The modulation of sugarcane growth and nutritional profile under aluminum stress is dependent on beneficial endophytic bacteria and plantlet origin

Godoy Labanca, Elaine Regina; Andrade, S.A.L.; Kuramae, Eiko; da Silveira, A.P.D.

published in
Applied Soil Ecology
2020

DOI (link to publisher)
10.1016/j.apsoil.2020.103715

document version
Publisher's PDF, also known as Version of record

Applied Soil Ecology 156 (2020) 103715

Contents lists available at ScienceDirect

Applied Soil Ecology

journal homepage: www.elsevier.com/locate/apsoil

The modulation of sugarcane growth and nutritional profile under aluminum stress is dependent on beneficial endophytic bacteria and plantlet origin

E.R.G. Labanca[a], S.A.L. Andrade[b], E.E. Kuramae[c,d], A.P.D. Silveira[a,*]

[a] Center of Soil and Environmental Resources, Agronomic Institute (IAC), Campinas, Sao Paulo, Brazil
[b] Department of Plant Biology, Institute of Biology, University of Campinas (UNICAMP), Campinas, São Paulo, Brazil
[c] Department of Microbial Ecology, The Netherlands Institute of Ecology, Wageningen, Netherlands
[d] Ecology and Biodiversity, Institute of Environmental Biology, Utrecht University, Utrecht, The Netherlands

ARTICLE INFO

Keywords:
Plant growth-promoting bacteria
Micropropagated plantlet
One-bud stalk plantlet
Abiotic stress
Acid soil
Plant nutrition

ABSTRACT

Plant growth-promoting bacteria (PGPB) are claimed to not only improve plant fitness but also alleviate plant stress. In this study, we evaluated the effect of five PGPB strains on plantlet growth and nutrient and aluminum (Al) uptake under acid soil conditions characterized by low P and K nutrient availability and high metal and aluminum (Al) bioavailability, which may represent a stress condition for crop plants. The PGPB strains were inoculated in sugarcane plantlets produced by meristem tissue culture (MCPs) or one-bud stalks (O-BSPs) and cultivated in soil at 37% Al saturation and pH 4.0. Biomass accumulation and Al and nutrient content in roots and shoots were determined after 30 days of growth. Bacterial inoculation increased root and shoot biomass. However, the magnitudes of these increases were dependent on bacterial strain and plantlet origin. The inoculated plantlets exhibited increased Al content and shifts in Al allocation and calcium (Ca) and boron (B) content among different plant parts (root or shoot), and these changes also depended on plantlet origin and the inoculated strain. The higher Ca uptake of inoculated MCPs and higher B uptake of inoculated O-BSPs may have contributed to reducing the damage caused by excessive Al content. The beneficial microbes also caused changes in plant uptake of micronutrients and slightly reduced macronutrient content. *Pseudomonas fluorescens* (IAC/BECa 141), *Kosakonia radicincitans* (IAC/BECa 95), *Paraburkholderia tropica* (IAC/BECa 135) and *Herbaspirillum frisingense* (IAC/BECa 152) showed potential for alleviating Al stress in sugarcane plantlets.

1. Introduction

Globally, Brazil is the largest producer of sugarcane, which is one of the country's most important commodities. Land in Brazil is dominated by intense weathered soil characterized by high acidity, low macronutrients, and high metal and aluminum (Al) bioavailability, which often represents a stress condition for crop plants. Sugarcane fields are planted with plantlets produced from mature stalk portions (with 1 to 3 buds) or meristem tissue culture; plantlets from these sources differ anatomically and microbiologically in the first stages of development but have the same life cycle (Chandra et al., 2010; Lal et al., 2015; Usman, 2015). Given these conditions and practices, continuous research on new technologies is necessary to improve yield and reduce cost and environmental impact.

Soil nutrient deficits suppress and delay growth, maturity and yield, resulting in stunted plants with reduced photosynthetic area and sugar storage (Guo et al., 2018). Toxic levels of metals such as aluminum (Al) and manganese (Mn) are often present in acid soils and can exacerbate nutritional imbalances, interfere with various physiological processes, disrupt structures such as chromosomes and lead to oxidative damage. The primary effect of Al in plants is apoplastic. The trivalent cation (Al^{3+}) forms very stable complexes with a wide range of biomolecules including proteins, nucleic acids, phenolics and pectin, impairing many plant processes and structures (Horst et al., 2010; Rao et al., 2016). In particular, the binding of Al^{3+} to cell wall pectin results in physical and chemical changes that disrupt cell wall structure and, consequently, membrane-related functions (Yang et al., 2017). Consequently, Al^{3+} is the major limitation on plant growth in acid soils (Singh et al., 2017). To overcome the limitations of acid soil with high Al saturation, considerable amounts of lime and fertilizers, especially nitrogen, must be added before installing a sugarcane plantation.

Plant growth-promoting bacteria (PGPB) are claimed as an effective

* Corresponding author.
E-mail address: apdsil@iac.sp.gov.br (A.P.D. Silveira).

https://doi.org/10.1016/j.apsoil.2020.103715
Received 28 January 2020; Received in revised form 17 June 2020; Accepted 12 July 2020

eco-friendly strategy to reduce costs, mitigate stress and improve the nutritional status of different species of plants (Majeed et al., 2018). PGPB inoculation may not only promote nutrient uptake under nutritional limitation conditions but also restrict the uptake and transport of toxic elements, thereby mitigating the stress caused by metal contamination (Ma et al., 2019; Sing et al., 2019). The effects of beneficial bacteria on plants under metal stress are well-documented. In general, PGPB enhance growth, stimulate the expression of genes related to tissue protection, exclusion and detoxification of Al, and reduce Al uptake (Panhwar et al., 2015; Sukweenadhi et al., 2015; Zerrouk et al., 2016; Farh et al., 2017) However, the role of PGPB in excess Al stress mitigation and plant nutrition has not been investigated, especially for sugarcane crops in tropical soil.

The mechanism by which beneficial bacteria mitigate stress is indirect and a consequence of plant hormonal signaling (Paungfoo-Lonhienne et al., 2016; J. Liu et al., 2017; S. Liu et al., 2017). By contrast, beneficial bacteria directly promote growth by fixing nitrogen and producing compounds that improve Fe and P uptake, such as siderophores and phosphate solubilizers, and phytohormone-related compounds that stimulate root growth, which enhances soil exploration for water and nutrients (Compant et al., 2019; Nascente et al., 2019). Moreover, PGPB inoculation can improve the nutritional state of plants and photosynthesis, enabling lower plant transpiration and increased water-use efficiency and biomass accumulation even under nutrient limitation (Dakora et al., 2015; Silveira et al., 2018; Rampazzo et al., 2018). Sorghum, lettuce, wheat, chrysanthemum, tomato, soybean and sugarcane are among the commercial crops that can benefit from bacterial inoculation (Silveira et al., 2016; Cipriano et al., 2016; Schlemper et al., 2017; Cipriano and Freitas, 2018; Orozco-Mosqueda et al., 2019/; Moretti et al., 2020).

Bacteria isolated from sugarcane roots in Brazilian plantations, including *Pseudomonas fluorescens* (IAC/BECa 141), *Kosakonia radicincitans* (IAC/BECa 95), *Paraburkholderia tropica* (IAC/BECa 135), *Herbaspirillum frisingense* (IAC/BECa 152) and *Paraburkholderia caribensis* (IAC/BECa 88), have been shown to stimulate growth, photosynthesis and nutrient uptake in micropropagated and stalk propagated plants under regular fertilization and N starvation (Silveira et al., 2018). Some of these strains produce indoleacetic acid, siderophores, and hydrogen cyanide and are antagonists to sugarcane pathogens (Marcos et al., 2016; Schlemper et al., 2018; Silveira et al., 2018; Silveira et al., 2019). However, the plant response to a strain may vary depending on the plant species/cultivar, soil physicochemical characteristics, and other biotic and abiotic factors.

Based on these features, we hypothesized that (1) these five strains are able to stimulate sugarcane growth and nutrient uptake and decrease Al uptake in an acid soil with high Al and metal availability and low nutrient supply and (2) the effect of bacterial inoculation is more intense for micropropagated plants than for stalk-propagated plants.

2. Material and methods

2.1. Soil conditions, bacterial strains and inoculum production

Soil was collected from a depth of 0–20 cm within an Oxisol at the Experimental Center of the Agronomic Institute (IAC), Campinas, Brazil. The soil chemical composition determined according to van Raij et al. (2001) was as follows: pH 4.0, organic matter 16 g dm^{-3}, P 5 mmol dm^{-3}, Ca 8 mmol dm^{-3}, Mg 3 mmol dm^{-3}, K 1 mmol dm^{-3}, Al 7 mmol dm^{-3} (m% 37), H + Al 41 mmol dm^{-3}, B 0.18 mg dm^{-3}, Cu 1.7 mg dm^{-3}, Fe 36 mg dm^{-3}, Mn 1.1 mg dm^{-3} and Zn 2.6 mg dm^{-3}.

