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Microbial consortia inoculants stimulate early growth of maize depending on nitrogen and phosphorus supply

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Abstract: Adoption of microbial consortia as plant growth-promoting microorganisms (PGPMs) instead of single-strain inoculants is discussed as an approach to increase the efficiency and flexibility of PGPM-assisted production strategies. This study provides the functional characterisation of a commercial microbial consortia product (MCP) in a series of greenhouse experiments with maize on a silty-loam field soil (pH 5.9). A 60%-increased abundance of bacteria that could be cultivated after rhizosphere extraction was measured after MCP inoculation at the end of the 42-days culture period. MCP inoculation did not stimulate shoot biomass production of maize fertilised with nitrate, but growth improvement was recorded in combination with stabilised ammonium, especially with reduced phosphorus (P) supply. The MCP inoculant improved the acquisition of ammonium-N but also increased shoot-P. MCP inoculation stimulated root length development under reduced P supply with stabilised ammonium by 52%. This was accompanied by the increased auxin production capacity of rhizosphere bacteria. C-, N-, and P-turnover in the rhizosphere were little affected by the MCP inoculation, as deduced from the analysis of activities of extracellular soil enzymes. The findings suggest that the form of N supply is crucial for the efficiency of plant-MCP interactions.

Keywords: biofertilisers; root-associated microbiome; P solubilisation; acid phosphatase; plant-microbe interactions

The use of selected rhizosphere microorganisms with well-characterised beneficial properties as plant inoculants, commonly termed as "plant growth-promoting microorganisms" (PGPM), is discussed as a strategy with the potential to improve soil quality, plant health, nutrient acquisition and abiotic stress tolerance in cropping systems (Kloepper et al. 1991, Glick 2014). However, the expression of the desired effects under real rhizosphere conditions strongly depends on the rhizosphere competence of inoculants, i.e., their ability for compatible root colonisation of the host plant in competition with the indigenous microflora (Rajasekar and Elango 2011, Rana et al. 2012) and on the resistance against the various abiotic stress factors. To improve the flexibility of PGPM products, combined formulations based on multiple PGPM strains with complementary properties, are increasingly employed as so-called microbial consortia (Nuti and Giovannetti 2015, Sekar et al. 2016). These may also contain non-microbial biostimulants and stress-protective nutrients. Due to high production costs of single-strain combinations, frequently even less-defined microbial populations, originating from fermentation of various natural substrates, farmyard manure or composting processes are used as inoculants (Higa and Parr 1994,

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Hadar 2011, Lopez-Cervantes and Thorpe 2013). However, in these applications, standardisation of the composition and the possibility to achieve predictable results is even more challenging than in case of single strain inoculants.

The objective of the study was to investigate the performance and the modes of action of a commercial microbial consortia product (MCP), based on a mixture of selected beneficial fungal and bacterial strains and microorganisms originating from a fermentation process of organic substrates, further supplemented with seaweed extracts and discussed as a more targeted approach (Lopez-Cervantes and Thorpe 2013). Based on the physiological properties of the inoculants, complementary effects on processes mediating mineralisation of organic C, N and P sources in soils, N_2 fixation, P solubilisation and hormonal effects on plant growth, as well as pathogen suppression have been hypothesised as modes of action involved in the MCP-induced plant growth promotion (Lopez-Cervantes and Thorpe 2013). Unfortunately, it is not possible to identify the individual contributions of the various microbial strains to plant growth promotion and their potential interactions within complex consortia under real rhizosphere conditions. Therefore, the starting point of this study was a functional characterisation of the MCP product as a whole, with respect to the postulated beneficial properties (Lopez-Cervantes and Thorpe 2013). For an exemplary demonstration of the expected effects under environmental conditions characteristic for the maize rhizosphere, a set of pot experiments was conducted with the application of both nitrate and also ammonium fertilisers which are increasingly used for starter fertilisation in maize production systems. Moreover, recent findings suggest a beneficial effect of ammonium fertilisation on plant-PGPM interactions with microbial genera also used in the investigated consortium (Mpanga et al. 2018, 2019a,b). As indicators for the postulated beneficial MCP effects on processes with impact on nutrient availability in the rhizosphere (Lopez-Cervantes and Thorpe 2013) activities of marker enzymes involved in N, P and C cycling were determined in the rhizosphere soil with and without MCP inoculation (Baldrian 2009). To address the MCP-related root growth-promoting potential, the auxin production capacity of bacterial populations, re-isolated from the rhizosphere of inoculated and non-inoculated plants was determined at different time points of the culture period. Plate-counting assays with the re-isolated bacteria were conducted as an indicator for the root colonisation efficiency of the inoculants.

