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Potentialities of plant growth promoting bacteria, isolated from *Portulaca oleracea* L. in soils with salinity

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Abstract

Objective: To determine the potential of rhizospheric and endophytic bacteria of *Portulaca oleracea* L. as growth promoters in soils with salinity.

Materials and Methods: Fifty four purslane plants were collected, from the Reque y Humedales de Eten district, in the Lambayeque region, Peru. From them isolations were made of rhizospheric and endophytic nitrogen-fixing bacteria. They were phenotypically identified, the ammonium, soluble phosphorus and indoles produced were quantified and the tolerance to 10 % NaCl was studied.

Results: The rhizospheric bacteria *Azospirillum* (23,1 %), *Azotobacter* (18,2 %), *Herbaspirillum* (15,85 %), *Beijerinckia* (12,2 %), *Burkholderia* (9,1 %), *Derxia* (6,01 %) and *Gluconacetobacter* (4,2 %) and the endophytic bacteria *Azospirillum* (43,3 %), *Herbaspirillum* (16,8 %), *Burkholderia* (14,4 %) and *Gluconacetobacter* (12,0 %) were isolated and identified.

Conclusions: The rhizospheric and endophytic bacteria of *P. oleracea* have as characteristics that they are nitrogen fixers, phosphate solubilizers, indole producers and tolerant to 10 % NaCl.

Keywords: PGPR, rhizospheric bacteria, plant growth promoters

Introduction

Soil salinization is one of the most serious problems faced by agriculture in the world (Zhang *et al.*, 2019). The affected soils are characterized by the accumulation of excessive quantities of soluble salts, exchangeable sodium or both and as consequence, fertility and biodiversity are lost and desertification is generated (Saier, 2010). In this context, the neglect of soils and deterioration of the structure and stability of agricultural communities generates rural workers' migration (Wicke *et al.*, 2011).

In the rhizosphere of plants in general, even in those that are salinity-tolerant, plant growth promoting rhizobacteria (PGPR) are found. These bacteria synthesize growth regulators, phosphate solubilizers and atmospheric nitrogen fixers, stimulate the ion absorption system, besides decreasing the attack of phytopathogens through competition, parasitism, enzymatic lysis and antibiosis and resistance induction (Nadeem *et al.*, 2014). PGPR constitute an alternative to decrease the use of chemical fertilizers. The recovery efficiency (percentage of the nutrient applied in the fertilizer absorbed by the plant) is different and is, as average 50, 30 and 60 % for

the applied N, P and K, respectively (Navarro-García and Navarro-García, 2014).

The species *Portulaca oleracea* L. proliferates on soils with salinity, due to its own mechanisms, such as tolerance to the sodium ion and osmotic adjustment; although in this performance a very important function is played by the microorganisms associated to the rhizosphere, which are adapted to the adverse conditions of the environment and, consequently, maximize their capacities as PGPR (Paul and Nair, 2008). Among these microorganisms, rhizospheric bacteria, after their inoculation in crops, can have a beneficial effect on them and make their establishment successful.

The Lambayeque region-Peru has problems of low production and salinity, derived from soil degradation. However, at present, the rhizospheric and endophytic bacteria of *P. oleracea* have not been isolated for their characterization, and later use as biofertilizers in crops planted on soils with high salinity. From these considerations, the objective of this work was to determine the potential of rhizospheric and endophytic bacteria of *P. oleracea* as plant growth promoters in soils with salinity.

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Materials and Methods

Plant material. The *P. oleracea* (purslane) plants were collected, from the Reque y Humedales de Eten district in the Lambayeque region, Peru. From them, isolations of rhizospheric and endophytic nitrogen-fixing bacteria were made.