Five bacterial strains from the IAC Beneficial Microorganisms Collection were used in this study: *Paraburkholderia caribensis* (IAC/BECa 088) (BC), *Kosakonia radicincitans* (IAC/BECa 95) (KR), *Paraburkholderia tropica* (IAC/BECa 135) (PBT), *Herbaspirillum frisingense* (IAC/BECa 152) (HF) and *Pseudomonas fluorescens* (IAC/BECa141) (PF). The five bacterial strains were previously characterized for plant growth promotion traits: antagonism to phytopathogenic fungus (PBT, PF, HF), indole-3-acetic acid (IAA) production (PF, HF), siderophore production (HF), hydrogen cyanide production (PF) and PCR amplification of nifH gene (BC, KR) (Marcos et al., 2016; Schlemper et al., 2018; Silveira et al., 2018). Each bacterial strain was plated in PDA (potato, dextrose, agar) medium and incubated for 60 h at 28 °C in the dark. A single colony was transferred to 5 mL of DYGS medium (2 g of glucose, 2 g of malic acid, 2 g of yeast extract, 0.5 g of K_2HPO_4, 0.5 g of $MgSO_4$, 1.5 g of glutamic acid, and 5 g of peptone in 1 L of water, pH 6) and incubated at 28 °C at 150 rpm. After 24 h, 100 µL was transferred to an Erlenmeyer flask containing 100 mL of the same medium and incubated under the same conditions. Bacterial cells in stationary growth phase were harvested by centrifugation at 4000 ×*g* for 10 min. The pellet was suspended in 10 mM $MgSO_4$, and the titer was adjusted spectrophotometrically (540 nm) to 10^8 CFU mL^{-1}. The control treatment was inoculated with sterile 10 mM $MgSO_4$.

2.2. Plantlet production, inoculation and pot cultivation

Sugarcane (cultivar IAC-SP 95-5000) plantlets produced from meristem tissue culture (MCPs) and one-bud stalk (O-BSPs) were provided by the IAC Sugarcane Center. One-bud stalks were planted in 400-mL pots containing sterilized commercial substrate (Plantmax®, composed of peat, *Pinus* bark and vermiculite) and inoculated with PGPB strains at planting time, 13 days after planting (DAP), and 27 DAP using 1, 2 and 10 mL of inoculum, respectively. The plantlets were grown in a greenhouse for 30 days under a shade screen (50%). Three days after the last inoculation, plantlets harboring two to three fully expanded leaves were transplanted to 2.5-L pots containing acid soil (pH 4.0) with 37% Al saturation and grown in a shadehouse for 30 days (Fig. 1).

MCP clumps were divided and transferred to 350-mL vials containing 15 mL of Murashige and Skoog (Sigma M5519) sterile basal medium supplemented with 20% (*w/v*) glucose and 100 µL of individual bacterium inoculum. The vials were housed in a BOD incubator (26 °C, 14 h light) for seven days, and then a second inoculation was performed (3 mL of inoculum, 1.5 h, 36 °C). The clumps were again divided, planted in 200-mL pots containing sterilized substrate, and housed in a growth chamber at 28 °C for 11 days. The plantlets were then transferred to a greenhouse with a shade screen (50%). After eight days, individual plantlets were transplanted to 400-mL pots containing sterilized substrate and received 2 mL of inoculum. Upon reaching the stage of 2 to 3 fully expanded leaves, the plantlets received 10 mL of inoculum. Three days later, they were transplanted into 2.5-L pots containing acidic soil (pH 4.0) and housed in a shadehouse for 30 days (Fig. 1).

The plantlets were watered with sterilized water until transplanted into soil. The controls received a corresponding volume of 10 mM sterilized $MgSO_4$ solution instead of inoculum.

2.3. Plant harvest and processing for nutrient quantification

Plants with 5 to 6 fully expanded leaves were harvested 30 days after transplanting (30 DAT). Soil attached to the plants was washed off with running water, and the roots and shoots were rinsed with distilled water. The shoots and roots were separated, dried at 60 °C until achieving a constant weight, and weighed. Roots and shoots were ground in a knife-mill (Tecnal TE-650/1), digested in HNO_3–$HClO_4$ (van Raij et al., 2001) and analyzed for concentrations of P, K, Ca, Mg, S, Fe, Mn, Cu, Zn, B and Al by inductively coupled plasma optical emission spectrometry (ICP-OES) at the IAC Soil Analysis Laboratory. Total N was determined using Kjeldahl analysis after sulfuric acid digestion of plant samples.

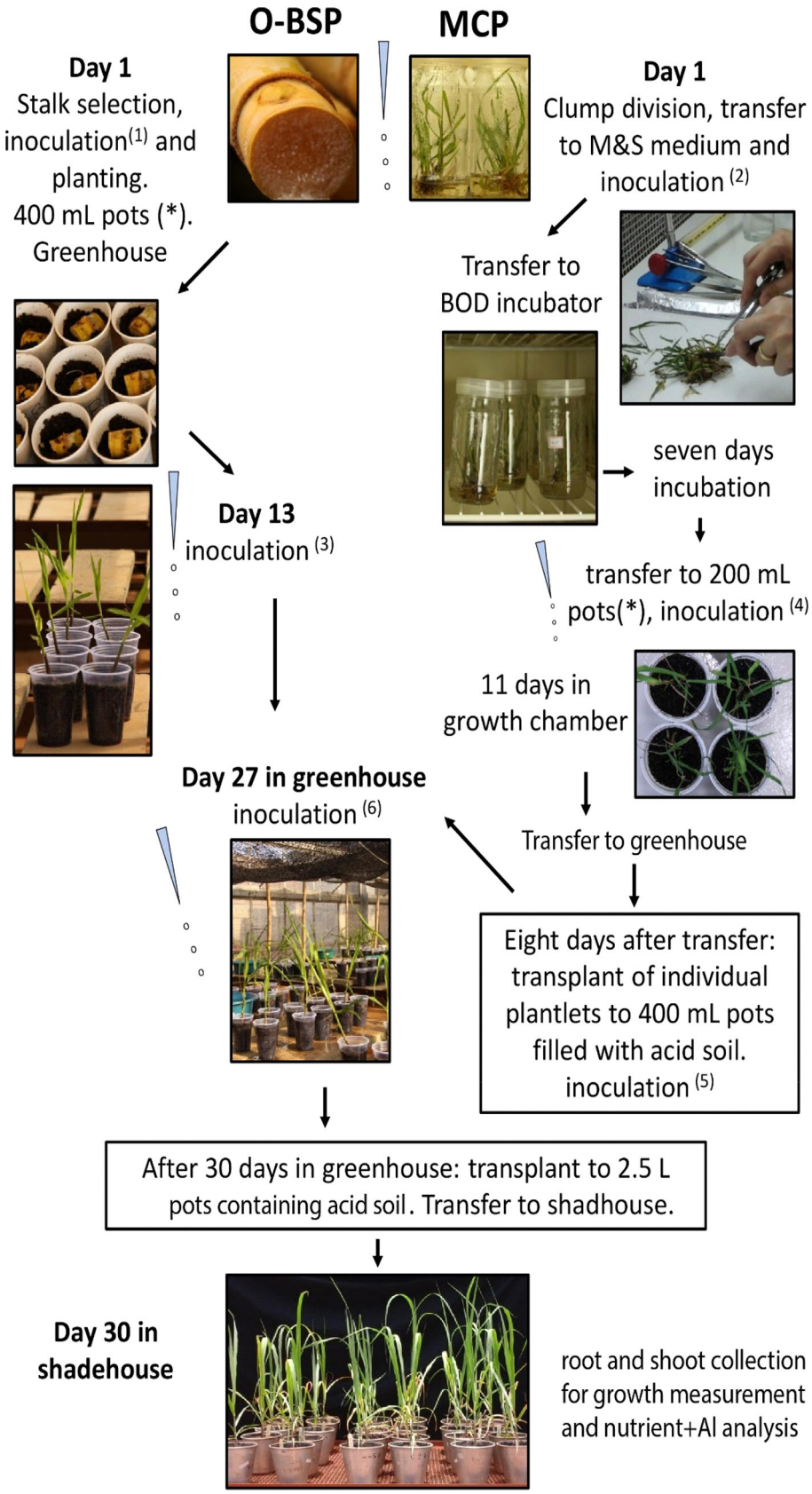

Fig. 1. Diagaram of experimental steps. O-BSP: one-bud stalk plantlet; MCP: meristem culture plantlet; (*) sterilized substrate. Plant inoculation was performed with 1 mL (1); 100 µL (2); 2 mL (3); 3 mL (4); 2 mL (5); and 10 mL (6) of bacterial cell suspension adjusted to 10^8 CFU mL^{-1}.