While the first part of the study exemplarily characterised the expression of potential plant growth-promoting properties of the MCP under rhizosphere conditions, the second part focused on the plant responses. The effects of the inoculants on shoot biomass production, root length development and the mineral nutritional status were evaluated in maize plants with different levels of N (nitrate *vs.* ammonium) and P supply.

MATERIAL AND METHODS

Plant cultivation. Greenhouse culture of maize (*Zea mays* cv. Jessy) was conducted on a silty loam field soil collected from the Ap horizon of a maize cultivation site in South-West Germany (Horb am Neckar, GMS coordinates: 48°26'39.23"N, 8°41'28.68"E): pH_{CaCl_2} 5.9; available P_{CAL}: 52 mg/kg soil (VDLUFA 1991); total N 0.15%; total C 1.1%. The soils were sieved with 2 mm mesh size, and the culture substrate was prepared as a 2:1 soil-sand mixture. Mineral nutrients were applied at different fertilisation levels (mg/kg soil): (*i*) (N140, P80, K150, Mg50), representing a standard full nutrient supply; (*ii*) reduced N/P fertilisation (N70, P0, K150, Mg50); (*iii*) reduced P fertilisation (N140, P0, K150, Mg50); (*iv*) an unfertilised control (0-Ctrl). Phosphate was applied as Ca $(H_2PO_4)_2$, K as K_2SO_4, Mg as $MgSO_4$ providing also sulfur in the sufficiency range for all treatments. Nitrogen fertilisation was performed as $Ca(NO_3)_2$ (calcinit; Yara International, Oslo, Norway) or $(NH_4)_2SO_4$ stabilised with the nitrification inhibitor DMPP (3,4-dimethylpyrazol-phosphate, NovatecSolub21; Compo Expert GmbH, Münster, Germany). Culture vessels contained 3 kg of the substrate, and the moisture content was regularly adjusted gravimetrically to 70% of the substrate water-holding capacity (WHC) throughout the culture period.

Application of MCP. A commercial microbial consortia product (EuroChem Agro GmbH, Mannheim, Germany) was used for inoculation. The MCP was based on bacterial and fungal strains including *Azotobacter vinlandii*, *Clostridium* sp., *Lactobacillus* sp., *Bacillus velezensis*, *B. subtilis* (SILo Sil® BS), *B. thuringiensis*, *Pseudomonas fluorescens*, *Acetobacter*, *Enterococcus*, *Rhizobium japonicum*, *Nitrosomonas*, *Nitrobacter*, *Saccharomyces*, *Penicillium roqueforti*, *Monascus*, *Aspergillus oryzae*, *Trichoderma harzia-*

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num T58 (TRICHOSIL®) and algae extracts from *Arthrospira platensis* (Spirulina) and *Ascophyllum nodosum* (Lopez-Cervantes and Thorpe 2013). For inoculation, a suspension of MCP 0.01325% (*w*/*w*) with non-chlorinated tap water was applied by soil-drenching close to the plants according to the instructions of the manufacturer (10 mL/plant) at 0, 14 and 28 days after sowing (DAS).