Isolation, identification and characterization of bacteria. Soils that show salinity problems, recognized for their hardness and white crusts on their surface, were selected. Fifty four samples of purslane roots with adhered soil were collected. The plants were extracted with a shovel and approximately 50 g of roots and adhered soil were selected. They were deposited in polyethylene bags and were transported in a thermal box (10 ± 1 °C) to the Microbiology and Parasitology Laboratory of the National University Pedro Ruiz Gallo of Peru.

Simultaneously to sampling, for the isolation of bacteria, a sample composed of 1 kg of root soil for its physical-chemical analysis (table 1) was collected. The soil is strongly alkaline, with sandy texture, low organic matter and nitrogen levels, as well as moderate level of available phosphorus and potassium.

Table 1. Physical-chemical analysis of sample composed by root soil of *P. oleracea*.

Indicator	Value
pH	9,3
CE, dS m ⁻¹	29,5
OM, %	0,3
N, %	0,1
P, ppm	14,6
K, ppm	287,0

The aerobic and microaerophilic, nitrogen-fixing, rhizospheric bacteria, were isolated from the roots with rhizospheric soil, which were previously dehydrated under shade for 72 h. The roots were fragmented (5 cm) and 10 g of roots with adhered soil were randomly taken. Afterwards, they were deposited in flasks with 90 mL of sterilized saline solution (NaCl 0,7 % m/v). After agitating the content manually, during 10 minutes, aliquots were taken. In order to isolate aerobic bacteria, the aliquots were inoculated through the isolation technique by depletion through striation on the surface of solid culture media, without nitrogen. The puncture technique was used in semisolid culture media, without nitrogen (one drop per tube) for the isolation of microaerophilic bacteria.

The aerobic bacteria were isolated in the solid culture media LG (*Azotobacter*), LGD (*Derxia*) and

BEIJ (*Beijerinckia* spp), according to the indications made by Frontera (1983). After incubation at 30 ± 2 °C during two days, with the morphotypes of the representative bacteria, suspensions were obtained in sterilized saline solution and were subcultivated for their purification in their respective solid culture media.

For microaerophilic bacteria, the semisolid culture media were NFb and LGI (*Azospirillum* spp), JNFb (*Herbaspirillum* spp), LGI-P (*Gluconacetobacter* spp) and JMV (*Burkholderia* spp), according to Silva-Froufe *et al.* (2009). After incubation at 30 ± 2 °C during seven days, the media in which a whitish film under the surface and the change of the indicator were observed were selected. In similar culture media, twice consecutively, subcultures were made.

For the isolation of microaerophilic bacteria with the bacterial film, a suspension was obtained in sterile saline solution and was inoculated in the respective solid media. Then, they were incubated at 30 ± 2 °C during 48 h. The diverse morphotypes of the isolated bacteria were cultivated again in the semisolid medium (third subculture), and later in the corresponding solid medium. With the colonies of the aerobic and microaerophilic bacteria Gram staining was made, and they were cultivated in nutrient agar and solid and semisolid media, without nitrogen. Thus, the pure cultures of nitrogen-fixing rhizospheric bacteria were constituted, which were kept in refrigeration (8 °C).

The endophytic, microaerophilic nitrogen-fixing bacteria were isolated from the shallowly disinfected roots. The roots were washed with drinkable water to remove the adhered soil and were cut into 5-cm fragments. Five grams were weighed and deposited in previously sterilized 500-mL flasks with lid, in order to wash them with 50 mL of distilled water plus 0,005 % m/v neutral detergent during one minute. Afterwards, they were rinsed four times consecutively with sterile distilled water, at a rate of one minute per rinsing. Then, they were agitated for 15 minutes in potassium phosphate buffer solution (0,05 mol L⁻¹, pH 7,0). They were subject to immersion in alcohol (70 % v/v) for one minute, and were agitated during five minutes in 5 % sodium hypochlorite solution (NaClO), to which a few drops of Tween 80 were added. The tissue was taken to previously sterilized flasks for their immersion during one minute in 70 % alcohol by agitation in buffer solution for 15 min. Finally, they were washed four times with sterilized distilled water.

