

# Modelling sugarcane root elongation in response to mechanical stress as an indicator of soil physical quality

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#### Summary

The root elongation rate represents a biophysical process that can be directly affected by mechanical, water, thermal, and gaseous stresses in the soil to be used as a soil physical quality indicator. The objective of this study was to determine sugarcane root growth parameters under soil physical stress for different root diameter classes in an Oxisol from the Southeast of Brazil. The experimental design was entirely randomized in a factorial scheme  $5 \times 2$  (mechanical  $\times$  water stress) with three replications. The factor mechanical stress was composed of five compaction levels (1.04; 1.12; 1.19; 1.28; 1.36 Mg m<sup>-3</sup>). The factor water stress was composed of two matric potentials (-6 kPa and -33 kPa). Soil samples were collected from the 0.0–0.2 m layer of an Oxisol with a clayey texture. Pre-sprouted sugarcane seedlings were transplanted and conditioned in a growth chamber. Root length, volume, surface area, and diameter were quantified to generate root growth models as a function of physical stresses in the soil. Soil penetration resistance increases from 1.4 to 5 MPa reduced root elongation rate from 3.5 to 1.35 cm day<sup>-1</sup> (-59%) and the average number of roots from 11 to 6 segments (-45%), respectively. The root volume, surface area, and length were reduced because of the increase in the compaction level. Coarse root diameter (1-2 mm) was weakly impacted by mechanical stress, whereas fine root diameter (0.5-1 mm) was more growth limited in compacted soils. The root elongation rate of sugarcane was modelled as a function of mechanical and water stress. Mechanical stress mainly affects the growth of sugarcane roots with small diameter.

Keywords: Saccharum sp.; soil penetration resistance; root growth; biophysical parameterization

# Introduction

The soil compaction as a result of bulk density increases impairs soil structure and changes the pore spaces (Oliveira *et al.* 2022), water dynamics (Rossetti & Centurion 2013), gas dynamics (Pandey & Bennett 2024), and mechanical conditions for root growth (Pandey *et al.* 2021) and, consequently, impairs sugarcane productivity (Esteban *et al.* 2019). The main physical factors that affect root growth, disregarding chemical and biological components, are thermal, gaseous, water, and mechanical stresses (Letey 1985; Moraes & Gusmão 2021). Combined in a domain of time and space, these soil physical stresses determine the establishment and development of roots, which affect water and nutrients availability for plants (Cherubin *et al.* 2016).

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Root elongation is the biophysical mechanism that represents the soil-root interaction (Pagès et al. 2010), which can be used to root growth modelling (Moraes et al. 2018). Despite the biophysical process of root elongation (Frene et al. 2024; Pandey & Bennett 2024) being at the frontier of knowledge (Tomobe et al. 2023; Yu et al. 2024b), only a few old studies have measured the root elongation responses to mechanical stress, for example, in soybean (Kaspar et al. 1984; Manavalan et al. 2010; Materechera et al. 1991), maize (Iijima & Kato 2007; Mirreh & Ketcheson 1973; Veen & Boone 1990), pea (Bengough et al. 1994; Iijima & Kato 2007), peanuts (Taylor & Ratliff 1969), cotton (Iijima & Kato 2007; Taylor & Ratliff 1969), and rice (Iijima & Kato 2007). However, there is no data about the responses of sugarcane roots to soil physical (mechanical, water, air or heat stress) stress, which could be used in the mechanistic root growth models such as Rootbox, CrootBox (Schnepf et al. 2018b, 2018a), or CPlantBox (Giraud et al. 2023; Zhou et al. 2020). In addition, the responses of root systems to compacted soil in the field are reproducible under controlled conditions (Colombi & Walter 2016; Giuliani et al. 2024), usually applied to the mechanistic models of root growth (Schnepf et al. 2022), whose measure and parameterization of root data are scarce in the literature (Moraes et al. 2019; 2020) and still are reason of call for collaboration in global context (Schnepf et al. 2020).

