as well as a greater fresh and dry matter of stems in response to the increased potassium doses.

Furthermore, in our study, with the application of different doses of K, an increase in its concentration in the cuttings was also noticed, which is due to its higher availability in the soil, facilitating its uptake by mother plants. Various important roles of K in plants, e.g., the growth of the whole plant, metabolic functions, stress tolerance, and growth and development of the root system, have been reported (Sustr, Soukup, & Tylova, 2019). The presence of K at higher concentrations in the cuttings, which can favor seedling development, particularly by both reducing dehydration and increasing the root and leaf expansion, promotes the development of the root system (Oosterhuis et al., 2014).

Moreover, cuttings exhibited reduced concentrations of Ca and Mg with the increase in K supply, possibly due to competition among these cations induced by an increase in K concentrations in the tissues. Plants preferably absorb K over other cations because it is a monovalent cation and has a smaller ionic radius (Wahab et al., 2025). In addition, an initial concentration of Ca for absorption by mother plants was low because it may not have been supplied by liming in time.

Additionally, the application of K<sub>2</sub>O had no effect on N concentrations in only the husk (Torres et al., 2022) and cuttings (Bazoni et al., 2020; Kolln et al., 2022) of coffee plants. Normally, N is predominant over K due to an antagonistic interaction between them (N×K). In other words, an increase in N level reduces K accumulation, while with an increase in K doses, no reduction in N accumulation occurs (Prado et al., 2008).

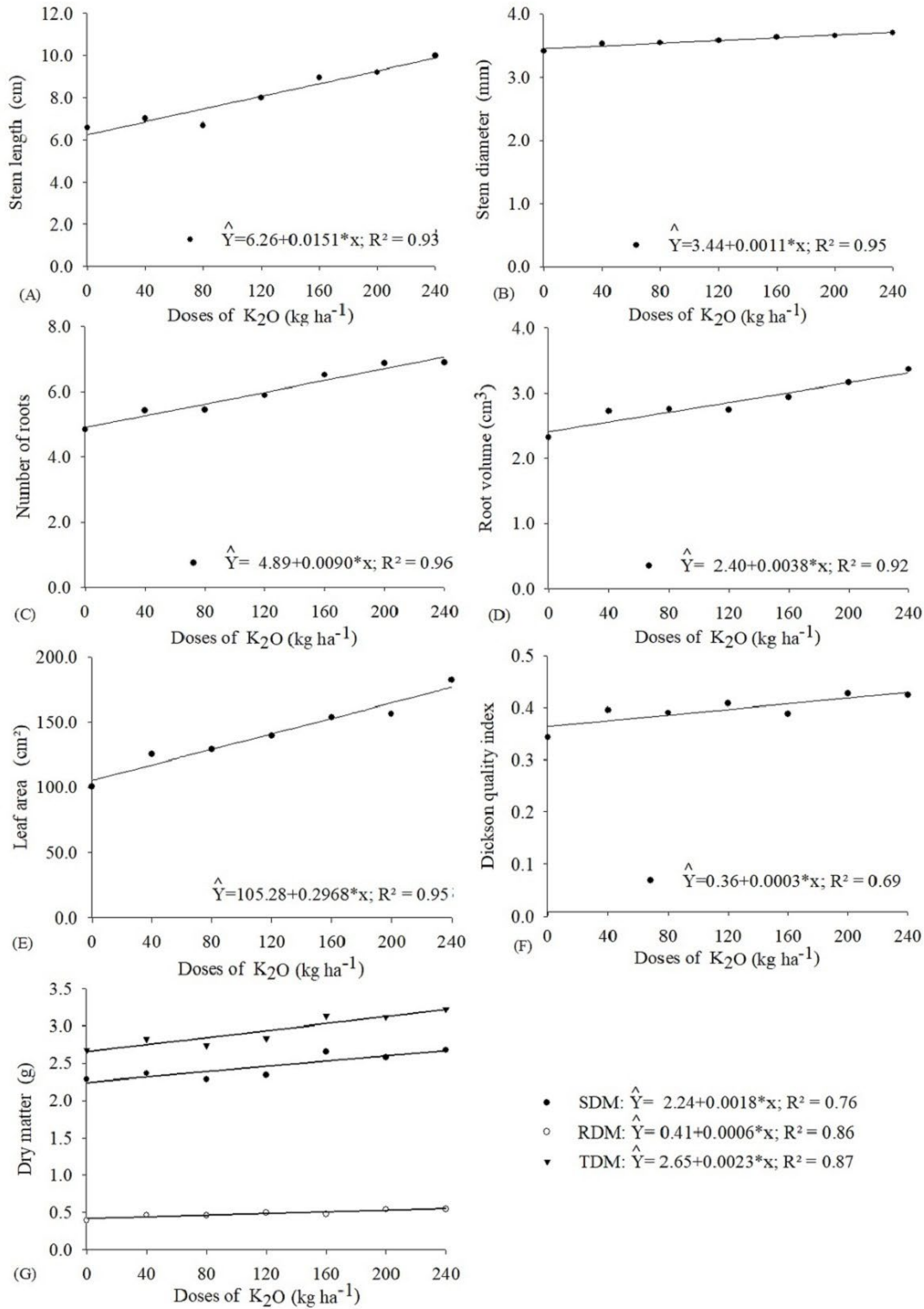
The cuttings of coffee plants exhibited the micronutrient accumulation order of K > N > Ca > P > Mg > S, which was similar in an experiment conducted by Kolln et al. (2022), who evaluated the effects of an increase in N doses, and also in a study by Bazoni et al. (2020) on Conilon coffee plants, in which, however, the accumulation of Mg was higher than that of P. This finding clearly highlights the importance of nutrients such as K, N, and Ca, while not neglecting the other macronutrients because the concentrations of mineral nutrients in the mother plant and, consequently, in cuttings are closely related to the adventitious root formation of propagules, and thus, play a decisive role in the production of high-quality seedlings (Ferreira et al., 2019).