The disinfected plant tissue was deposited on drying paper to eliminate the moisture excess, and then it was taken to polyethylene bags of 16 x 15 cm, where it was crushed re-suspended in 0,5 mL of sterile saline solution (NaCl 0,87 % p/v). With a syringe 1 mL of the macerated material was extracted, and one drop was immediately inoculated by double puncture in the semisolid culture media without nitrogen: NFb, LGI, JNFb, LGI-P and JMV (Garrido-Rubiano, 2007). After inoculation, a similar procedure to the above-described one in microaerophilic nitrogen-fixing rhizospheric bacteria was followed.

The genus identification of the nitrogen-fixing rhizospheric and endophytic bacteria was carried out depending on the morphological and physiological characteristics, according to systematic bacteriology (Argüello-Navarro *et al.*, 2016). With all the bacteria, catalase, oxidase and motility tests were conducted. For aerobic bacteria of the *Azotobacter* genus, the nitrate reduction and glucose, sucrose, maltose and fructose acidification tests were carried out. For the genera *Derrxia* and *Beijerinckia*, the tests were indole production, utilization of citrate as carbon source, growth in 1 % peptone and acidification of glucose, sucrose and mannitol.

The quantification of the ammonium from the *in vitro* nitrogen fixation was done with Berthelot colorimetric method, the quantification of phosphorus product of *in vitro* phosphate solubilization

through the molybdate colorimetric method, and the quantification of *in vitro* produced indoles according to Salkowski colorimetric reaction (Lara-Mantilla *et al.*, 2007). Rhizospheric and endophytic bacteria were cultivated in nutrient broth with 5 % NaCl (m/v) at 30 °C, during 24 h. The bacterial growth was shown by turbidity or surface film, and cultivation in nutrient broth with 10 % NaCl (m/v) was done immediately. The bacteria were considered tolerant by the growth that was observed. This criterion, along with the higher values in the ammonium, soluble phosphorus and indole concentration, was considered for the selection of rhizospheric bacteria (eight aerobic, eight microaerophilic and eight endophytic ones).

Statistical analysis. Descriptive statistics was applied and the statistical program SPSS®, version 15.0, and Minitab® 15 were used.

Results

Bacteria isolation, identification and characterization. In the three solid culture media without nitrogen (LG, LGD and BEIJ) aerobic nitrogen-fixing bacteria were isolated, whose presence was shown by the developed colonies and the change of the indicator to yellow (table 2). The root sample frequency, which was calculated from the total of the isolations with rhizospheric soil and aerobic nitrogen-fixing bacteria, was 62,9, 33,3 and 14,8 % in LG, BEIJ and GD, respectively. In turn, in the

Table 2. Differential characteristics of aerobic nitrogen-fixing rhizospheric bacteria.

Characteristics	<i>Azotobacter</i>	<i>Derrxia</i>	<i>Beijerinckia</i>
Cells	Straight bacilli	Small bacilli	Small bacilli
Gram staining	-	-	-
Motility	+	+	+
Catalase	+	+	+
Oxidase	+	+	+
Indole production		+	-
Citrate utilization		+	+/-
Growth in 1% peptone		+	-
Acidification of:			
- Glucose	+	+	+
- Sucrose	+	+	+
- Fructose	+		
- Maltose	+		
- Mannitol		+	+
Reduction of nitrates	+		

isolation 73 pure cultures of aerobic nitrogen-fixing bacteria were obtained, whose provenance corresponded to 47,9, 32,8 and 19,17 % in LG, BEIJ and GD, respectively.