Table 1
Total biomass (shoot and root) of plantlets after 30 days of Al stress.

	O-BSP (g plant^{-1})	MCP (g plant^{-1})
T	7.01 ± 0.16 a A	4.75 ± 0.20 b B
BC	5.61 ± 0.19 b A	5.05 ± 0.27 b A
KR	7.82 ± 0.13 a A	6.70 ± 0.14 a B
PF	7.52 ± 0.57 a A	5.80 ± 0.43 b B
PBT	7.75 ± 0.22 a A	5.27 ± 0.42 b B
HF	7.67 ± 0.05 a A	6.86 ± 0.26 a B

T: control; BC: *Paraburkholderia caribensis*; KP: *Kosakonia radicincitans*; PF: *Pseudomonas fluorescens*; PBT: *Paraburkholderia tropica*; HF: *Herbaspirillum frisingense*. O-BSP: one-bud stalk plantlet; MCP: meristem culture plantlet. Same letters (capital letters in rows and lowercase letters in columns) are not significantly different according to the Scott-Knott test ($p < 0.05$).

2.4. Statistical analysis

The experiment was performed using a completely randomized factorial design considering bacterial treatment and plantlet origin as independent factors. For each treatment, five replicates were used for biomass quantification, and four replicates were used for Al and nutrient content. The data were analyzed using two-way ANOVA. Mean significant differences were compared by the Scott-Knott test at a significance level of 5% using SISVAR software (Ferreira, 2011). Principal component analysis of data was performed using PAST software (Hammer et al., 2001).

3. Results

3.1. Plant growth

Bacterial strain and plantlet origin impacted plant biomass (shoot + root). Plantlet growth promotion was observed only in MCPs treated with KR and HF (Table 1). However, when roots and shoots were assessed separately, significant increases were observed in dry root biomass for all inoculated MCPs and shoot biomass for O-BSPs inoculated with KR, PBT, HF and PF (Fig. 2A, B). Inoculated MCPs had up to 60% greater root biomass than the control (Fig. 2B). However, dry root and shoot biomass were increased only in MCPs inoculated with HF and KR (Fig. 2A). There was no effect of bacterial inoculation on O-BSP root growth. However, four of the five bacterial strains, KR, PBT, HF and PF, promoted O-BSP shoot growth by up to 30%, whereas BC reduced O-BSP growth (Fig. 2A and B).

Bacterial inoculation in MCPs promoted an increase in the root-to-shoot biomass ratio (R:S ratio), whereas this ratio was reduced in O-BSPs (Fig. 2C). The R:S ratio was 0.64 in the control regardless of plantlet origin. Plantlet origin affected biomass accumulation. In general, O-BSP shoots grew more than MCP shoots (Fig. 2A), regardless of inoculation. The root dry biomass of plantlets inoculated with BC, HF and PF did not differ significantly between MCPs and O-BSPs. However, KR-inoculated MCPs had greater root growth than KR-inoculated O-BSPs, while the opposite pattern was observed in response to PBT treatment (Fig. 2B).

3.2. Effect of bacterial strains on plantlet nutritional profile

Zinc was the only element that was not altered by inoculation of the different bacterial strains in both MCPs and O-BSPs (Figs. S1 and S2). Bacterial inoculation changed shoot Fe and N content and root Mg and Cu content (Fig. 3). The content of Al, P, K, Ca, S, B and Mn in shoots and/or roots varied depending on the inoculated strain and plantlet origin. Each strain promoted unique changes in the nutritional profile of the plantlets (Figs. 3, 4A and B) compared with the control. The profiles of O-BSPs inoculated with KR PF, PBT and HF were similar

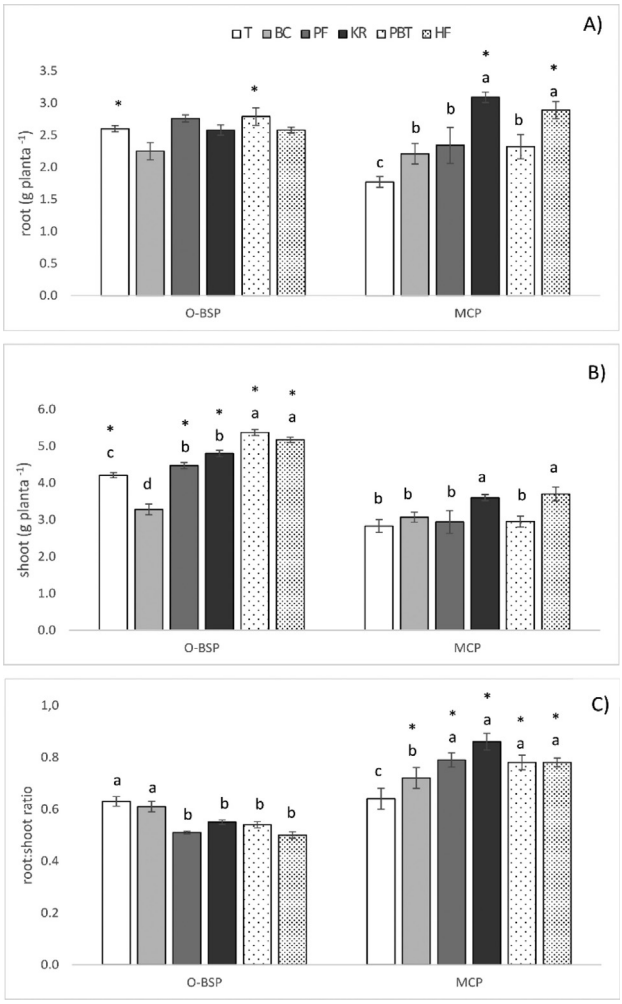

Fig. 2. Shoot (A) and root (B) growth and root-to-shoot ratio (C) 30 days after transplanting to soil with high Al content. T: control; BC: *Paraburkholderia caribensis*; KP: *Kosakonia radicincitans*; PF: *Pseudomonas fluorescens*; PBT: *Paraburkholderia tropica*; HF: *Herbaspirillum frisingense*. O-BSP: one-bud stalk plantlet; MCP: meristem culture plantlet. Columns represent mean values, and bars represent the SE of five replicates. Different letters indicate significant differences between bacterial treatments, and asterisks indicate significant differences between plantlet origin (Scott Knott test $p < 0.05$).

(Fig. 3B, C, D and E) and differed from those of the control and O-BSPs inoculated with BC (Figs. 3A, 4A).

In general, there was a reduction in macronutrient content, except Ca, in both O-BSPs and MCPs inoculated with KR, PF, PBT and HF (Fig. 3). Inoculation with BC reduced shoot B content in both O-BSPs and MCPs compared with the control (Fig. 3). HF treatment had no impact on Al content but promoted increased Ca content in the roots of O-BSPs (70%) and increased Mn content in the roots and shoots of both MCPs and O-BSPs (Fig. 3).

Distinct variations in B, Al, Fe and Ca content were observed in the inoculated plantlets compared with the control (Figs. 3, S1 and S2). In general, Al content was higher in the shoots and lower in the roots of inoculated O-BSPs compared with the control. Interestingly, the opposite pattern was observed in MCPs. The increase in Al content in O-BSPs that exhibited growth promotion was accompanied by an increase in B content. In MCPs, B content was reduced, whereas Ca content was increased. In all treatments, the changes in Al and B content were large; for example, shoot Al content was three times higher in O-BSPs treated with BC than in the control, whereas the B content decreased by half (Fig. 3).