### 4.2 Experiment 2 - Dry matter accumulation growth rates and vegetative characteristics of seedlings at 122 days after sticking the cuttings (DAS)

There was a slow growth and dry matter accumulation during the initial phase of seedling development (48 DAS). Since during this phase, the plant's roots have not yet

developed, the uptake of water and nutrients from the substrate is limited, and thus, it is considered a critical period for its survival (Petry et al., 2012). Overall, the nutrient reserves and

photoassimilates in cuttings are necessary for survival and the promotion of root formation (Cunha et al., 2009; Dias et al., 2012).



**Figure 5:** Vegetative characteristics of the seedlings of *Coffea canephora* coffee plants formed from cuttings grown under different doses of potassium at 122 DAS; Stem length (A), stem diameter (B), leaf area (C), number of roots (D), root volume (E), Dickson quality index (F), and shoot, root, and total dry matter (G).

The establishment of the root system and development of shoots occur simultaneously, which support the accumulation of dry matter until the final period of evaluation (164 DAS). The seedlings produced from the cuttings of the same weight started to differ from each other at DAS under the elevated concentrations of K. The cuttings with a higher K concentration formed seedlings that produced a higher dry matter, which emphasizes the importance of K for the plant root formation and growth (Cunha et al., 2009). In addition, the K<sup>+</sup> ion plays fundamental roles in various processes in plants, including homeostatic control, internal transport of substances and energy, response mechanisms to biotic and abiotic stresses, plant growth, and metabolic control (Sardans & Peñuelas, 2021).

The AGR and RGR curves were similar and showed two peaks of growth. The analysis of these curves describes the growth patterns and the physiological aspects of the plants throughout the experimental period (França et al., 2014). During the two periods of evaluation, e.g., from 48 to 61 and 143 to 164 DAS, the growth rates of seedlings were higher, that not only is attributed to the emergence and formation of roots, which may be attributed not only to the emergence and formation of roots, favoring water and nutrient absorption from the substrate, but also possibly to the fertilization with calcium nitrate.