In five semisolid culture media without nitrogen, microaerophilic nitrogen-fixing rhizospheric and endophytic bacteria were isolated, whose presence was proven by a whitish film formed under the surface of the culture medium and by the change of the indicator (table 3). The frequency of root samples with rhizospheric soil, which was calculated from the total of the isolations of microaerophilic nitrogen-fixing rhizospheric bacteria, after three subcultivations, was 48,1; 38,8; 29,6; 27,7 and 6,6 % in JNFb; LGI; Nfb; JMV and LGI-P, respectively. Ninety one pure cultures of nitrogen-fixing bacteria were obtained, whose provenance corresponded to 29,6, 24,1; 18,6; 17,5 and 9,8 % in JNFb; LGI; Nfb; JMV and LGI-P, respectively (figure 1).

The frequency of root samples, calculated from the total isolations with nitrogen-fixing endophytic bacteria after three subcultivations, was 44,1; 29,6; 25,9; 20,4 and 20,3 %, in JNFb, LGI, Nfb, JMV and

LGI-P, respectively. In the isolation 83 pure cultures of nitrogen-fixing bacteria were obtained (figure 2).

In the nitrogen-fixing rhizospheric bacteria *Azospirillum* (23,1 %), *Azotobacter* (18,2 %), *Herbaspirillum* (15,8 %), *Beijerinckia* (12,1 %), *Burkholderia* (9,1 %), *Derxia* (6,09 %) and *Gluconacetobacter* (4,2 %) were identified. In the nitrogen-fixing endophytic bacteria *Azospirillum* (43,3 %), *Herbaspirillum* (16,8 %), *Burkholderia* (14,4 %), and *Gluconacetobacter* (12,04 %) were found. All this from the total isolated strains (figure 3).

Regarding the isolations of rhizospheric bacteria, among the ones of the LG medium *Azotobacter* (85,7 %) was identified; within BEIJ, *Beijerinckia* (83,3 %); and from the LGD medium, *Derxia* (71,4 %). Among the isolated bacteria in Nfb and LGI, *Azospirillum* (100 %) was recognized, and among those isolated in JNFb *Herbaspirillum* (96,2 %) was found. With regards to the ones of JMV medium, *Burkholderia* (88,2 %) was identified; while among the ones in LGI-P *Gluconacetobacter* (77,7 %) was found.

From the endophytic bacteria in the Nfb and LGI media *Azospirillum* (90 %) was identified, and within

Table 3. Differential characteristics of microaerophilic nitrogen-fixing bacteria.

Charateristics	<i>Azospirillum</i> spp.	<i>Herbaspirillum</i> spp.	<i>Gluconacetobacter</i> spp.	<i>Burkholderia</i> spp.
Cells	Pleomorphic	Curve bacilli	Small bacilli	Straight bacilli
Gram staining	-	-	-	-
Motility	+	+	+	+
Catalase	+	+/-	+	+
Oxidase	+	+	+	+
Urea hydrolysis	+	+/-	+	+
Jelly hydrolysis	-/+		+	+
Nitrate reduction	+		+	
Starch hydrolysis			+	
Lysine decarboxylation				
Growth in Nfb broth		+		+
Source of C (N fixation):				
Malic acid	+	+	+	+
Glucose	-/+	+	+	+
Mannitol	-/+	+	+	+
Sucrose	-/+	+	+	+
PHA granules	+		+	
Resistance Polymyxin B(300J)				+

* (+) positive; (-) negative.
; (-) negativo.