Plant growth and nutritional status. At the final harvest (42 DAS), the root systems were washed out of the soil substrate, and loosely adhering rhizosphere soil was collected by shaking and stored at –20 °C until further analysis. Root length was determined after digitalisation using the WinRhizo root analysis system (Regent Instruments, Quebec, Canada) and root and shoot dry matter was determined gravimetrically after oven-drying at 60 °C. For analysis of the P status, 250 mg of dried plant shoot material was subjected to 1.5 h microwave digestion at 1400 W (ETHOS.lab Professional Microwave System, MLS, Leutkirch, Germany) after 30 min extraction in 5 mL HNO_3 (conc.) 1:3, 3 mL H_2O_2 (30%) and 2 mL deionised water. Spectrophotometric determination of orthophosphate was conducted after the addition of molybdate-vanadate color reagent according to Gericke and Kurmis (1952) using a Hitachi U-3300 spectrophotometer (Hitachi Ltd, Tokyo, Japan). Total shoot N was measured by elemental analysis with a Vario Max CN macro-elementar analyser (Elementar Analysensysteme, Hanau, Germany).

Isolation of rhizosphere bacteria and auxin production assay. After final harvest at 42 DAS, bacteria from two grams of root samples with adhering rhizosphere soil were isolated into 50 mL of 0.1% protease peptone with two grams of sterile glass beads according to the method of Broadbent et al. (1971). Plate-count assays were performed after transfer to Standard 2 Nutrient Agar (Merck, Darmstadt, Germany) and Kings B Medium (Sigma-Aldrich, Germany for detection of fluorescent Pseudomonades (Naglitsch 1996), documented under UV light (354 nm). The auxin production potential of the bacterial isolates in 0.1% protease peptone was determined spectrophotometrically according to the method of Glickmann and Dessaux (1995). Bacterial dry biomass in the assay solution was estimated gravimetrically after 5 min centrifugation at 8 000 × g and subsequent oven drying of the bacterial pellet.

Enzymes involved in C, N, and P cycling in the rhizosphere. The activity of marker enzymes, mediating C, N and P cycling in the rhizosphere was assayed with fluorogenic substrates containing the fluorescence indicator 4-methylumbelliferone (4-MUF; Sigma-Aldrich, St. Louis, USA) according to Stemmer (2004). A microplate reader (Microplate Fluorescence reader FLx800, BioTek Instruments Inc., Winooski, USA) was used for monitoring the enzymatic hydrolysis of the MUF substrates for β-D-glucosidase (Glu, EC 3.2.1.21); L-leucin peptidase (LLpep, EC 3.4.11.1); L-tyrosin peptidase (LTpep, EC 3.4.11.1); cellulase (Cell, EC 3.2.1.21); xylosidase (Xyl, EC 3.2.1.37), acid (EC 3.1.3.2) and alkaline phosphomonosterase (EC 3.1.3.1) at 360/460 nm.

Experimental design and statistical evaluation. Experiments were arranged in a completely randomised design with five replicates per treatment. Prior to the analysis, outliers were eliminated according to Chromiński and Tkacz (2010). To ascertain significant differences, a one-way ANOVA analysis with a Tukey-test ($P \leq 0.05$) was performed. The results are presented as adjusted means ± standard deviations (SD). The SAS/STAT software package of SAS® 9.4 (2016) (SAS Institute Inc., Cary, USA) was used for statistical analysis.

RESULTS AND DISCUSSION

MCP effects on the abundance of rhizosphere bacteria. Re-isolation of cultivable bacteria from the rhizosphere of maize plants, using plate-count assays with standard 2 nutrient agar revealed a significant increase in total colony forming unit (CFU) of the inoculated plants by 60% (Figure 1) at six weeks after sowing (WAS). This point to a long-lasting rhizosphere effect, reflecting an increased abundance of MCP bacteria and/or a stimulatory effect on indigenous rhizosphere microbial communities induced by the repeated application of the inoculants, as recently shown also by Eltlbany et al. (2019). An interesting observation was the decline of fluorescent Pseudomonades induced by MCP inoculation, after plating of re-isolated bacterial communities on the selective Kings-B medium (Naglitsch 1996). Members of the genus *Pseudomonas* are known as efficient rhizosphere colonizers with plant growth-promoting but also pathogenic properties (Waschkies et al. 1994, Erlacher et al. 2014) and are included in the MCP inoculum. Therefore, a more detailed characterisation at the species level would be required to evaluate the potential consequences of declining *Pseudomonas* populations for the host plant. However, the observation exemplarily demonstrates a significant MCP