Given the clear shifts in B and Al content in roots and shoots

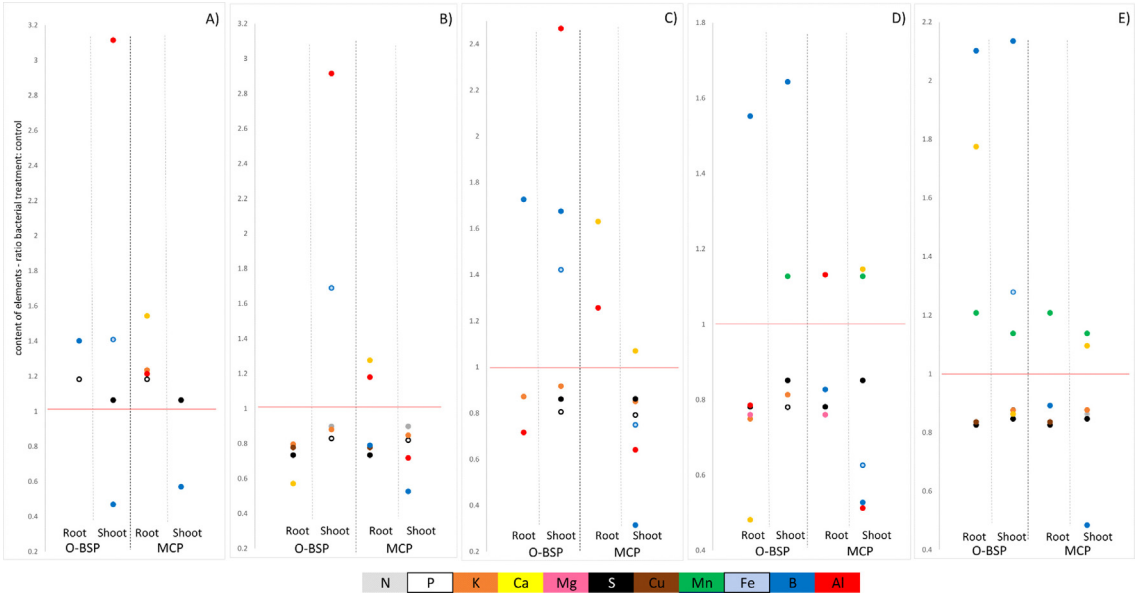

Fig. 3. Nutrient and Al content in inoculated plantlets relative to the control. A) *Paraburkholderia caribensis*; B) *Kosakonia radicincitans*; C) *Pseudomonas fluorescens*; D) *Paraburkholderia tropica* and E) *Herbaspirillum frisingense*. O-BSP: one-bud stalk plantlet; MCP: meristem culture plantlet. The plotted dots indicate whether the element content was higher or lower compared with the control (Scott-Knott test, $p < 0.05$).

promoted by the bacterial strains, we determined the ratios of total Al and B accumulation between roots and shoots (Al_R:Al_S and B_R:B_S ratios). Bacterial inoculation promoted a drastic reduction in the Al_R:Al_S ratio in O-BSPs but an increase in the Al_R:Al_S ratio in MCPs (Fig. 5A). Compared with the other treatments, BC increased the B_R:B_S ratio threefold.

The plantlet content of nutrients and Al was a function of bacterial strain and plantlet origin (Fig. 4C). The interaction between 'plantlet origin' and 'bacterial strain' is evident in Fig. 4A and B, especially for the distribution of Al in roots and shoots. In general, nutrient levels were higher in MCPs (Figs. S1 and S2), with the exception of root levels of K, B and Mn, which were higher in O-BSPs than in MCPs.

In general, the bacterial strains that promoted greater biomass also promoted higher total (root + shoot) nutrient accumulation (Fig. 6), with the exception of B and P accumulation in MCPs and Al, Fe, S and P accumulation in O-BSPs. None of the strains affected Al accumulation in O-BSPs (Fig. 6A). In MCPs, inoculation resulted in less B accumulated per plantlet (Fig. 6B).

4. Discussion

4.1. Influence of bacterial strains on biomass accumulation and partitioning between roots and shoots

In this study, we determined the effects of five different PGPB strains on biomass production and nutritional homeostasis in sugarcane plantlets of different origin but the same genetic background exposed to high soil Al saturation. Plantlet origin determined bacterial effects. O-BSPs grew better than MCPs, probably as a result of different microbiological and/or anatomical features. The MCP plantlet production system is expected to support a very limited microbiome, and when planted in soil, MCPs undergo rapid colonization by autochthonous microorganisms. The colonization process is not random, and plantlet recruitment of beneficial microbes is an energy and metabolic investment for the plant (Da Costa et al., 2014; Yan et al., 2017). Considering the reduced microbiome present in MCPs, it is possible that this energy cost, in combination with Al stress, was higher in MCPs than in O-BSPs, resulting in slower growth. The delayed growth of MCPs may also be attributable to anatomical deficiencies. The MCPs were maintained for approximately 45 days in a growth chamber and greenhouse for adaptation before transplanting to soil in order to reach the same phenological stage as the O-BSPs. MCPs require a period of hardening to prevent damage due to anatomical peculiarities, such as the high number and size of stomata and the abnormal structure of the epidermis and epicuticular wax (Chandra et al., 2010). However, it was not possible to affirm that the MCPs were anatomically and physiologically similar to the O-BSPs after the adaptation period, and therefore an influence of the plantlets' anatomical differences on growth cannot be ruled out.

Each bacterial strain promoted different growth responses depending on plantlet origin. HF promoted root and shoot growth in MCPs but only shoot growth in O-BSPs. BC reduced shoot growth in O-BSPs and stimulated root growth in MCPs. Previous studies of the same sugarcane variety and bacterial strains in the absence of stress reported root and shoot growth promotion for MCPs (Silveira et al., 2018) and O-BSPs (Silveira et al., 2019). Therefore, we conclude that some of the variability observed in PGPB performance in this study may be related to the influence of Al stress.

Inoculation of different bacterial strains in MCPs increased the root-to-shoot ratio without reducing shoot biomass. When plant stressors are present below ground, plants tend to allocate more biomass to roots, and as a consequence, the root-to-shoot ratio frequently increases because root growth occurs at the expense of shoot growth (Poorter et al., 2012). PGPB often stimulate root growth, either by the production of phytohormones or by interference with plant hormonal homeostasis, without loss of shoot growth (Paungfoo-Lonhienne et al., 2014; Straub et al., 2013). Therefore, the stimulation of root growth promoted by the five bacterial strains was probably due to modulation of the stress response allowing greater access to water and soil nutrients, leading to higher biomass production.

The Al stress mitigation effect of the PGPB strains in sugarcane plantlets was confirmed by visual symptoms of Al toxicity, which reduces root growth (Ryan and Delhaize, 2017). Increased root growth in soil with excessive Al saturation is widely associated with higher Al tolerance (Giannakoula et al., 2008; Li et al., 2018; Silva et al., 2013; Zerrouk et al., 2016).