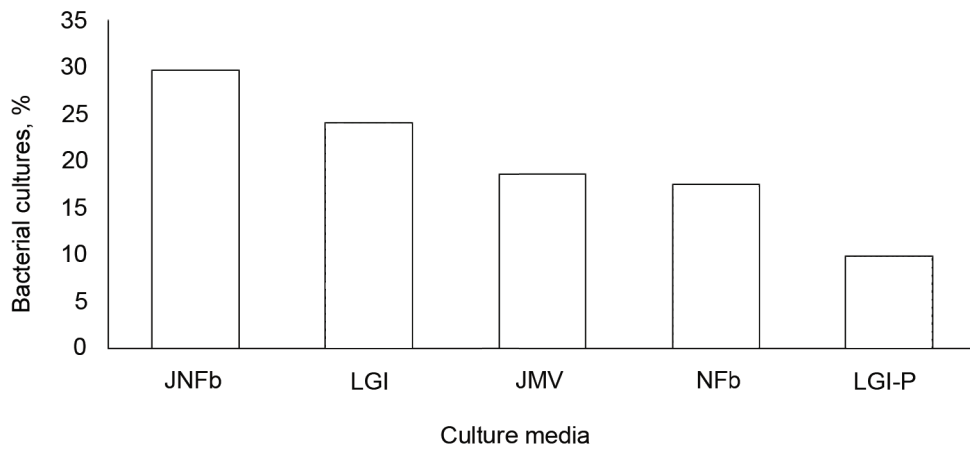


Figure 1. Percentage of microaerophilic nitrogen-fixing rhizospheric bacteria, isolated in five semisolid media.

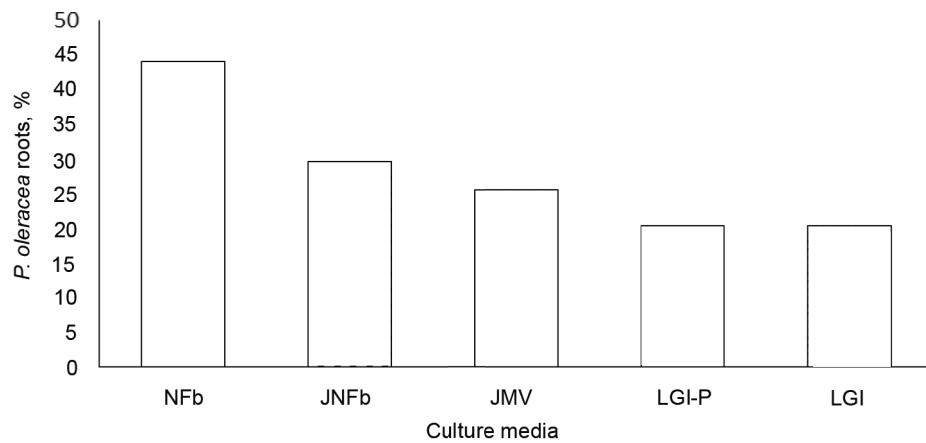


Figure 2. Frequency of *P. oleracea* roots with nitrogen-fixing endophytic bacteria, isolated in five semisolid media after three subcultivations.

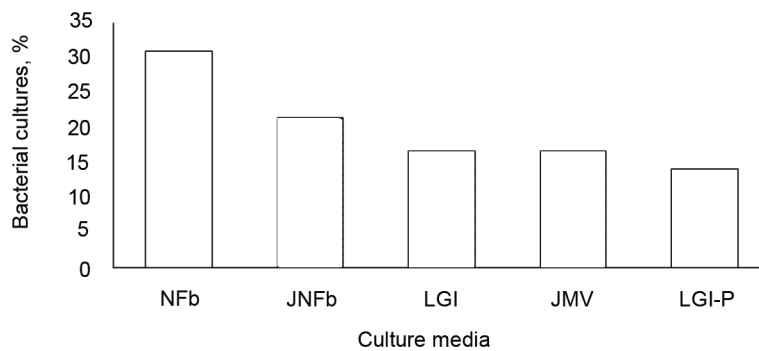


Figure 3. Percentage of nitrogen-fixing endophytic bacteria, isolated in five semisolid media.

the ones of the JNFb medium *Herbaspirillum* (82,6 %) was found. From the ones corresponding to JMV, *Burkholderia* (85,7 %) was identified and from the ones isolated in LGIP, *Gluconacetobacter* (83,3 %).

From the cultures of rhizospheric bacteria, 88,9 % fixed nitrogen *in vitro*. Of the bacteria, 36,5 % were anaerobic and 52,4 %, microaerophilic. In the cultures of isolated bacteria, 86,7 % of the bacteria were endophytic.