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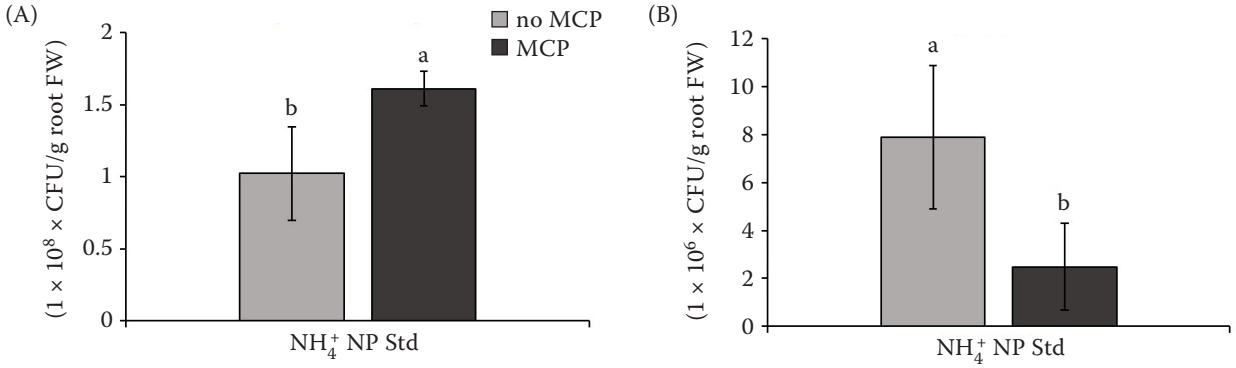

Figure 1. (A) Total cultivable rhizosphere bacteria on Std2 Medium and (B) colonies of fluorescent Pseudomonades cultivated on Kings B (KB) medium. Bacteria isolated from the rhizosphere of maize plants supplied with 130 mg ammonium N and 80 mg P per kg soil with and without microbial consortia product (MCP) inoculation. Data represent means and standard deviations of 5 replicates per treatment. Significant differences (Tukey-test, α < 5%) are indicated by different letters. CFU – colony forming unit; FW – fresh weight

interaction with the native soil microbiome as similarly shown in a more recent follow-up study, using an amplicon sequencing approach in a field experiment with tomato. In this case, the effects persisted even four months after the last inoculation, when no more changes in the abundance of the inoculated genera where detectable (Bradáčová et al. 2019a).

Marker enzyme activities for C, P, and N cycling in the rhizosphere. Based on the hypothesis that the MCP inoculants induce stimulatory effects on nutrient cycling in the rhizosphere (Lopez-Cervantes and Thorpe 2013), a functional characterisation was performed by measuring marker enzyme activities for turnover of C (β-glucosidase, cellulase, xylosidase), N (leucine and tyrosine peptidases) and P (acid and alkaline phosphatases) in rhizosphere soil samples according to Stemmer (2004), collected at 42 DAS. However, with the exception of a moderate decline in acid phosphatase activity by approximately 20%, and reduced cellulase activity, no inoculant effects were detectable (Table 1). Increased secretion of acid phosphatases is a typical response to P limitation by fungi and plant roots (Neumann and Römheld 2007), and the moderately declined activity may, therefore, reflect a slight improvement of the plant P status. The data do not support any direct involvement of the microbial inoculants in rhizosphere nutrient cycling, at least at the investigated sampling time. This is in line with recent follow-experiments, which showed significant stimulatory effects on the respective enzymatic activities in the maize rhizosphere only in a soil substrate with extremely low background activities of the respective enzymes, reflecting a low microbial activity due to long-term dry storage of the respective soil for more than 20 years. By contrast, on a freshly collected field soil, as similarly used in the