The root elongation rate is reduced by mechanical impedance (Colombi *et al.* 2018), which is accentuated when combined with water and gaseous stress (Moraes & Gusmão 2021). This root elongation process indicates the interaction between the soil and roots (Bengough 2012), which can be described by a process-based model that can be used to predict root system growth (Moraes *et al.* 2018). The process-based models, based on quantitative descriptions for understanding the relationships that govern soil–root biophysical interaction, are innovative (Moraes *et al.* 2018; Mulazzani *et al.* 2022) and still underexplored for modelling the root growth of sugarcane (Lovera *et al.* 2021). The important step to create a biophysical root growth model is the parameterization, especially the water and mechanical stress on the root elongation rate. To this end, process-based models help identify which measurements need to be carried out in laboratory experiments with both preserved (Chapman *et al.* 2012) and packed (Bai *et al.* 2019) soil structures for root growth modelling. The actual knowledge gap for sugarcane root growth modelling is due to the absence of data on root growth responses in the function of soil physical stresses.

Most field studies on the effect of soil physical restrictions on sugarcane root system (Saccharum spp.) have only quantified the total root length density (Lovera et al. 2021) and root biomass in the soil profile (Esteban et al. 2019; Cury et al. 2014), overlooking the processes and mechanisms of soil-root biophysical interaction during root elongation. Studies carried out in the laboratory using samples with reconstructed soil structure under a controlled environment (Bai et al. 2019) have contributed important information on the impact of mechanical stress on root growth (Huang et al. 2022), especially for the understanding of the process (Kong et al. 2024) and mechanisms (Tomobe et al. 2023; Pandey & Bennett 2024) involved in the soil-root interaction. Most studies commonly disregard the effect of mechanical and water stress on root diameter classes (Giuliani et al. 2024; Kumi et al. 2023), which can be distinguished by sensitivity to physical stresses in the soil (Materechera et al. 1992). Thus, the advances in root growth modelling depend on measurements of the root response as a function of mechanical and water stress, as these factors have been neglected in most agricultural crops (Seidel et al. 2022). Furthermore, parameterizations based on the relationship between soil mechanical impedance, water stress, and sugarcane root elongation allow for the establishment of mechanistic models (Moraes et al. 2018) to predict root growth in field conditions using the architectural root growth models (Schnepf et al. 2018b). The aim was to determine sugarcane root growth parameters under soil physical stress for different root diameter classes in an Oxisol from the Southeast of Brazil.



**Figure 1.** Experimental procedures for quantifying the impact of soil mechanical and water stress on sugarcane root elongation rate, including soil sampling (a), sieving (b), reconstruction into compaction levels (c), hydrostatic equilibrium in Richards chambers (d), transplanting and incubation of sugarcane seedlings in growth chambers (e), and analysis of root attributes using WinRHIZO software (f).

# Material and methods

# Experimental design

The experiment design was based on a completely randomized design with a  $5 \times 2$  factorial scheme (mechanical stress × water stress) and three replications. Pre-sprouted sugarcane seedlings were used, which were grown for 92 h in a growth chamber. The mechanical factor consisted of five compaction levels, quantified by the soil penetration resistance (MPa), and expressed by five bulk densities (1.04; 1.12; 1.19; 1.28; 1.36 Mg m<sup>-3</sup>). The water factor was expressed by two soil water matric potentials (-6 and -33 kPa), by means of hydrostatic equilibrium in Richards pressure chambers (Jacob *et al.* 2017). The soil structure was reconstructed using PVC cylinders with a height of 15 cm and an internal diameter of 5 cm.