Table 4 shows the maximum ammonium concentrations for each genus: 64 ppm (*Azospirillum* sp. NfB19R); 27,2 ppm (*Azotobacter* sp. LG11A); 25,6 ppm (*Burkholderia* sp. JMV22bR); 20,8 ppm (*Herbaspirillum* sp. JNFb20R); 16 ppm (*Beijerinckia* sp. 9AR) and 8 ppm (*Derxia* sp. LGD5B and *Gluconacetobacter* sp. LGI-P45).

Of the rhizospheric bacteria cultures, 96 % solubilized phosphate *in vitro*. From the isolated bacteria,

54 % corresponded to aerobic and 42 % to microaerophilic ones; while, in the cultures of isolated bacteria, 95 % of the bacteria were endophytic (table 4).

The maximum concentrations of soluble phosphorus were: 64,9 ppm (*Burkholderia* sp. JMV53bR); 51,4 ppm (*Gluconacetobacter* sp. LGI-P33R); 19,2 ppm (*Herbaspirillum* sp. JNFb49R); 16,9 ppm (*Azospirillum* sp. LGI38R); 49,2 ppm (*Derxia* sp. LGD5C); 14,4 ppm (*Azotobacter* sp. LG11B) and 14,3 ppm (*Beijerinckia* sp. 4R). With the endophytic bacteria, the maximum concentrations corresponded to the following values: 16,02 ppm (*Azospirillum* sp. LGI54E); 11,89 ppm (*Gluconacetobacter* sp. LGI-P12E); 11,86 ppm (*Burkholderia* sp. JMV19E) and 11,39 ppm (*Herbaspirillum* sp. JNFb15E).

A percentage of 99,3 % (162) of the cultures of rhizospheric bacteria (43,0 % and 56,3 %, aerobic and microaerophilic ones, respectively) and 100 % (80) of the

Table 4. Characteristics of rhizospheric and endophytic bacteria, isolated from *Portulaca oleracea* L.

Code bacteria	Ammonium, ppm	Phosphorus, ppm	Indoles, ppm	Tolerance 10% NaCl
<i>Azotobacter</i> sp. LG11ARA	27,2	4,4	19,1	+
<i>Azotobacter</i> sp. LG11BRA	7,2	14,7	19,6	+
<i>Azotobacter</i> sp. LG33RA	8,8	11,2	30,0	+
<i>Azotobacter</i> sp. LG25RA	8,0	8,6	34,7	+
<i>Azotobacter</i> sp. LG27RA	8,0	8,6	56,7	+
<i>Beijerinckia</i> sp. 53AA	1,6	15,3	95,6	+
<i>Derxia</i> sp. LGD5CRA	2,4	6,7	49,2	+
<i>Derxia</i> sp. LGD7RA	4,0	10,9	19,1	+
<i>Azospirillum</i> sp. NfB42RM	14,4	13,9	23,7	+
<i>Azospirillum</i> sp. NfB447RM	8,8	12,8	51,4	+
<i>Azospirillum</i> sp. LGI49RM	10,4	9,3	3,6	+
<i>Azospirillum</i> sp. LGI12RM	9,6	7,4	84,3	+
<i>Burkholderia</i> sp. JMV47RM	12,0	6,7	27,7	+
<i>Gluconacetobacter</i> sp. LGI-P40RM	1,6	8,8	73,3	+
<i>Herbaspirillum</i> sp. JNFb22RM	10,4	9,6	18,5	+
<i>Herbaspirillum</i> sp. JNFb20RM	20,8	6,4	67,1	+
<i>Azospirillum</i> sp. NfB46E	3,2	11,4	54,2	+
<i>Azospirillum</i> sp. NfB50E	13,6	4,7	85,3	+
<i>Azospirillum</i> sp. LGI34AE	21,6	6,9	46,5	+
<i>Azospirillum</i> sp. LGI38E	4,8	8,4	30,9	+
<i>Burkholderia</i> sp. JMV1E	23,2	11,5	20,0	+
<i>Burkholderia</i> sp. JMV44E	3,2	13,2	36,2	+
<i>Gluconacetobacter</i> sp. LGI-P20E	3,2	11,1	26,2	+
<i>Herbaspirillum</i> sp. JNFb15E	13,2	4,7	85,3	+