Table 1. The activity of rhizosphere marker enzymes for C, N, P turnover in the rhizosphere of maize plants fertilised with N in the form of Ca-nitrate (NO_3^-) *versus* DMPP-stabilised ammonium (NH_4^+) in comparison to an unfertilised control (0-Ctrl,) as affected by microbial consortia product (MCP) inoculation

Enzymatic activity (nmol × g/soil/h)		0-Crtl	NO_3^-		NH_4^+	
			no MCP	MCP	no MCP	MCP
C-turnover	β-D-glucosidase	601.45^a	490.73^a	526.97^a	667.35^a	549.09^a
	xylosidase	85.82^a	69.63^a	71.11^a	93.29^a	72.71^a
	cellulase	67.73^{ab}	88.10^a	63.26^b	57.90^a	76.50^{ab}
N-turnover	L-leucin-peptidase	441.70^a	359.30^{ab}	306.03^b	336.52^b	307.02^b
	L-tyrosin-peptidase	216.60^a	185.56^{ab}	151.01^b	164.85^b	170.97^b
P-turnover	acid-phosphatase	449.53^a	411.02^{ab}	333.92^b	411.15^{ab}	342.07^b
	alkaline-phosphatase	169.97^a	173.80^a	167.80^a	165.31^a	188.00^a

Presented data represent the means of five replicates. One-way ANOVA with Tukey test was performed. Different letters indicate significant differences between treatments ($P < 0.05$)

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present study, the inoculant effects were completely superimposed by high background activities of the rhizosphere marker enzymes exceeding the recorded MCP-induced changes by two orders of magnitude (Bradáčová et al. 2019b). This point to beneficial MCP effects on nutrient cycling and mineralisation preferentially expressed in heavily disturbed soil environments with limited microbial activities but not in fertile agricultural soils.

Auxin production potential of rhizosphere bacteria. Microbial production of auxins and molecules interfering with plant-hormonal signaling are among the best-documented features of PGPMs with beneficial effects on the root development of the host plants (Patten and Glick 2002, Ahmed and Hasnain 2010). Interestingly, rhizosphere bacteria re-isolated from MCP-inoculated maize plants with stabilised ammonium fertilisation revealed an increased auxin production potential as compared with the non-inoculated control, but exclusively in plants directly after the last MCP inoculation (28 DAS). This effect was lost at later stages of plant development at two weeks after the last inoculation (42 DAS) (Table 2). This is in line with earlier reports, demonstrating an increased auxin production potential detected for certain PGPM strains, such as of *Bacillus amyloliquefaciens* FZB42, *Pseudomonas* sp. DMSZ13134; *Pseudomonas putida* UB1 and *Acetobacter diazotrophicus* L1 (Patil et al. 2011, Bharucha et al. 2013, Mpanga et al. 2019a) supplied with ammonium instead of nitrate as mineral nitrogen source. These bacterial genera were also present in the MCP inoculant used in this study. Accordingly, Mpanga et al. (2019b) also reported beneficial effects of PGPM inoculants particularly during early growth of field-grown maize supplied with stabilised ammonium fertilisation, potentially related to the limited stability of the nitrification inhibitor. Consequently, nitrification and uptake of ammonium could explain the decline of beneficial effects on bacterial auxin production in the later stages of plant development. Similarly, Bradáčová et al. (2019b) reported root growth effects and expression of auxin-responsive genes in the root tissue induced by MCP inoculation of maize plants with stabilised ammonium fertilisation mainly during the first four weeks of the culture period, while the effects declined in later stages of plant development when inhibitory effects on nitrification were no longer detectable. However, the bacterial production of indole 3-acetic acid (IAA) has not only been related to the potential for stimulation of root growth but also with the ability for root colonisation, since IAA production mutants of various PGPR strains were also inefficient root colonisers (Spaepen et al. 2007). Therefore, the improved IAA production potential of bacterial populations in the rhizosphere of MCP-inoculated plants in response to ammonium supply may also contribute to improved root colonisation.