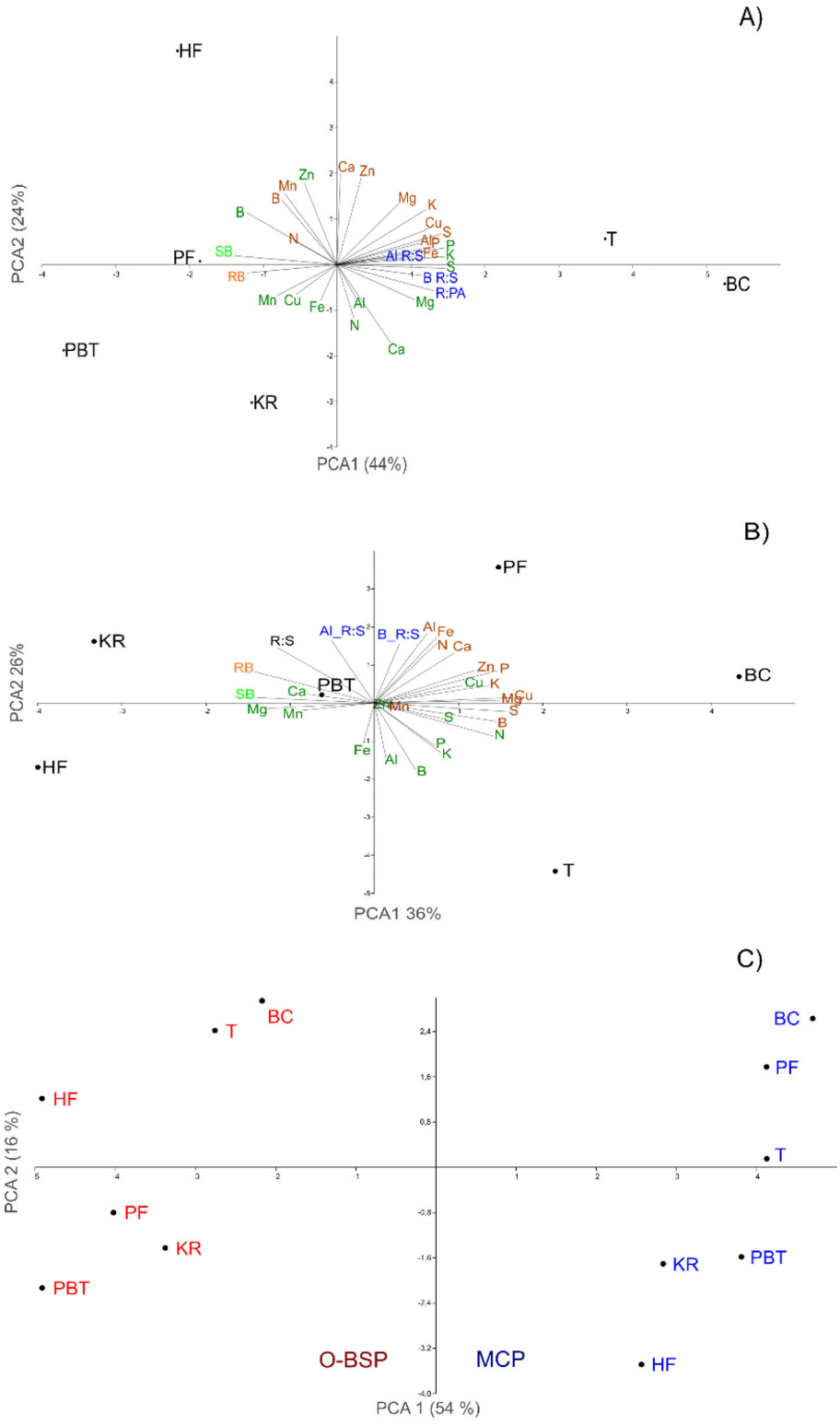

Fig. 4. Principal component analysis for (A) one-bud stalk plantlets (O-BSPs); (B) meristem tissue culture plantlets (MCPs) and (C) O-BSPs (in blue) and MCPs (in red). T: control; BC: *Paraburkholderia caribensis*; KP: *Kosakonia radicincitans*; PF: *Pseudomonas fluorescens*; PBT: *Paraburkholderia tropica*; HF: *Herbaspirillum frisingense*. O:S: root-to-shoot ratio; Al-R:S: root-to-shoot ratio of accumulated Al; B-R:S: root-to-shoot ratio of accumulated B; R:B: root biomass; SB: shoot biomass. Element contents are shown in brown for roots and green for shoots. (For interpretation of the references to color in this figure legend, the reader is referred to the web version of this article.)

4.2. Impact on nutritional status and partitioning of Al and B between roots and shoots

PGPB have been evaluated as a biological tool to improve plant fitness in metal-contaminated soils and for phytoremediation due to their ability to (1) change the distribution of metals between roots and shoots; (2) secrete compounds that can complex metals, thereby reducing metal bioavailability; and (3) absorb metals (Ma et al., 2016b, 2016a; T. Wang et al., 2017; Gupta et al., 2018).

Compared with other metals, information about the mechanisms of Al stress alleviation activated by PGPB is sparse. A few studies have reported positive effects of PGPB on the root system and the expression of genes controlling Al tolerance and exclusion from plant tissues (Sukweenadhi et al., 2015; Zerrouk et al., 2016; Farh et al., 2017). The nutritional approach used in this study allowed us to make assumptions about Al tolerance mechanisms activated by PGPB in the different sugarcane plantlets.

PGPB inoculation in sugarcane plantlets did not alter total Al

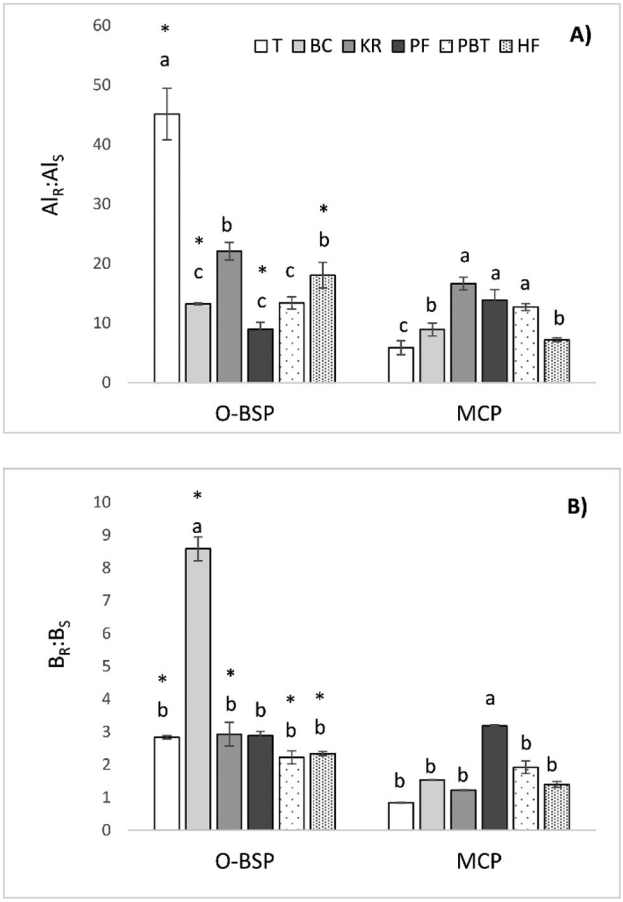

Fig. 5. Ratio of total Al (A) or B (B) accumulation between roots and shoots 30 days after transplanting to soil with high Al content. T: control; BC: *Paraburkholderia caribensis*; KP: *Kosakonia radicincitans*; PF: *Pseudomonas fluorescens*; PBT: *Paraburkholderia tropica*; HF: *Herbaspirillum frisingense*. O-BSP: one-bud stalk plantlet; MCP: meristem culture plantlet. Columns represent mean values, and bars represent the SE of four replicates. Different letters indicate significant differences between bacterial treatments, and asterisks indicate significant differences between plantlet origin (Scott-Knott test $p < 0.05$).

accumulation in O-BSPs, but total Al accumulation increased in MCPs. These results are in contrast to other reports for Poaceae crop species such as wheat, barley and sorghum, where Al tolerance has been related mainly to its exclusion from plant tissues (Kochian et al., 2015; Moustaka et al., 2016; Ramesh et al., 2015). However, in highly Al-tolerant wild Poaceae (Ezaki et al., 2013; Reyna-Llorens et al., 2015) and rice (Yokosho et al., 2016; Y. Wang et al., 2017) several internal mechanisms have been implicated in Al detoxification in response to high Al, with a minor role of Al exclusion mechanisms. Nonetheless, Maia et al. (2018) reported that lower accumulation of Al in sugarcane tissues is not always correlated with enhanced metal tolerance. Therefore, we suggest that the inoculated PGPB strains may have induced mechanisms of Al detoxification such as those found in wild Poaceae species or rice.

The changes in allocation of Al to roots and shoots promoted by the PGPB strains suggest that plantlets of sugarcane cultivar IACSP-95-5000 harbor internal mechanisms for Al detoxification in both the root and shoot compartments. In O-BSPs, inoculation resulted in an increase in Al content in shoots and a decrease in roots, whereas the opposite pattern was observed in MCPs. Thus, the effects of the bacterial strains on the distribution of toxic metals between roots and shoots (Ma et al., 2016b) seems to be dependent on plantlet origin.

Calcium, B, Al and Fe content varied most significantly between the inoculated sugarcane plantlets. The inoculated bacterial strains promoted an increase in Ca content in MCPs, whereas in O-BSPs, almost no change in Ca content was observed. Ca^{2+} bridges molecules at the membrane surface and cell walls and is crucial for the organization and functioning of the two structures (Pilbeam and Morley, 2006). Al^{3+} displaces basic cations such as Ca^{2+}, modifying wall structure and reducing Ca uptake (Postma et al., 2005; Horst et al., 2010; Rao et al., 2016; Singh et al., 2017). Some authors attribute higher Al tolerance to higher Ca and Mg uptake capacity and the maintenance of higher levels of these elements in both the roots and shoots of plants (Giannakoula et al., 2008; Moustaka et al., 2016). The increased Ca content in MCPs might be a plant response mediated by the bacterial strains that protects cell walls and membrane function and reduces the potential negative impact of higher Al content. Similar to Ca, the changes in B content were dependent on plantlet origin. Inoculation resulted in enhanced B content in O-BSPs and but reduced levels in MCPs. The increase in B content mediated by PGPB in O-BSPs may be related to stress relief. In adequate concentrations, B can reverse Al damage by triggering alkalization of the root zone (Li et al., 2018), modulating the antioxidant defense system (Riaz et al., 2018) or increasing Al immobilization by wall pectin in border cells (Li et al., 2017). While B content increased in O-BSPs, its distribution between roots and shoots ($B_R:B_S$) was dependent on bacterial strain. Compared with control plants, the $B_R:B_S$ ratio was approximately 2.6 times higher in O-BSPs inoculated with PF, KR, PBT or HF. By contrast, BC interfered with plant performance and caused growth depletion, with an increase in the $B_R:B_S$ ratio of approximately threefold compared with the control and plants inoculated with the other strains.