RA= Aerobic rhizospheric, RM= Microaerophilic rhizospheric, E= Endophytic

endophytic bacteria produced indoles *in vitro* (table 4). The maximum indole concentrations were: 142,2 ppm (*Azospirillum* sp. LGI18R); 92,2 ppm (*Herbaspirillum* sp. JNFb13R); 80,8 ppm (*Burkholderia* sp. JMV53bR); 64,9 ppm (*Gluconacetobacter* sp. LGI-P45R); 56,7 ppm (*Azotobacter* sp. LG27); 51,4ppm (*Beijerinckia* sp. 1R) and 49,2 ppm (*Derrxia* sp. LGD 5C). With the endophytic bacteria, the maximum concentrations corresponded to the following values: 85,3 ppm (*Herbaspirillum* sp. JNFb15E); 64,9 ppm (*Gluconacetobacter* sp. LGI-P45E); 60 ppm (*Azospirillum* sp. Nfb 33E) and 49,6 ppm (*Burkholderia* sp. JMV21E).

From the cultures of rhizospheric bacteria, 73,3 % tolerated 5 % of sodium chloride. Aerobic and microaerophilic were 26,8 and 46,9 %, respectively. In the cultures of isolated bacteria, 78,5 % of the bacteria were endophytic.

On the other hand, 30,4 % of the cultures of rhizospheric bacteria tolerated 10 % of sodium chloride. From them, 12,19 % were aerobic and 18,29 %, microaerophilic.

The bacteria that tolerated 10 % of sodium chloride, and reached the highest values in the concentration of ammonium, soluble phosphorus and indoles, were studied to determine their effect on the vegetative development of tomato. For such purpose, 16 cultures of rhizospheric bacteria (eight of aerobic and eight of microaerophilic), and eight of endophytic ones were considered.

The genera of the other bacteria were identified with the utilization of glucose, mannitol and sucrose, as carbon sources for nitrogen fixation. In addition, the growth in Nfb broth, for *Herbaspirillum*; urea hydrolysis, jelly hydrolysis and nitrate reduction for *Gluconacetobacter*; and jelly hydrolysis, lysine decarboxylation and resistance to 300 UI of Polymixin B, in the case of *Burkholderia*, were utilized.

In 85,7 % (30) of the rhizospheric bacteria isolated in the LG medium, the *Azotobacter* genus was identified. In 83,3 % (20) of the ones isolated in BEIJ, the *Beijerinckia* genus was recognized, and in 71,4 % (10) of those isolated in the LGD medium, *Derrxia*. In 100 % (38) of the bacteria that were isolated in Nfb and LGI *Azospirillum* was identified; in 6,2 % (26) of the ones isolated in JNFb, *Herbaspirillum* could be observed. From those that were in the JMV medium, in 88,2 % (15) *Burkholderia* was recorded, and 77,7 % (7) from those that were isolated in LGI-P, was identified as belonging to the *Gluconacetobacter* genus.

In 90 % (36) of the endophytic bacteria, isolated in the Nfb and LGI media, the *Azospirillum* genus was observed; in 82,6 % (14) from the ones that were found in the JNFb medium, *Herbaspirillum* was recognized. Meanwhile, in 85,7 % (12) of the bacteria that were in the JMV medium, *Burkholderia* was identified; and in 83,3 % (10) of the isolated bacteria in LGIP, the *Gluconacetobacter* genus was recorded.