Table 2. Auxin production potential (relative values mg/microbial biomass) of rhizosphere bacteria isolated from maize roots inoculated with microbial consortia product (MCP) (with MCP) and non-inoculated control plants (no MCP) with nitrate or DMPP-stabilised ammonium fertilisation

Treatment		Harvest time			
		28 DAS		42 DAS	
		mean	SD	mean	SD
NH_4^+	no MCP	38.14^b	17.69	272.88^a	36.26
	with MCP	147.57^a	27.88	330.94^a	45.93
NO_3^-	no MCP	50.83^a	23.01	361.51^a	53.72
	with MCP	53.61^a	53.00	386.59^a	77.91

Data represent the means and standard deviation (SD) of five replicates. One-way ANOVA with Tukey test ($P < 0.05$) was performed. For each N form, different characters indicate significant differences between inoculated and non-inoculated plants. DAS – days after sowing

Effects on plant growth and mineral nutritional status. To assess potential benefits of the rhizosphere effects induced by MCP inoculation for plant growth and nutrient acquisition, a pot experiment was established comparing MCP responses of maize plants supplied with sufficient or reduced N and P supply and nitrate or ammonium as dominant nitrogen sources. Nitrogen was identified as the major limiting nutrient, and shoot growth was stimulated by 25% in comparison with an unfertilised control after the application of full N supply (140 mg N/kg soil) (Figure 2). This view is supported by the low N-status of control plants, indicating N deficiency (Figure 3A). The limited response to P fertilisation is in line with the moderate P availability of the investigated field soil (50 mg P_{CAL}/kg soil). Accordingly, the shoot P status (Figure 3C) at the end of the 42 days culture period reached 0.25–0.30% (*w*/*w*) even without additional P fertilisation, as a typical shoot P concentration under sufficient P supply (Campbell 2000). Nitrogen fertilisation increased both the N status (Figure 3A, B) and shoot growth, without significant differences between N forms (Figures 2 and 3).

Interestingly, MCP application improved shoot growth as compared with the non-inoculated controls in combination with ammonium supply but not with

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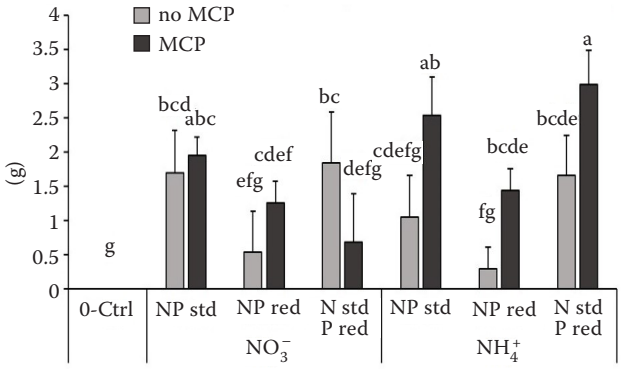

Figure 2. Increments in shoot dry biomass production of maize plants supplied with different levels of N (nitrate *vs.* ammonium) and P supply at 42 DAS (days after sowing) with and without microbial consortia product (MCP) inoculation as compared to the unfertilised control (0-Ctrl) (8.28 g dry matter (DM)). NP std = 140 mg N + 80 mg P/kg soil; NP red = 70 mg N + 0 mg P/kg soil; N std P red = 140 mg N + 0 mg P/kg soil. Data represent means and standard deviations of 5 replicates per treatment. Significant differences (Tukey-test, α < 5%) are marked with different letters above the bars

nitrate (Figure 2), as previously reported also for other inoculants based on strains of *Bacillus*, *Paenibacillus*, *Pseudomonas* and *Trichoderma* (Mpanga et al. 2019a,b). The increased shoot biomass production of MCP-treated plants was independent of P fertilisation, suggesting that MCP inoculation mainly improved the acquisition of the limiting nutrient N, supplied in the form of ammonium. Similarly, Mpanga et al. (2019b) also reported improved acquisition of stabilised ammonium fertilisers in combination with PGPM inoculants. Even after a reduction of N supply by 50% without additional P application, plants treated with the MCP-ammonium combination had similar biomass as those receiving the full dose of nitrate and P fertilisation (Figure 2).