Between 61 and 80 DAS, growth rates declined, which, however, recovered in subsequent evaluation periods. This unexpected reduction may have taken place as a result of the low water content in the substrate during this period because from 30 to 90 DAS, irrigation was applied to the seedlings every 20 minutes for 30 seconds, which indicates the necessity of the application of water at greater intensity and lower frequency throughout this period.

At the end of the evaluation period, dry matter did not accumulate at a constant speed, which is generally due to the low volume of the container (Espindula et al., 2018; Giuriatto Júnior, 2020), regardless of the doses of K applied. Nevertheless, there was a nutrient limitation, because from 122 to 143 DAS, stable growth was witnessed, and subsequently, the growth peaked only at 164 DAS after nutrient supply.

The seedlings were evaluated on day 42 after 122 DAS for their status of development after they passed through the normal growing period in the nursery to check whether any type of limitations, like the volume of the container, arose (Espindula et al., 2018). The results of growth characteristics of plants receiving calcium nitrate Ca (NO<sub>3</sub>)<sub>2</sub> suggest that container volume was not a limiting factor for their growth. The substrate used, however, showed nutrient limitation. The application of substrate supplements is recommended to be adopted as a management practice to promote the growth of seedlings in organic substrates, even with the use of controlled-release complex fertilizers. The substrate used,

however, may have presented nutrient limitations. This suggests that supplementing organic substrates could be a useful management practice to improve seedling growth, even when controlled-release fertilizers are applied.

In both the cuttings and the substrate, calcium availability was low because of the lack of Ca in the Basacote® fertilizer used in the preparation of the substrate. The roles this nutrient has in plants are the involvement in auxin metabolism, cell division, peroxidase activity, and cell elongation (Cunha et al., 2009). In addition, it not only contributes to the structural rigidity of the plant due to its pivotal function in cell-wall formation but also makes up the middle lamella (Taiz et al., 2017). The growth of the seedlings of woody species may be particularly constrained by the lack of Ca (Gonçalves et al., 2013). Therefore, an effective nutritional management that aims to increase the Ca concentrations in the substrate allows for the better development of seedlings, which have higher dry matter (Gonçalves et al., 2013), height, and diameter (Estevez et al., 2020).

The increase in the application rate of potassium fertilizer to mother plants in the clonal garden was found to improve all the evaluated vegetative traits, leading to the production of seedlings with better physiological quality. The higher availability of nutrients present in the cuttings is the reason for this observation, since K<sup>+</sup> performs both physiological and metabolic functions in the plant, such as the internal transport of substances and energy and the improvement of its ability to respond to biotic and abiotic stresses (Sardans & Peñuelas, 2021).

The positive effect of K on plant shoots was manifested as increases in SL, SD, LA, and SDM, which resulted in greater shoot growth. The potential of K<sup>+</sup> in stimulating plant growth (Nieves-Cordones et al., 2016) due to its various role in the maintenance of cell turgidity and control of the osmotic pressure in the stomatal guard cells, and consequently, stomatal opening and the synthesis of abscisic acid (ABA) and auxin and their interactions, explains the positive effect of this nutrient on shoot growth parameters (Osakabe et al., 2013).

Furthermore, in the presence of K, the assimilates are translocated from the phloem to the shoots, which is conducive to the greater length and development of seedlings (Sardans & Peñuelas, 2021). The greater leaf areas, indicating the optimal plant growth and development, which is related to a better physiological state (Partelli et al., 2006) and higher dry matter, corroborate our obtained result (Cruz et al., 2007). The higher leaf area enhances the interception of sunlight, resulting in the increased assimilation of carbon, and thus higher photosynthetic rates (Sales et al., 2017; Valadares et al., 2016).