Discussion

Due to the constant application of chemical products in the cultivation fields, the production cost of harvests, the environmental quality of soil and water have been affected (Solomon *et al.*, 2000). Rhizospheric bacteria, which inhabit the soil of the root surface, establish a relation with plants, although not obligate. They are considered endophytic because they live in the tissues (Agua-do-Santacruz, 2012) during one stage (facultative) or during their entire life cycle (obligate), for which they were isolated from the previously disinfected roots.

Rhizobacteria are capable of stimulating plant growth through various mechanisms, which include the improvement of nutrition, phytohormone production and regulation, as well as the suppression of disease-causing organisms (Martínez-Viveros *et al.*, 2010).

In the endophytic bacteria, the genera *Azospirillum*, *Herbaspirillum*, *Burkholderia* and *Gluconacetobacter*, previously referred in *Zea mays* L., were identified. Endophytic bacteria have been isolated from monocotyledonous and dicotyledonous plants, which include from ligneous tree species (Brooks *et al.*, 1994) to herbaceous plants, such as sugar beet (Jacobs *et al.*, 1985) and *Z. mays* (Gutiérrez-Zamora and Martínez-Romero, 2001).

In the rhizospheric bacteria, *Azospirillum*, *Azotobacter*, *Beijerinckia*, *Burkholderia*, *Herbaspirillum*, *Gluconacetobacter* and *Derrxia* spp. were recognized. These bacterial genera have been reported before in the rhizosphere of diverse agricultural crops: *Gluconacetobacter* in *Z. mays* (Mehnaz *et al.*, 2006, *Azospirillum*, *Azotobacter*, *Beijerinckia* and *Burkholderia* in *Oryza sativa* L. (Ahmadi-Rad *et al.*, 2016). *Herbaspirillum*, *Burkholderia*, *Gluconacetobacter*, *Azospirillum*, *Azotobacter*, *Beijerinckia* and *Derrxia* in *Megathyrsus maximus* (Jacq.) B.K.Simon & Jacobs, *Dichanthium aristatum* (Poir.) C.E. Hubb and *Brachiaria* sp. (Garrido-Rubiano, 2007; Garrido *et al.*, 2010)

PGPR often have more than one mechanism to improve plant growth. The experimental evidence

shows that their stimulation is the net result of multiple processes that are simultaneously activated (Martínez-Viveros *et al.*, 2010). The isolated bacteria in this study fixed nitrogen, solubilized phosphate and synthesized indoles. These solubilizing microorganisms show effects on plant growth, which include production of indoleacetic, gibberellic acid, cytokinins and ethylene; as well as in nitrogen fixation, necessary characteristics for a microorganism to be considered growth promoter (Banerjee *et al.*, 2010; Souza *et al.*, 2015). Likewise, it is stated that these microorganisms increase height, root and aerial biomass of the plants, which follows the release of chemical products that favor growth (González and Fuentes, 2017). In inoculation trials they also showed significant increase in the dry weight of the sprout under controlled conditions, an increase in the P solubilization capacity and an improvement in the N assimilation efficiency (Montañez *et al.*, 2012).

Conclusions

The rhizospheric and endophytic bacteria of *P. oleracea* are nitrogen fixers, phosphate solubilizers, indole producers and tolerant to 10 % NaCl.

Authors' contribution

- Marbil Corrales-Lozada. Participated in the genesis of the idea, project design, data collection and interpretation, result analysis, as well as in the manuscript preparation.
- Victoria Lumbres. Participated in the genesis of the idea, project design, data collection and interpretation, result analysis, as well as in the manuscript preparation.
- Sebastian Iglesias-Osores. Participated in the genesis of the idea, project design, data collection and interpretation, result analysis, as well as in the manuscript preparation.
- Carmen Carreño-Farfán. Participated in the genesis of the idea, project design, data collection and interpretation, result analysis, as well as in the manuscript preparation.

Conflicts of interests

The authors declare that there are no conflicts of interests among them.

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