Although P was not a growth-limiting nutrient (Figure 3), a general trend for an increased P status, both with respect to shoot concentration, as well as in the P content, were observed in the MCP variants. The increase of the P concentration by the MCP treat-

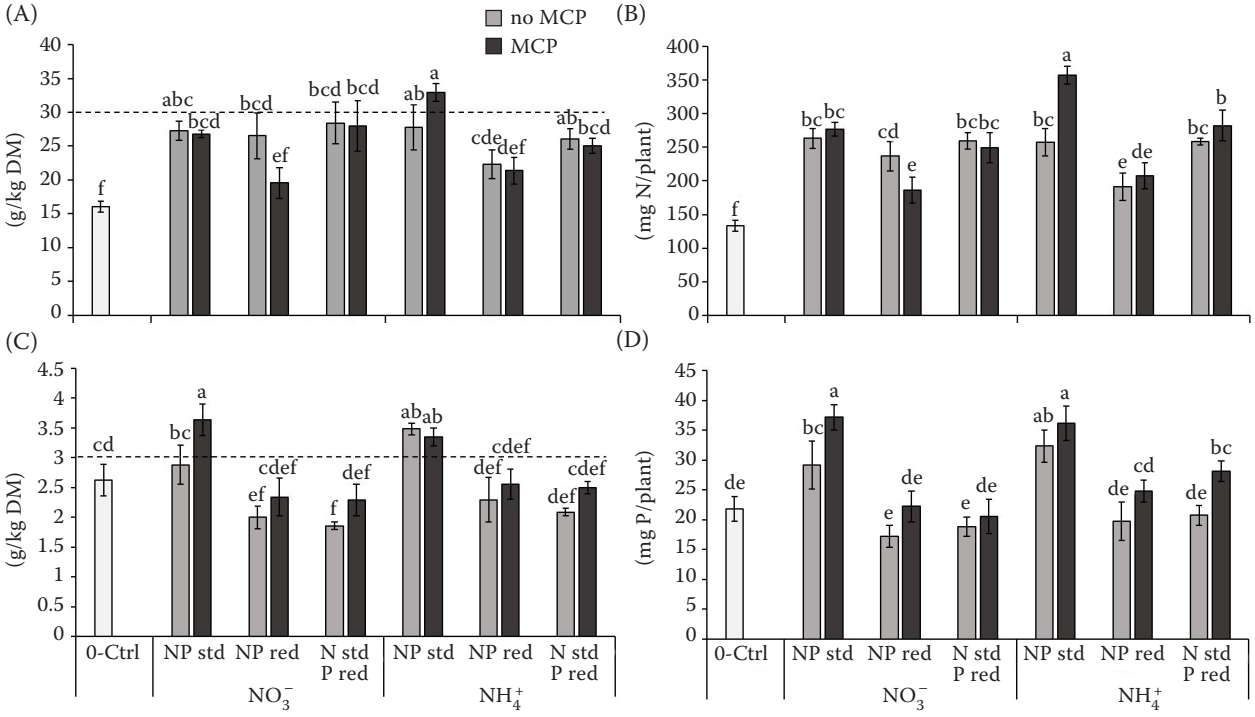

Figure 3. Concentrations and contents of N and P in the shoot tissue of maize plants with nitrate (nitrate) or stabilized ammonium fertilisation (ammonium) at 42 DAS (days after sowing) with and without microbial consortia product (MCP) inoculation. Values below the dashed lines indicate nutrient deficiencies, according to Campbell (2000). (A) N concentration in shoot tissue; (B) N content in shoot tissue; (C) P concentration in shoot tissue and (D) P content in shoot tissue. NP std = 140 mg N + 80 mg P/kg soil; NP red = 70 mg N + 0 mg P/kg soil; N std P red = 140 mg N + 0 mg P/kg soil. Data represent means and standard deviations of 5 replicates per treatment. Significant differences (Tukey-test, α < 5%) are marked with different letters