Soil unpreserved samples were collected at 0.0-0.2 m depth (Figure 1a) from an Oxisol (Soil Survey Staff 2022) with a clayey texture (510 g kg<sup>-1</sup> clay, 330 g kg<sup>-1</sup> sand, and 160 g kg<sup>-1</sup> silt), located in Piracicaba, SP, Brazil (22°42'7.61"S, 47°37'54.60"W, 546 m a.s.l.). The soil samples were sieved with a mesh size of 2 mm, to ensure a homogeneous structure, and then stored in plastic bags (Figure 1b). To compress the soil in the PVC cylinders, soil water content was adjusted by adding water uniformly until it reached a friable consistency and corresponded to the optimum moisture for compaction (i.e. 0.32 kg<sup>-1</sup>), estimated by the pedotransfer function of Proctor test (Marcolin & Klein 2011). Afterwards, the plastic bags containing the soil samples were sealed to redistribute the water and maintain the water content throughout the soil mass.

The cylinders with different compaction levels were prepared by determining the equivalent of soil drymass as a function of the bulk density (i.e., 1.04; 1.12; 1.19; 1.28; 1.36 Mg m<sup>-3</sup>) and cylinders volume (294.5 cm<sup>3</sup>), according to the steps and procedures illustrated in Figure 1c. In order to ensure structural uniformity of the soil throughout the cylinder body, compression was carried out in three soil layers, with the surface of each layer being slit to avoid discontinuity between them.

# Soil physical analysis

Physical characterizations of the soil pore volume (i.e. macroporosity, microporosity, and total porosity) were made based on the hydrostatic equilibrium of the samples in Richards chambers

(Figure 1d). The samples, previously saturated by capillarity with water, had their mass determined and were subjected to a soil water matric potential of -6 kPa (group I), corresponding to the lower energy state of the water in the macropores (pore diameter >50 µm), according to the Laplace equation (de Jong van Lier 2020), and half of the soil samples (15) were subsequently submitted to the soil water matric potential of -33 kPa (group II). After the equilibrium at each soil water matric potential, the soil samples groups (-6 kPa and -33 kPa) were used for sugarcane root growth measurement. The volume of micropores was quantified by the volume of pores  $\leq 50$  µm (at -6 kPa). Total porosity was determined by the sum of macroporosity and microporosity.

#### Sugarcane root growth

Pre-sprouted sugarcane seedlings, 50 days old, from tillers were used to measure the root elongation rate. The developed roots were cutted, immersed in a 0.525% sodium hypochlorite (NaOCl) solution for 60 seconds, and washed in deionized water to disinfect the material (Beyerle *et al.* 1994). For emition of new nodular roots, the pre-sprounted sugarcane seedlings were placed on a double-layered filter paper (Cunha *et al.* 2021) and conditioned in growth chambers (B.O.D) for 91 h, with controlled temperature of 28 °C, relative humidity of ~70%, and photoperiod of 12 h (Melloni *et al.* 2015; Yadav *et al.* 2020), with a view to the emission of new basal roots to adapt the initial root growth to the evaluation of the cylinder.

Once the new roots had sprouted (<5 mm), transplanting was carried out by inserting each root segment into holes created in the soil 5–10 mm deep using a 3 mm diameter spiral drill (Bengough *et al.* 2016). The initial root length was measured before transplanting. After transplanting the pre-sprounted sugarcane seedlings, a 10 cm cylinder with the same diameter was positioned at the top end of the samples to deposit a soil cover with the same gravimetric water content as each sample to provide physical support for the sugarcane seedling.

The cylinders with the plants were conditioned in a growth chamber (B.O.D) for 92 h at same conditions of temperature, relative humidity, and photoperiod that initial emition of roots (Melloni *et al.* 2015; Yadav *et al.* 2020). Into the growth chamber the cilinders and pre-sprounted seedlings were placed for root growing (~92 hours) without additional irrigation or control of water content and matric potential of the soil. After removing the cylinders from the growth chamber, the soil penetration resistance was assessed until 60 mm depth, using a bench penetrometer (model CT3TM texture analyser, Brookfield Amatek<sup>®</sup>, cone 3 mm diameter, 30<sup>°</sup> angle, and penetration rate of 20 mm min<sup>-1</sup>) (Moraes *et al.* 2014b). In the same time, soil wet mass were quantified. The soil was extracted in such a way as to preserve the roots, which were separated and washed. For each cylinder a soil sample (~30g) was dried in a oven at 105°C to quantity the soil water content.