Inoculation also altered Fe and Mn content in O-BSPs. However, it is difficult to determine whether these modifications were a direct effect

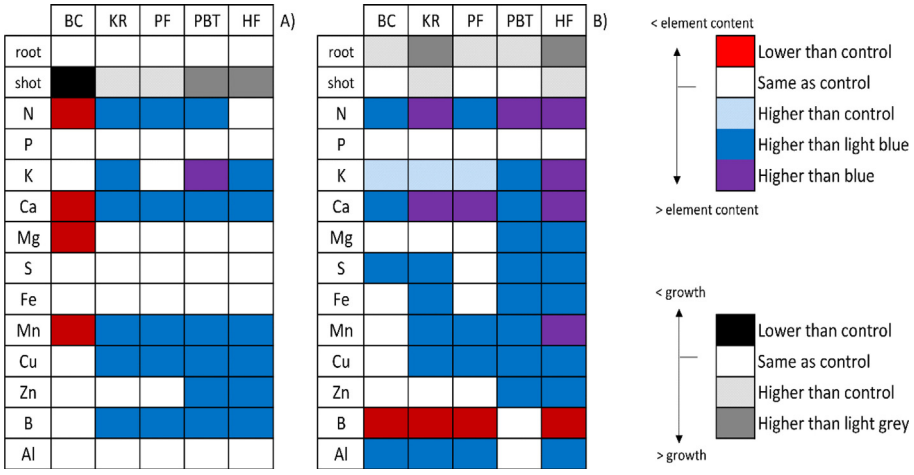

Fig. 6. Heatmap of the accumulation of each element in plantlets (root + shoot) and growth of plantlets inoculated with bacteria relative to control non-inoculated plantlets. A) One-bud stalk plantlets; B) meristem culture plantlets. BC: *Paraburkholderia caribensis*; KR: *Kosakonia radicincitans*; PF: *Pseudomonas fluorescens*; PBT: *Paraburkholderia tropica*; HF: *Herbaspirillum frisingense*. Different colors on the same line indicate a significant difference between treatments (Scott-Knott test, $p < 0.05$).

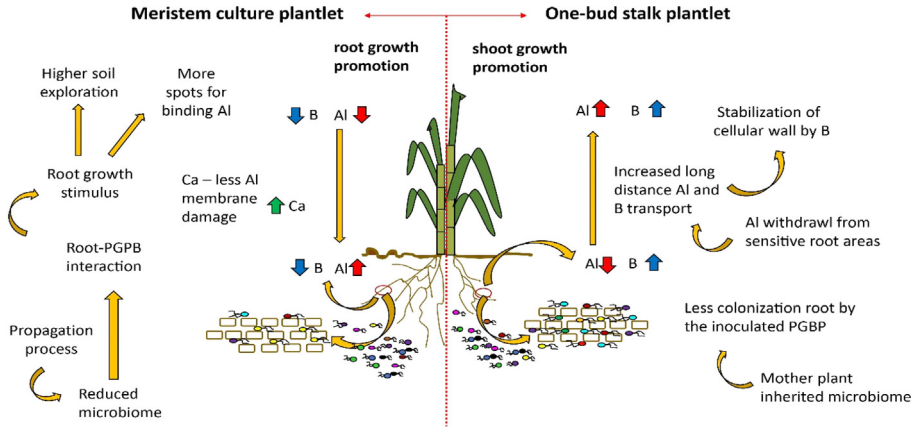

Fig. 7. Conceptual framework of the results of this study.

of the bacterial strains on Fe and Mn uptake or a side effect of altered uptake of other metallic micronutrients or Al. Metals are usually chelated by organic compounds in the cellular environment for compartmentalization and/or for long-distance transport via the xylem stream (Peng and Gong, 2014). In addition, different metals can share the same membrane transporters for short-distance transport and compartmentalization. Disruption of metal uptake or distribution by excessive Al content can cause shifts in the homeostasis of nutrients such as Fe and Mn (Bityutskii et al., 2017; Liu et al., 2017a).

Sugarcane plantlets inoculated with the studied bacterial strains did not show increases in macronutrient content except Ca; in fact, inoculation caused 10–25% depletion of macronutrients. These results are contrary to the increases in plant N and P uptake normally reported in the presence of PGPB (Martins et al., 2018; Richardson et al., 2009; Silveira et al., 2016).

PGPB traits are common guidelines for beneficial bacteria selection. Nonetheless, tests involving different plants and environmental conditions must be performed to confirm bacteria ability to promote growth or mitigate stress. Although BC and KR have nifH gene in their genomes they were not able to improve N uptake but the capacity of production of IAA (PF and HF), HCN (PF) and siderophore (HF) may have contributed to plant growth. Plant growth is stimulated by IAA (Kudoyarova et al., 2017) and both HCN and siderophore are able to form non-toxic complex with metals (Chen et al., 2017; Faramarzi et al., 2020; Rijavec and Lapanje, 2016) and may have mitigated metal stress. Fig. 7 illustrates a conceptual framework summarizing the results of this study, including the potential roles of PF (IAC/BECa141), KR (IAC/BECa 95), PBT (IAC/BECa 135) and (IAC/BECa 152) HF in the alleviation of Al stress in sugarcane plantlets and some possible explanations.

5. Conclusions

Of the five PGPB strains previously identified as promoting sugarcane growth in the absence of stress, IAC/BECa 135 (*Paraburkholderia tropica*), IAC/BECa 152 (*Herbaspirillum frisingense*), IAC/BECa141 (*Pseudomonas fluorescens*) and IAC/BECa 95 (*Kosakonia radicincitans*) were able to enhance plantlet fitness under nutritional limitation conditions and Al stress by modulating nutritional and Al homeostasis. The effects of bacterial inoculation in the presence of excessive Al were dependent on the interaction between the bacterial strain and sugarcane plantlet origin.

Declaration of competing interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

Acknowledgments

The first author was financially supported by CAPES foundation. This research was supported by CNPq/NWO Program (Project Number 456420/2013-4) and NWO (NWO-729.004.013) grants. Publication number 7011 of The Netherlands Institute of Ecology (NIOO-KNAW). The authors thank Dr. Silvana Creste de Souza and Dr. Mauro Xavier for providing the plant material, from Sugarcane Center (IAC).

Appendix A. Supplementary data

Supplementary data to this article can be found online at https://doi.org/10.1016/j.apsoil.2020.103715.

References

Bityutskii, N.P., Yakkonen, K.L., Petrova, A.I., Shavarda, A.L., 2017. Interactions between aluminium, iron and silicon in *Cucumber sativus* L. grown under acidic conditions. J. Plant Physiol. 218, 100–108. https://doi.org/10.1016/j.jplph.2017.08.003.

Chandra, S., Bandopadhyay, R., Kumar, V., Chandra, R., 2010. Acclimatization of tissue cultured plantlets: from laboratory to land. Biotechnol. Lett. 32, 1199–1205. https://doi.org/10.1007/s10529-010-0290-0.

Chen, Y., Yang, W., Chao, Y., Wang, S., Tang, Y.T., Qiu, R.L., 2017. Metal-tolerant *Enterobacter* sp. strain EG16 enhanced phytoremediation using *Hibiscus cannabinus* via siderophore-mediated plant growth promotion under metal contamination. Plant Soil 413, 203–216. https://doi.org/10.1007/s11104-016-3091-y.

Cipriano, M.A.P., Freitas, S.S., 2018. Effect of *Pseudomonas putida* on chrysanthemum growth under greenhouse and field conditions. Afr. J. Agric. Res. 13, 302–310. https://doi.org/10.5897/AJAR2017.12839.

Cipriano, M.A.P., Lupatini, M., Lopes-Santos, L., da Silva, M.J., Roesch, L.F.W., Destéfano, S.A.L., Freitas, S.S., Kuramae, E.E., 2016. Lettuce and rhizosphere microbiome responses to growth promoting *Pseudomonas* species under field conditions. FEMS Microbiol. Ecol. 92. https://doi.org/10.1093/femsec/fiw197.

Compant, S., Samad, A., Faist, H., Sessitsch, A., 2019. A review on the plant microbiome: ecology, functions, and emerging trends in microbial application. J. Adv. Res. https://doi.org/10.1016/j.jare.2019.03.004.

Da Costa, P.B., Granada, C.E., Ambrosini, A., et al., 2014. A model to explain plant growth promotion traits: a multivariate analysis of 2,211 bacterial isolates. PLoS One 9, 1–25. https://doi.org/10.1371/journal.pone.0116020.

Dakora, F.D., Matiru, V.N., Kanu, A.S., 2015. Rhizosphere ecology of lumichrome and riboflavin, two bacterial signal molecules eliciting developmental changes in plants. Front. Plant Sci. https://doi.org/10.3389/fpls.2015.00700.