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ment, which was found in most treatments, except for the full ammonium-N and full P dose, indicates that the P utilisation within the plant changed. Furthermore, the P shoot accumulation was always increased by MCP inoculation, irrespective of the N-form supply (Figure 3C, D). The largest biomass was recorded with MCP inoculated plants with stabilised ammonium-N and reduced P fertilisation (Figure 2) and in this fertilisation regime, MCP inoculation was most beneficial for improving the P content of plants (Figure 3D). Direct effects of potential ammonium-induced rhizosphere acidification on P availability as described by Mpanga et al. (2019b), were not detectable in this study since the P status of the plants showed no N-form dependent differences in the absence of MCP inoculation (Figure 3C, D). Improved spatial acquisition of the native soil P *via* massively increased root length (+52%, Figure 4) is likely causal for the MCP effects under ammonium fertilisation (Table 2) and this effect can also improve the acquisition of other nutrients as recently shown by Mpanga et al. (2019a). This is also in accordance with the increased auxin production potential, detected for rhizosphere-bacterial populations of MCP inoculated plants. MCP inoculation increased shoot P concentrations also with full nitrate and P supply (Figure 3C, D) without any effect on root length (Figure 4). Thus, in this case, P acquisition of soluble P was stimulated by MCP inoculation *via* other, probably indirect mechanisms. A direct nutrient effect of the inoculum is unlikely since the effects were not detectable in all MCP treatments; the total P input of 0.19 mg/kg soil by the inoculum can be excluded as this was negligible.

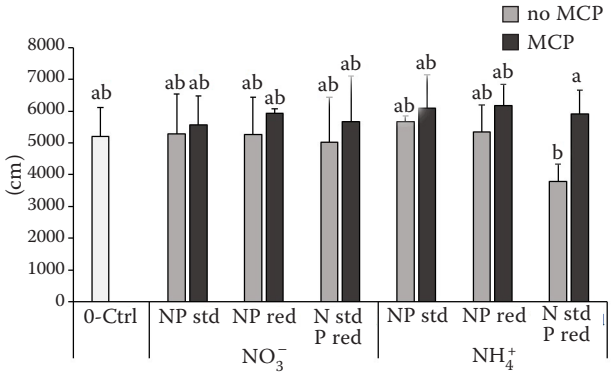

Figure 4. Total root length of maize plants with nitrate (nitrate) or stabilised ammonium fertilisation (ammonium) at 42 DAS (days after sowing) with and without microbial consortia product (MCP) inoculation. NP std = 140 mg N + 80 mg P/kg soil; NP red = 70 mg N + 0 mg P/kg soil; N std P red = 140 mg N + 0 mg P/kg soil. Data represent means and standard deviations of 5 replicates per treatment. Significant differences (Tukey-test, $\alpha < 5\%$) are marked with different letters

Taken together, the characterisation of the investigated MCP inoculant in a maize culture system revealed clear rhizosphere effects on the rhizosphere-bacterial abundance and composition, still detectable at the end of the 42 days culture period. Beneficial MCP effects on plant growth and nutrient acquisition were detected particularly in combination with stabilised ammonium fertilisation. The most intense growth stimulation was associated with significant MCP effects on root elongation for improved spatial nutrient acquisition under ammonium nutrition and increased auxin production of bacterial populations re-isolated from the rhizosphere of the respective plants. Similar results were recently reported for ammonium effects on maize plants inoculated with *Bacillus velezensis* (Mpanga et al. 2019b) as a bacterial species also present in the MCP inoculant. By contrast, there was no indication for longer-lasting MCP effects on nutrient cycling in the rhizosphere of the investigated soil. The results suggest that the form of N supply plays a crucial role in the expression of MCP effects and could offer a tool to improve the efficiency of plant-MCP interactions. However, the expression of MCP effects on different soils, different crops, the impact of variable environmental stress factors in the field and the relevance for real production conditions are aspects requiring further attention.

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