The roots, after being washed and separated from the soil, were digitalized using a flatbed scanner. The number and length of basal roots developed from each pre-sprounted sugarcane seedlingswere measured. The roots were preserved in a ethanolsolution 70%. The root elongation rate corresponded to the quotient of the averange root length by the established growth period (92 h). The root system extracted from each sample was digitalized and analysed using WinRHIZO to determine the total root length, surface area, volume, and average diameter of the roots. In addition, the root system was segmented into root diameter classes of  $\leq 0.5$  mm, 0.5–1.0 mm, and 1.0–2.0 mm (Bieluczyk *et al.* 2023), and the same growth attributes were evaluated for each class. The relative length Eq. (1) per root diameter class was determined to assess the effect of stress levels on the relative proportion of classes and is defined as follows:

$$REL = \left(\frac{RLi}{TRL}\right) \times 100 \tag{1}$$

where REL is the relative length in percentage (%), RLi is the root length for each diameter class (i) in cm, and TRL is the total root length in cm, and 100 is the conversion factor from decimal to percentage.

Bulk density	Total porosity	Macroporosity	Microporosity	Soil penetration resistance <sup>*</sup>
(Mg m <sup>-3</sup> ) <sup>*</sup>		(m <sup>3</sup> m <sup>-3</sup> )		(MPa)
1.04	0.67a	0.38a	0.29e	1.35c
1.12	0.64b	0.33b	0.31d	2.35bc
1.19	0.61c	0.28c	0.33c	3.03b
1.28	0.58d	0.22d	0.35b	4.08a
1.36	0.56e	0.18e	0.38a	4.11a
<i>p</i> -value	< 0.001	< 0.001	< 0.001	<0.001
CV (%)	2	5	5	17

Table 1. Soil physical characterization (bulk density, soil penetration resistance, and soil pore space) of packed soil samples from an Oxisol.

\*Average of soil penetration resistance measured at -6 and -33 kPa. Averages in the same column with the same letters do not differ by the Scott-Knott test (5% significance level).



Figure 2. Relationships between soil bulk density (BD) and soil penetration resistance (SPR) (a) and total porosity (Pt), macroporosity (MaP), and microporosity (MiP) (b) of packed soil samples of an Oxisol.

#### Statistical analysis of soil and root data

The parameterization consisted of the relationship between root elongation rate, other root growth attributes (i.e. total length, surface area, total volume, and average root diameter), and soil penetration resistance by means of non-linear regressions. Porous volume was related to compaction levels, expressed by bulk density. Analyses of variance were carried out to verify the independent and integrated effect of mechanical and water stress in the soil on the response variables. The means of each variable were compared (Scott-Knott test, p < 0.05) according to the soil physical stress levels. Regression analyses were developed to adjust the equations expressing the behaviour of the response variables as a function of the main factors.

#### Results

#### Changes in the pore volume and soil strength

The increase in bulk density, corresponding to the compaction levels established, reduced the soil pore space (Table 1). The increase in bulk density from 1.04 to 1.36 Mg m<sup>-3</sup> reduced total porosity from 0.67 to 0.55 m<sup>3</sup> m<sup>-3</sup> (-18%), and the same behaviour was observed for soil macroporosity (diameter > 50  $\mu$ m), which decreased from 0.38 to 0.18 m<sup>3</sup> m<sup>-3</sup> (-53%), respectively (Figure 2).

