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Isolation and characterization of three new PGPR and their effects on the growth of *Arabidopsis* and *Datura* plants

Bilal Rahmoune a, Abdelkader Morsli b, Madjda Khelfi-Slaoui a, Lakhdar Khelfi a, Andrew Strueh b, Alexander Erban c, Joachim Kopka c, Jürgen Preti b and Joost T. van Dongen b

aLaboratory of Genetic Resources and Biotechnology, École Nationale Supérieure d’Agronomie (ENSA), Algiers, Algeria; bLaboratory of Molecular Ecology of the Rhizosphere, Institute of Biology I – RWTH Aachen University, Worringerweg, Aachen, Germany; cMax-Planck-Institut für Molekulare Pflanzenphysiologie, Potsdam, Germany

**ABSTRACT**

This study characterizes three bacterial strains isolated from plant rhizospheres and evaluates their performance as plant-growth-promoting rhizobacteria. *Pseudomonas plecoglossicida* strain Pp20 was isolated from the rhizosphere of a date palm in Bechar (Algerian Sahara), *Bacillus* spec. strain Bt04 was isolated from the rhizosphere of pear in Ghardaïa (Algerian Sahara) and *Lysinibacillus fusiformis* strain LF89 was isolated from the rhizosphere of tomato in Ain Defla (northern Algeria). Their effects on plant growth and development were analyzed in different in vitro cultures: an *Arabidopsis thaliana* plate assay and two hydroponic systems for *Datura stramonium* and *Datura tatula*. Our results show that all strains significantly improve plant growth of the species tested and some strains produce a shift in the C/N ratio in *A. thaliana*. Inoculation had no effect on alkaloid production per gram leaf dry weight in *D. stramonium*, but specific plant-growth-promoting rhizobacteria interactions may alter the alkaloid composition in the shoot.

**Introduction**

Plant-growth-promoting rhizobacteria (PGPR) are soil bacteria which live in the rhizosphere of plants, where they stimulate plant growth and development of their hosts. PGPR belong to the most important and agronomically useful soil microbiota that involve free-living bacteria (Lugtenberg & Kamilova 2009; Bhattacharyya & Jha 2012). They are characterized by the following properties: (1) they must be able to colonize the root surface, (2) they must be able to compete with other microbiota to express their plant-growth-promoting/protecting activities and (3) they must promote plant growth (Kloepper 1994).

PGPR can support plants by the production of several metabolites, including siderophores and hydrocyanic acid (HCN) (Bhatia et al. 2005), plant hormones such as indole acetic acid (IAA) and some other auxins, gibberellins, cytokinins (García de Salamone et al. 2001; Persello-Cartieaux et al. 2001; Patten & Glick 2002) and ethylene (Glick 1995).

Some strains are found to protect plants from pathogen attack by killing parasites. These PGPR produce, for example, antibiotics such as phenazines, pyoluteorin and pyrrolnitrin. PGPR can also help plants to grow by providing soluble phosphate converted by acidification from insoluble mineral phosphates, or via mobilization of other essential nutrients that can also help in growth improvement of plants (De Freitas et al. 1997; Bertrand et al. 2001).

The genus *Datura* (family *Solanaceae*) consists of nine (annual and tree) species (Geeta & Gharaiheb 2007). They are known since antiquity for their narcotic and medicinal potentials (Dafni & Yaniv 1994; Higgins 2006). Ingestion of *Datura* plant parts or extracts can cause serious poisoning or hallucinations (Chan et al. 1995; Birmes et al. 2002). The toxic components mainly consist of the tropine alkaloids hyoscyamine and atropine (which are stereoisomers) and scopolamine (Bruneton 1999). Atropine and scopolamine have anticholinergic properties and are used in legitimate medical applications in very low doses. Scopolamine (also called hyoscine) is an antimuscarinic agent and a smooth muscle relaxant. It is also an antispasmodic agent with antinauseant properties and is extensively used in the treatment of motion sickness and in pre-operative medication (Van Wyk et al. 2002). Atropine causes blurred vision, suppressed salivation, vasodilation, increased heart rate and delirium (Bruneton 1999). It also reduces rigidity in Parkinsonism and is used as an antidote to poisoning with parasympathomimetic agents, for example, nerve gases and organophosphorus insecticides.

Research on the effects of PGPR on medicinal plants is limited (Lenin & Jayanthi 2012), and mechanisms involved have not been described. Indigenous plant-growth-promoting microorganisms of medicinal plants and their effects in the plant rhizosphere can be used to increase the growth of such species (Sharma et al. 2015). Moreover, inoculation of medicinal plants with PGPR could also influence the quality and quantity of bioactive constituents and their potential in agriculture, pharmaceutical and medicine (Sharma et al. 2015). Only very few studies have been published specifically addressing the effects of PGPR on medicinal plants. The mechanisms reported for interactions with plants in general may also apply for medicinal plants. The objective of this study was to evaluate the effect of inoculation with PGPR on the growth of *Arabidopsis* and the C/N ratio and also on the growth of *Datura* and its ability to produce alkaloids.
Materials and methods

Datura seeds origin

*Datura stramonium* and *Datura tatula* seeds were collected from plants grown in the National School of Agronomy (ENSA-Algiers) and harvested in September 2009. For *Arabidopsis thaliana*, the ecotype colombia-0 was used.