Ezaki, B., Jayaram, K., Higashi, A., Takahashi, K., 2013. A combination of five mechanisms confers a high tolerance for aluminum to a wild species of Poaceae, *Andropogon virginicus* L. Environ. Exp. Bot. 93, 35–44. https://doi.org/10.1016/j.envexpbot.2013.05.002.

Faramarzi, M.A., Mogharabi-Manzari, M., Brandl, H., 2020. Bioleaching of metals from wastes and low-grade sources by HCN-forming microorganisms. Hydrometallurgy 191, 105228. https://doi.org/10.1016/j.hydromet.2019.105228.

Farh, M.E.A., Kim, Y.J., Sukweenadhi, J., et al., 2017. Aluminium resistant, plant growth promoting bacteria induce overexpression of aluminium stress related genes in *Arabidopsis thaliana* and increase the ginseng tolerance against aluminium stress. Microbiol. Res. 200, 45–52. https://doi.org/10.1016/j.micres.2017.04.004.

Ferreira, D.F., 2011. Sisvar: a computer statistical analysis system. Ciênc. Agrotecnol. 35, 1039–1042. https://doi.org/10.1590/S1413-70542011000600001.

Giannakoula, A., Moustakas, M., Mylona, P., et al., 2008. Aluminum tolerance in maize is correlated with increased levels of mineral nutrients, carbohydrates and proline, and decreased levels of lipid peroxidation and Al accumulation. J. Plant Physiol. 165, 385–396. https://doi.org/10.1016/j.jplph.2007.01.014.

Guo, P., Qi, Y.P., Cai, Y.T., Yang, T.Y., Yang, G.L.T., Huand, Z.R., Chen, L.S., 2018. Aluminum effects on photosynthesis, reactive oxygen species and methylglyoxal detoxification in two *Citrus* species differing in aluminum tolerance. Tree Physiol. 38, 1548–1565.

Gupta, P., Rani, R., Chandra, A., Kumar, V., 2018. Potential applications of *Pseudomonas* sp. (strain CPSB21) to ameliorate Cr^{6+} stress and phytoremediation of tannery effluent contaminated agricultural soils. Sci. Rep. 8, 1–10. https://doi.org/10.1038/s41598-018-23322-5.

Hammer, Ø., Harper, D.A.T., Ryan, P.D., 2001. PAST: Paleontological statistics software package for education and data analysis. http://palaeo-electronica.org/2001_1/past/issue1_01.htm.

Horst, W.J., Wang, Y., Eticha, D., 2010. The role of the root apoplast in aluminium-induced inhibition of root elongation and in aluminium resistance of plants: a review. Ann. Bot. 106, 185–197. https://doi.org/10.1093/aob/mcq053.

Kochian, L.V., Piñeros, M.A., Liu, J., Magalhaes, J.V., 2015. Plant adaptation to acid soils: the molecular basis for crop aluminum resistance. Annu. Rev. Plant Biol. 66, 571–598. https://doi.org/10.1146/annurev-arplant-043014-114822.

Kudoyarova, G.R., Vysotskaya, L.B., Arkhipova, T.N., Kuzmina, L.Y., Galimsyanova, N.F., Sidorova, L.V., Gabbasova, I.M., Melentiev, A.I., Veselov, S.Y., 2017. Effect of auxin producing and phosphate solubilizing bacteria on mobility of soil phosphorus, growth rate, and P acquisition by wheat plants. Acta Physiol. Plant. 39, 1–8. https://doi.org/10.1007/s11738-017-2556-9.

Lal, M., Tiwari, A.K., Gupta, G.N., et al., 2015. Commercial scale micropropagation of sugarcane: constraints and remedies. Sugar Tech. 17, 339–347. https://doi.org/10.1007/s12355-014-0345-y.

Li, X.W., Liu, J.Y., Fang, J., et al., 2017. Boron supply enhances aluminum tolerance in root border cells of pea (*Pisum sativum*) by interacting with cell wall pectins. Front. Plant Sci. 8, 1–11. https://doi.org/10.3389/fpls.2017.00742.

Li, X., Li, Y., Mai, J., et al., 2018. Boron alleviates aluminum toxicity by promoting root alkalization in transition zone via polar auxin transport. Plant Physiol. 177, 1254–1266. https://doi.org/10.1104/pp.18.00188.

Liu, J., Zhou, M., Delhaize, E., Ryan, P.R., 2017a. Altered expression of a malate-permeable anion channel, OsALMT4, disrupts mineral nutrition. Plant Physiol. 175, 1745–1759. https://doi.org/10.1104/pp.17.01142.

Liu, S., Hao, H., Lu, X., et al., 2017b. Transcriptome profiling of genes involved in induced systemic salt tolerance conferred by *Bacillus amyloliquefaciens* FZB42 in *Arabidopsis thaliana*. Sci. Rep. 7, 10795. https://doi.org/10.1038/s41598-017-11308-8.

Ma, Y., Oliveira, R.S., Freitas, H., Zhang, C., 2016a. Biochemical and molecular mechanisms of plant-microbe-metal interactions: relevance for phytoremediation. Front. Plant Sci. 7, 1–19. https://doi.org/10.3389/fpls.2016.00918.

Ma, Y., Rajkumar, M., Zhang, C., Freitas, H., 2016b. Inoculation of *Brassica oxyrrhina* with plant growth promoting bacteria for the improvement of heavy metal phytoremediation under drought conditions. J. Hazard. Mater. 320, 36–44. https://doi.org/10.1016/j.jhazmat.2016.08.009.

Ma, Y., Vosátka, M., Freitas, H., 2019. Beneficial microbes alleviate climatic stresses in plants. Front. Plant Sci. https://doi.org/10.3389/fpls.2019.005595.

Maia, C., Almeida, C., Costa, P., et al., 2018. Phenotypic plasticity of sugarcane genotypes under aluminum stress. J. Exp. Agric. Int. 22, 1–11. https://doi.org/10.9734/JEAI/2018/40984.

Majeed, A., Muhammad, Z., Ahmad, H., 2018. Plant growth promoting bacteria: role in soil improvement, abiotic and biotic stress management of crops. Plant Cell Rep. 37, 1599–1609. https://doi.org/10.1007/s00299-018-2341-2.

Marcos, F.C.C., Iório, R.P.F., Silveira, A.P.D., Ribeiro, R.V., Machado, E.C., Lagôa, A.M.A., 2016. Endophytic bacteria affect sugarcane physiology without changing plant growth. Bragant. 75, 1–9. https://doi.org/10.1590/1678-4499.256.

Martins, M.R., Jantalia, C.P., Reis, V.M., et al., 2018. Impact of plant growth-promoting bacteria on grain yield, protein content, and urea-15 N recovery by maize in a Cerrado Oxisol. Plant Soil 422, 239–250. https://doi.org/10.1007/s11104-017-3193-1.

Moretti, L.G., Crusciol, C.A.C., et al., 2020. Effects of growth-promoting bacteria on soybean root activity, plant development and yield. Agron. J. 112, 418–428. https://doi.org/10.1002/agj2.20010.

Moustaka, J., Ouzounidou, G., Bayçu, G., Moustakas, M., 2016. Aluminum resistance in wheat involves maintenance of leaf Ca^{2+} and Mg^{2+} content, decreased lipid peroxidation and Al accumulation, and low photosystem II excitation pressure. BioMetals 29, 611–623. https://doi.org/10.1007/s10534-016-9938-0.

Nascente, A.S., Lanna, A.C., Sousa, T.P., Chaibub, A.A., Souza, A.C.A., Filippi, M.C.C., 2019. N fertilizer dose-dependent efficiency of *Serratia* spp. for improving growth and yield of upland Rice (*Oryza sativa* L.). Int. J. Plant Prod. 13, 217–226.

Orozco-Mosqueda, M., Duan, J., DiBernardo, M., Zetter, E., Campos-García, J., Glick, B.R., Santoyo, G., 2019. The production of ACC deaminase and trehalose by the plant growth promoting bacterium Pseudomonas sp. UW4 synergistically protect tomato plants against salt stress. Frontiers in Microbiology 10, 1392. https://doi.org/10.3389/fmicb.2019.01392.

Panhwar, Q.A., Naher, U.A., Radziah, O., et al., 2015. Eliminating aluminum toxicity in an acid sulfate soil for rice cultivation using plant growth promoting bacteria. Molecules 20, 3628–3646. https://doi.org/10.3390/molecules20033628.

Paungfoo-Lonhienne, C., Lonhienne, T.G.A., Yeoh, Y.K., et al., 2014. A new species of *Burkholderia* isolated from sugarcane roots promotes plant growth. Microb. Biotechnol. 7, 142–154. https://doi.org/10.1111/1751-7915.12105.

Paungfoo-Lonhienne, C., Lonhienne, T.G.A., Yeoh, Y.K., et al., 2016. Crosstalk between sugarcane and a plant-growth promoting *Burkholderia* species. Sci. Rep. 6, 1–14. https://doi.org/10.1038/srep37389.