Bulk density (Mg m <sup>-3</sup> )*	Root length (cm)	Root diameter (mm)	Root volume (cm <sup>-3</sup> )	Root area (cm <sup>2</sup> )	
Soil water matric potential of -6 kPa					
1.04	109.0a	0.82ns	0.60a	28.6a	
1.12	94.5a	0.89	0.54a	25.0a	
1.19	59.6b	0.85	0.42b	17.5b	
1.28	57.6b	0.77	0.26b	13.7b	
1.36	28.0c	0.87	0.17b	7.6c	
Soil water matric potential of –33 kPa					
1.04	102.8a	0.88ns	0.64a	28.7a	
1.12	70.1a	0.89	0.44a	19.6a	
1.19	54.1b	0.93	0.35b	15.3b	
1.28	42.3b	0.94	0.30b	12.6b	
1.36	21.6c	0.94	0.15b	6.4c	
<i>p</i> -value	<0.001	0.02	< 0.001	< 0.001	
CV (%)	36	9	36	33	

Table 2. Sugarcane root growth attributes as a function of soil bulk density and soil water matric potential.

\*Average of soil penetration resistance measured at –6 and –33 kPa. ns: not significant at the 5% confidence level using the Scott-Knott test. Averages in the same column with the same letters do not differ by the Scott-Knott test (5% significance level).

The volume of soil micropores (diameter  $\leq 50 \ \mu$ m) increased from 0.29 to 0.38 m<sup>3</sup> m<sup>-3</sup> (+31%), with an increase in bulk density from 1.04 to 1.36 Mg m<sup>-3</sup>. Therefore, changing the compaction level of the soil samples alters the volume of macropores and micropores.

As a reflection of the rearrangement of the pore space, soil penetration resistance showed an exponential and positive relationship with the soil compaction level, represented by soil bulk density, although there was no effect from the matric potentials of water in the soil (-6 and -33 kPa). Soil penetration resistance varied from 1.35 to 4.11 MPa (+204.4%) for densities of 1.04 and 1.36 Mg m<sup>-3</sup>, respectively.

## Impact of soil physical stress on root growth

The changes in the levels of mechanical impediment, related to soil penetration resistance and caused by the increase in bulk density, reduced the root elongation rate of the sugarcane (Table 2). The values for length, volume, and root surface area were reduced by the higher soil mechanical impediment, in both matric potentials of -6 and -33 kPa (Figure 3). The increase in soil penetration resistance from 1.35 MPa to 4.11 MPa reduced, on average for both matric potentials, total root length from 105.9 to 24.8 cm (-76.6%) (Figure 3a), and total surface area and root volume from 28.65 to 7.0 cm<sup>2</sup> (-75.56%) and 0.62 to 0.16 cm<sup>3</sup> (-74.19%) (Figure 3b), respectively. Meanwhile, the average root diameter had no difference under mechanical stress but was altered by water stress (Table 2). The average root diameter was 0–13% greater under the matric potential of -33 kPa compared to -6 kPa.

The root elongation rate was negatively affected by the mechanical stress but was insensitive to the matric potential of the water in the soil (Figure 3d). The root elongation rate decreased by 53.4% due to the increase in soil penetration resistance from 1.35 to 4.11 MPa (Figure 3d). The number of seminal roots decreased exponentially from 11 to 5 roots (-54.5%) as a function of the variation in mechanical stress, regardless of the matric potential (Figure 3c).

#### Effect of soil physical stress on root diameter classes

Mechanical and water stress impacted the length and volume of roots (Figure 4) over diameter classes of sugarcane. In regard to root length, the diameter class of  $\leq 0.5$  mm was affected by the soil mechanical impediment and water stress (Figure 4a). The smaller-diameter roots (diameter of  $\leq 0.5$  mm) length responses to soil mechanical stress of 1.35 and 4.11 MPa under a soil water



**Figure 3.** Impact of mechanical stress, expressed by soil penetration resistance (SPR), on root growth attributes in presprouted sugarcane seedlings: (a) total root length (TRL); (b) total root volume (TRV) and total root surface area (TRA); (c) number of seminal roots (NR); and (d) root elongation rate (ER).