Similarly, potassium fertilization of the coffee mother plants yielded better results for NR, RV, and RDM in roots, which are linked to the propagule quality. The reason is the

physiological status of the mother plant, which affects the process of rooting in cuttings (Picolotto et al., 2015), and the nutrient concentrations of the propagules determine the formation, quantity, length, and density of adventitious roots (Cunha et al., 2009).

The efficiency of the root system is dependent on dry matter and root volume (Albertino et al., 2012), and seedlings with a well-developed root system have a higher drought stress tolerance and survive better after transplanting into the field (Grossnickle, 2012).

The growth and development of the root system were positively influenced by potassium fertilization owing to the participation of  $K^+$  in metabolism, growth, and stress adaptation in the plant (Nieves-Cordones et al., 2016). It has multiple functions in root growth, the establishment of root system architecture, root growth at the cellular level, root hair growth, and responses of the root system to stress factors (Sustr, Soukup, & Tylova, 2019).

The combined evaluation of all vegetative traits of seedlings revealed that with the increase in K doses, better results were produced for the DQI, which is an indicator of seedling quality and shows the balance of the distribution of biomass throughout the plant. Both the shoot and root dry matter increased in response to K addition, which was reflected by total dry matter.

The balance between the shoot and root of the plant facilitates the reduction of water loss by transpiration and its uptake by roots (Grossnickle, 2012). This occurs due to the association between dry matter production in the shoot and dry matter accumulation in the leaf, since the leaf is an organ responsible for the photosynthesis process and has a central role in plant growth. Likewise, dry matter production in roots is dependent on the efficient uptake of water and nutrients, which is also related to vital plant functions in the shoot, thus contributing to plant growth (Costa et al., 2022). In this context, due to its physiological and metabolic functions in the plant, e.g., the internal transport of substances and energy and responses to biotic and abiotic stresses,  $K^+$  can affect the root/shoot biomass ratio (Sardans & Peñuelas, 2021).

These overall results emphasize the importance of the nutritional conditions of mother plants in the physiological quality of the cuttings, which is in line with the findings of other studies (Cunha et al., 2009; Dias et al., 2012; Epedu et al., 2025; Oliveira et al., 2019). In our study, the positive results of the fertilization of mother plants with  $K_2O$  reinforce the need for optimum K fertilizer application to the donor plant from which the cuttings were taken.

It is crucial to acknowledge that potassium was applied in the form of potassium chloride (KCl). Consequently, potential implications of chloride ( $Cl^-$ ), including its influence on nutrient uptake and salinity tolerance, cannot be ruled out and should be considered a limitation of this investigation.

## 5 CONCLUSIONS

Increasing  $K_2O$  doses applied to mother plants did not affect dry matter in cuttings but increased  $K^+$  and reduced  $Ca^{2+}$  and  $Mg^{2+}$  concentrations. Nutrient accumulation in clonal cuttings followed the order  $K > N > Ca > P > Mg > S$ , regardless of K dose. Potassium rates up to 240 kg  $K_2O$   $ha^{-1}$  improved vegetative growth and Dickson Quality Index, enhancing clonal seedling quality, although no optimal dose was identified and potential  $Cl^-$  effects should be considered.

## 6 AUTHOR CONTRIBUTIONS

Conceptual Idea: Kolln, A.M.; Espindula, M.C.; Rocha, R.B.; Araújo, L.F.B.; Methodology design: Espindula, M.C.; Rocha, R.B.; Data collection: Kolln, A.M.; Espindula, M.C.; Kolln, F.T.; Data analysis and interpretation: Kolln, A.M.; Espindula, M.C.; Rocha, R.B.; Araújo, L.F.B.; Campanharo, M.; Writing and editing: Kolln, A.M.; Espindula, M.C.; Rocha, R.B.; Araújo, L.F.B.; Campanharo, M.; Kolln, F.T.

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## 8 DATA AVAILABILITY STATEMENT

Data available upon request to authors.

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