Peng, J.S., Gong, J.M., 2014. Vacuolar sequestration capacity and long-distance metal transport in plants. Front. Plant Sci. 5, 15. https://doi.org/10.3389/fpls.2014.00019.

Pilbeam, D.J., Morley, P.S., 2006. Calcium. In: Backer, A.V., Pilbean, D.J. (Eds.), Handbook of Plant Nutrition, 2nd edn. CRC Press, Boca Raton, pp. 121–144.

Poorter, H., Niklas, K.J., Reich, P.B., et al., 2012. Biomass allocation to leaves, stems and roots: meta-analyses of interspecific variation and environmental control. New Phytol. 30–50. https://doi.org/10.1111/j.1469-8137.2011.03952.x.

Postma, J.W.M., Keltjens, W.G., Van Riemsdijk, W.H., 2005. Calcium-(organo)aluminum-proton competition for adsorption to tomato root cell walls: experimental data and exchange model calculations. Environ. Sci. Technol. 39, 5247–5254. https://doi.org/10.1021/es048138v.

van Raij, B., Andrade, J.C., Cantarella, H., Quaggio, J.A., 2001. Análise química para avaliação da fertilidade de solos tropicais. Instituto Agronômico, Campinas.

Ramesh, S.A., Tyerman, S.D., Xu, B., et al., 2015. GABA signaling modulates plant growth by directly regulating the activity of plant-specific anion transporters. Nat. Commun. 6, 1–9. https://doi.org/10.1038/ncomms8879.

Rampazzo, P.E., Marcos, F.C.C., Cipriano, M.A.P., Marchiori, P.E.R., Freitas, S.S., Machado, E.C., Nascimento, L.C., Brocchi, M., Ribeiro, R.V., 2018. Rhizobacteria improve sugarcane growth and photosynthesis under well-watered conditions. Ann. Appl. Biol. 172, 309.

Rao, I.M., Miles, J.W., Beebe, S.E., Horst, W.J., 2016. Root adaptations to soils with low fertility and aluminium toxicity. Ann. Bot. 118, 593–605. https://doi.org/10.1093/aob/mcw073.

Reyna-Llorens, I., Corrales, I., Poschenrieder, C., et al., 2015. Both aluminum and ABA induce the expression of an ABC-like transporter gene (FeALS3) in the Al-tolerant species *Fagopyrum esculentum*. Environ. Exp. Bot. 111, 74–82. https://doi.org/10.1016/j.envexpbot.2014.11.005.

Riaz, M., Yan, L., Wu, X., et al., 2018. Boron alleviates the aluminum toxicity in trifoliate orange by regulating antioxidant defense system and reducing root cell injury. J. Environ. Manag. 208, 149–158. https://doi.org/10.1016/j.jenvman.2017.12.008.

Richardson, A.E., Barea, J.M., McNeill, A.M., Prigent-Combaret, C., 2009. Acquisition of phosphorus and nitrogen in the rhizosphere and plant growth promotion by microorganisms. Plant Soil 321, 305–339. https://doi.org/10.1007/s11104-009-9895-2.

Rijavec, T., Lapanje, A., 2016. Hydrogen cyanide in the rhizosphere: not suppressing plant pathogens, but rather regulating availability of phosphate. Front. Microbiol. 7, 1–14. https://doi.org/10.3389/fmicb.2016.01785.

Ryan, P.R., Delhaize, E., 2017. Adaptatiom to aluminium toxicity. In: Shabala, S. (Ed.), Plant Stress Physiology, 2nd edn. CABI, Boston, pp. 171–193.

Schlemper, T.R., Leite, M.F., Lucheta, A.R., Shimels, M., Bouwmeester, H.J., van Veen, J.A., Kuramae, E.E., 2017. Rhizobacterial community structure differences among sorghum cultivars in different growth stages and soils. FEMS microbiology Ecology 93 (8). https://doi.org/10.1093/femsec/fix096.

Schlemper, T.R., Dimitrov, M.R., Silva Gutierrez, F.A.O., et al., 2018. Effect of *Burkholderia tropica* and *Herbaspirillum frisingense* strains on sorghum growth is plant genotype dependent. PeerJ 6, e5346. https://doi.org/10.7717/peerj.5346.

Silva, S., Pinto, G., Correia, B., et al., 2013. Rye oxidative stress under long term Al exposure. J. Plant Physiol. 170, 879–889. https://doi.org/10.1016/j.jplph.2013.01.015.

Silveira, A.P.D., Sala, V.M.R., Cardoso, E.J.B.N., et al., 2016. Nitrogen metabolism and growth of wheat plant under diazotrophic endophytic bacteria inoculation. Appl. Soil Ecol. 107, 313–319. https://doi.org/10.1016/j.apsoil.2016.07.005.

Silveira, A.P.D., Iório, R.P.F., Marcos, F.C.C., et al., 2018. Exploitation of new endophytic bacteria and their ability to promote sugarcane growth and nitrogen nutrition. Antonie Van Leeuwenhoek. https://doi.org/10.1007/s10482-018-1157-y. (0123456789).

Silveira, A.P.D., Iório, R.D.P.F., Marcos, F.C.C., Fernandes, A.O., de Souza, S.A.C.D., Kuramae, E.E., Cipriano, M.A.P, 2019. Exploitation of new endophytic bacteria and their ability to promote sugarcane growth and nitrogen nutrition. Antonie van Leeuwenhoek 112 (2), 283–295.

Sing, S., Kumar, V., Sidhu, G., et al., 2019. Plant growth promoting rhizobacteria from heavy metal contaminated soil promote attributes of *Pisum sativum* L. Biocatal. Agric. Biotechnol. https://doi.org/10.1016/j.bcab.2019.01.035.

Singh, S., Tripathi, D.K., Singh, S., et al., 2017. Toxicity of aluminium on various levels of plant cells and organism: a review. Environ. Exp. Bot. 137, 177–193. https://doi.org/10.1016/j.envexpbot.2017.01.005.

Straub, D., Yang, H., Liu, Y., et al., 2013. Root ethylene signalling is involved in *Miscanthus sinensis* growth promotion by the bacterial endophyte *Herbaspirillum frisingense* GSF30T. J. Exp. Bot. 64, 4603–4615. https://doi.org/10.1093/jxb/ert276.

Sukweenadhi, J., Kim, Y.J., Choi, E.S., et al., 2015. *Paenibacillus yonginensis* DCY84 Tinduces changes in *Arabidopsis thaliana* gene expression against aluminum, drought, and salt stress. Microbiol. Res. 172, 7–15. https://doi.org/10.1016/j.micres.2015.01.007.

Usman, S., 2015. Biotechnology interventions for production of good quality seed canes. Int. J. Sci. Res. Innov. Technol. 2, 96–104.

Wang, T., Sun, H., Ren, X., et al., 2017a. Evaluation of biochars from different stock materials as carriers of bacterial strain for remediation of heavy metal-contaminated soil. Sci. Rep. 7, 1–10. https://doi.org/10.1038/s41598-017-12503-3.

Wang, Y., Li, R., Li, D., et al., 2017b. NIP1; 2 is a plasma membrane-localized transporter mediating aluminum uptake, translocation, and tolerance in *Arabidopsis*. Proc. Natl. Acad. Sci. 114, 5047–5052. https://doi.org/10.1073/pnas.1618557114.

Yan, Y., Kuramae, E.E., De Hollander, M., et al., 2017. Functional traits dominate the diversity-related selection of bacterial communities in the rhizosphere. ISME J. 11, 56–66. https://doi.org/10.1038/ismej.2016.108.

Yang, D., Chen, D., Wang, P., Jiang, D., Xu, H., Pang, X., Chen, L., Yu, Y., Li, K., 2017. Aluminium-inhibited NO3-uptake is related to Al-increased H_2O_2 content and Al-

decreased plasma membrane ATPase activity in the root tips of Al-sensitive black soybean. Funct. Plant Biol. 44, 198–207.

Yokosho, K., Yamaji, N., Fujii-Kashino, M., Ma, J.F., 2016. Functional analysis of a MATE gene OsFRDL2 revealed its involvement in Al-induced secretion of citrate, but a lower contribution to Al tolerance in rice. Plant Cell Physiol. 57, 976–985. https://doi.org/10.1093/pcp/pcw026.

Zerrouk, I.Z., Benchabane, M., Khelifi, L., et al., 2016. A *Pseudomonas* strain isolated from date-palm rhizospheres improves root growth and promotes root formation in maize exposed to salt and aluminum stress. J. Plant Physiol. 191, 111–119. https://doi.org/10.1016/j.jplph.2015.12.009.