matric potential of -6 kPa were 1.2 to 1.9-fold greater than in the soil water matric potential of -33 kPa, which indicates the high sensitivity of this root class. In the root diameter class of 0.5–1 mm, length was reduced solely because of mechanical impediment, with a linear decrease of approximately 68.0% from 1.35 to 4.11 MPa (Figure 4b). Roots with an average diameter of 1–2 mm showed no response to either mechanical or water stress levels, with a negligible decrease in length at higher penetration resistance values. In contrast, root volume with an average diameter of  $\leq$ 0.5 mm and 0.5–1 mm was reduced by 28.6 and 80.65%, respectively, due to mechanical stress in the soil (Figure 4d). The 1–2 mm classes did not differ between levels of physical stress in the soil.

Changes were observed in the relative proportion of the root diameter classes as a function of the total physical stresses (Figure 4c). Roots with an average diameter of  $\leq 0.5$  mm increased their relative length (proportion of total root length) as only a function of mechanical impediment. This class of root showed a linear increase in relative length from approximately 15.7 to 25.4% (+61.2%) as a function of the variation in soil penetration resistance from 1.35 to 4.11 MPa. Meanwhile, the relative length of roots with an average diameter of 0.5–1 mm decreased from approximately 53.7–36.51% (–32.0%) at the same mechanical stress variation. In contrast, the relative root length of roots with a diameter of 1–2 mm was not altered by the levels of physical stress in the soil.



**Figure 4.** Impact of physical stresses in the soil on the different classes of root diameter in pre-sprouted sugarcane seedlings: (a) absolute length (RL) of roots with a diameter  $\leq$ 0.5 mm as a function of matric potential and mechanical impedance; (b) absolute length (RL) of roots with a diameter of 0.5–1 mm and 1–2 mm; (c) relative length (REL) of roots with a diameter  $\leq$ 0.5, 0.5–1 and 1–2 mm as a function of soil penetration resistance (SPR); and (d) absolute volume (REL) of roots with a diameter of  $\leq$ 0.5, 0.5–1 mm and 1–2 mm.

# Discussion

#### Changes in soil pore space and mechanical impediment

Soil structure is expressed by the arrangement of the solid fraction and especially the architecture of its pore space (Rabot *et al.* 2018). This means that changes in soil bulk density of natural occurrence, such as densification or caused by compression during agricultural operations (i.e. compaction) (Bareta Junior *et al.* 2022), affect the volume of pores in the soil. Our experimental model indicated that increasing compaction level, as expressed by bulk density, resulted in an exponential 19% reduction in total soil porosity in the 1.04–1.36 Mg m<sup>-3</sup> range., in a test with packed soil structure under three soil bulk densities using volumetric cylinders, reported a decrease of 18 and 70% in total porosity and macroporosity, respectively, which also corroborates the high reduction (53%) in macropores volume observed in our study.

The volume of macropores showed greater sensitivity to increasing soil compaction levels, as observed by Martínez *et al.* (2024) when studying the immediate impact of soil compression due to machine traffic in agricultural areas. Larger diameter pores are attributed to important gas

diffusion processes (Jin *et al.* 2023), crucial participation in water movement (Jarvis *et al.* 2024), and providing important pathways for root growth, notably under greater mechanical stress (Xiong *et al.* 2022). Therefore, a reduction in the volume of macropores alters the properties such as permeability (i.e. water infiltration rate) (Neto *et al.* 2023) and physical processes in the soil, especially root growth.

Contrary to larger-diameter pores, the volume of soil occupied by micropores increased. This increase, also observed by Silva *et al.* (2023) in a long-term experiment, can be explained by the rearrangement of particles in response to the pressure applied to the soil. The absolute volume of micropores, as well as macropores, decreased with increasing soil compaction levels, although there was an increase in their relative volume, that is, in proportion to total porosity. Increases in the volume of smaller-diameter pores provide greater available water content for plants in the soil (Viana *et al.* 2023) at the same matric potential. However, it can also lead to a decrease in the rate of gas diffusion, particularly under high matric potential, which causes gaseous stress on root growth (Moraes & Gusmão 2021). Thus, in addition to changes in the total pore volume of the soil, it is necessary to assess the effects on different pore-size classes, as these are associated with various soil processes and properties that directly or indirectly affect root development.

Soil penetration resistance increased more than 3-fold, reaching 4.11 MPa in response to the increase in bulk density, with the rearrangement of particles and consequent reduction of pore space. The increase in soil penetration resistance reflects the effect of pressure applied to the soil during intense tillage and machine traffic in areas under conventional sugarcane cultivation (Tweddle *et al.* 2021), especially when the pressure applied exceeds the soil load-bearing capacity (Toledo *et al.* 2021).

Soil compaction is a frequent problem in sugarcane crops due to the large biomass involved in the sugarcane harvesting process, which on average exceeds 75 Mg ha<sup>-1</sup>, in addition to the weight of the machines, normally above 10 Mg. In sugarcane fields, soil penetration resistance from 3.97 to 4.21 MPa was reported after four sugarcane harvests (Esteban et al. 2019) and 3.6 MPa after six harvests (Jimenez et al. 2021). Those differences between studies are due to variations in soil texture, structure, and water content at the time of measurement. Soil penetration resistance, measured with penetrometers, can be used as an indicator of physical limitations for root growth in agricultural crops (Moraes et al. 2014a). Soil penetration resistance values of 2 MPa (conventional system), 3 MPa (minimum tillage system), and 3.5 MPa (no-tillage system) have been reported as limiting values for root growth in grain-producing crops (Moraes et al. 2014a). However, critical soil penetration resistance limits for sugarcane roots are scarce in the literature. In some studies, values of 2 MPa have been suggested (Resende et al. 2023), but these have been refuted in other studies (Barbosa et al. 2018), as these values also depend on soil texture and vary from 1.5 MPa to 2.5 MPa. In general, it is known that soil structure and plant species (Bengough et al. 2011; Pott et al. 2023) influence the critical limits of soil penetration resistance for root growth. Therefore, calibration of these values is necessary for different edaphoclimatic conditions (Moraes et al. 2018).

In this study, we observed that sugarcane root growth was highly affected by soil compaction, as measured by soil penetration resistance, indicating that higher compaction levels correspond to environments with greater physical stress on sugarcane root growth. This root-soil interaction may be valid for the proposed model, considering that the soil structure was packed from soil sieved through 2 mm mesh, followed by compression. However, in soils with preserved structure under conservationist tillage systems, the presence of continuous and interconnected macropores (notably biopores) can mitigate the negative impact of mechanical restrictions on root growth (Moraes *et al.* 2016), which highlights the need to investigate the relationship between root growth and pore-related variables (e.g. pore-size distribution, pore continuity and interconnectivity, and the nature of large pores – whether they are biopores or interaggregate pores) (Xiong *et al.*, 2022).

## Impact of soil physical stress on root growth

Our experimental model for parameterizing sugarcane root growth attributes was sensitive to changes in soil physical stress. Except for root diameter and the root length with a diameter class  $\leq$ 0.5 mm, the other attributes were not affected by the water factor, but the impact of mechanical stress was evident. This is due to the narrow range of soil water potentials, which, combined with a small fraction of pores with diameters corresponding to the -6 and -33 kPa potential range, resulted in little change in soil water content between the two levels.

The length, surface area, and volume of a plant root system are associated with its ability to explore water and nutrients in the soil, but they are negatively affected by physical stresses in the soil, notably mechanical impedance. In areas under sugarcane cultivation with different tillage systems, Oliveira *et al.* (2022) observed a reduction in root surface area in response to increased soil compaction levels. Yu *et al.* (2024a), in a more detailed study on the impacts of compaction on some wheat root growth attributes during its early developmental stage, demonstrated a systemic reduction in root volume, root surface area, and root length as soil bulk density increased. Similarly, sugarcane roots exhibited high sensitivity to mechanical soil impedance, with a reduction in root emission and growth.

The reduction in the root elongation rate, commonly measured in studies of soil-root biophysical interactions (Bengough *et al.* 2011; Moraes *et al.* 2019a), is explained by the greater growth pressure required to overcome the impedance imposed by the soil (Ogilvie *et al.* 2021), resulting in a lower rate of cell division and axial expansion of the root system. The maximum root elongation rate occurred under low compaction levels and, consequently, minimal soil penetration resistance, corresponding to a rate close to  $3.5 \text{ cm d}^{-1}$ . This value is consistent with Glover (1967), who observed an average maximum root elongation rate of 2.8 cm day<sup>-1</sup> for roots originating from tillers in clayey soils with low mechanical impedance. Under the maximum mechanical restriction established in the study, that is, at 5 MPa, the root elongation rate decreased to  $1.35 \text{ cm day}^{-1}$  (-59%). Materechera *et al.* (1992), studying root growth of different agronomic species under mechanical impedance, reported a reduction between 88 and 97% in root elongation rate, accompanied by an increase in root diameter. This behaviour was not observed in this study for sugarcane roots, indicating a greater tolerance to higher mechanical stress levels in the soil during growth compared to other crops.

#### Effect of soil physical stress on root diameter classes

Regarding the different root diameter classes, the impact and intensity of physical soil stresses did not occur uniformly across each class. The effect of the moisture factor on the length of finer roots ( $\leq$ 0.5 mm) may be associated with a decrease in soil water uptake by these roots. This, although on a very small scale, was enough to affect the development of these roots in the soil matrix, especially considering their smaller diameter and, consequently, lower growth pressure (Materechera *et al.* 1992). However, in relative terms, the root length with a diameter  $\leq$ 0.5 mm increased, whereas a diameter of 0.5–1 mm reduced relative length. This highlights an adaptive strategy to aid the growth of finer roots in soils with higher compaction levels, as observed in soil bulk densities greater than 1.28 Mg m<sup>-3</sup>, since these finer roots contribute to the exploration of pores with smaller diameters (Clark *et al.* 2003).

The greater root radial development in conditions of higher resistance is a physiological and morphological response also reported in other studies with different crops (Colombi *et al.* 2018; Correa *et al.* 2019). However, our model was not sufficiently sensitive to identify the effect of soil physical stress on roots with larger diameters (1-2 mm), despite these roots showing a notable increase in relative length under conditions of higher soil penetration resistance. Moreover, the fact that coarser roots (1-2 mm) are not affected by compaction levels is supported by the greater growth pressure they exert on the soil (Materechera *et al.* 1992). Meanwhile, roots with smaller

diameters (0.5–1 mm) were the most sensitive, being strongly affected by mechanical stress. This is due to the reduction in pore space, particularly macropores, caused by soil compaction, which leads to an increased impediment to the growth of these roots.

Mechanistic models that explain the biophysical interaction between soil and roots have been studied and proposed, notably for conditions of physical stress on root system growth (Bengough *et al.* 2016; Moraes *et al.* 2019a; Pandey & Bennett 2024), but without greater detail on the behaviour of different root classes, even less for sugarcane. Our experimental model for sugarcane was able to identify changes in growth attributes and their variation among roots with different diameters in response to soil physical stress levels. This contributes to the parameterization of the impacts of these stresses for root growth modelling (Schnepf *et al.* 2020), as well as to a better understanding of biophysical processes and mechanisms of interactions with sugarcane crops.

# Conclusion

Sugarcane root elongation was highly affected by soil physical stress, particularly by mechanical impedance. The sugarcane root class of 0.5–1 mm was the most impacted by the mechanical stress in the Oxisol. The soil physical limitations due to mechanical stress had less impact on the root growth of sugarcane in the 1–2 mm diameter class compared to roots in diameter classes smaller than 1 mm.

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