Bacterial strain and culture media

Isolation and screening of bacteria strains

Three bacterial strains were isolated from different regions in Algeria. Strains Pp20 and Bt04 were isolated from a pear tree rhizosphere in Ghardaia, date palm in Bechar and tomato in Ain Della.

Soil rhizosphere samples were serially diluted and the suspensions were used to isolate bacterial strains on plates containing King B agar medium (King et al. 1954) at 30°C for 48 h.

Identification of bacterial isolates by 16S rRNA amplification and sequencing

Three bacterial strains were isolated from different regions in Algeria. Strains Pp20 and Bt04 were isolated from a pear tree rhizosphere in Ghardaia: 32° 29’ 00” N, 3° 40’ 00” E and a date palm rhizosphere (in Bechar: 31° 33’ 40” N, 2° 14’ 24” W) in the Sahara region of Algeria, respectively. Strain Lf89 was isolated from the rhizosphere of a tomato plant in the north of Algeria in Ain Della (36°15’50” N, 1°58’04” E).

Genomic DNA was isolated from each strain and partial 16S rDNA sequence, including the V1–V8 regions, was amplified using universal primers BAC 27f (5’-GAG TTT TGA CCT TGT TAC GAC TT-3’) and BAC 1492r (5’-CGG TTA CCT TGT TAC GAC TT-3’).

16S rDNA amplicons were custom sequenced from both ends at SeqLab (Göttingen, Germany). The nucleotide sequences obtained were processed to remove low-quality ends at SeqLab. The resulted high-quality sequences were analyzed using BLASTn (NCBI) to identify the nearest neighbor.

16S rDNA sequence identity of the first isolated showed 100% identity to *Pseudomonas plecoglossicida* NBRC 103162 (accession NR_114226) and *P. plecoglossicida* FPC951 (accession NR_024662) and will therefore be referred to as *P. plecoglossicida* strain Pp20 here. The second isolate showed 99% identity to *Bacillus toyonensis*, *Bacillus thuringiensis* and *Bacillus cereus* strains and will therefore be referred to as *Bacillus* spec. strain Bt04 here; the third isolate showed 99% identity to *Lysinibacillus fusiformis* NBRC15717 (accession NR_112569) and *L. fusiformis* DSM 2898 (accession NR_042072) and will therefore be referred to as *L. fusiformis* strain Lf89 here.

All partial 16S rRNA sequence data have been deposited in the NCBI GenBank Nucleotide database under the accession numbers KU321233 for Pp20, KU321234 for Bt04 and KU321235 for Lf89.

Growth promotion of bacterial isolates on *A. thaliana*

An in vitro assay to study growth promotion of bacterial isolates on *A. thaliana*, WT colombia-0 variety, was used as described in Schwachtje et al. (2011). Briefly, seeds were pre-incubated in a bacterial KB-solution for 1 h, prior to placing them on agar plates with 0.5 MS. The plates were then transferred to a Percival growth chamber where plants were grown with a photoperiod of 12/12 h (light/dark) and temperatures of 22°C/20°C (light/dark) for 4 weeks.

Parameters measured

Root length is the length of the main root from the origin at the seed to the tip. The number of small roots around the main root was counted and the rosette diameter was measured.

Total carbon and nitrogen

Carbon and nitrogen content of the shoots of *A. thaliana* were determined by dry combustion using a Vario EL III elemental analyzer (Elementar Analysetechnik, Hanau, Germany). Around 10–20 mg of heat dried, water-free samples for each treatment were used.

Germination of the seeds and growing conditions for Datura

*Datura* seeds were scarified using paper glass (80 point) (Khefli-Slaoui et al. 2005) and germinated in soil. For the establishment of hydroponics, seedlings were transferred from soil to hydroponic systems containing nutritional MS (Murashige & Skoog 1962) mineral solution [macro-elements, micro-elements and iron (no sugar and vitamins added), pH: 5.8].

Datura experimental setup 1

*D. stramonium* plants of five weeks old were transferred from soil to a hydroponic system. The hydroponic system consisted of six PVC tubing containing five plants each. Each PVC tube was linked to a separate tank containing nutrient solution. The circulation of nutrient solution was assured by six pumps, one pump per PVC tube and tank. Root irrigation was assured by spray nozzles. Each treatment was performed in two PVC tubes each. Bacterial inocula (Strain Pp20 and Bt04) were applied for 16 days. Growth promotion parameters were measured 2, 8 and 16 d.a.i. (days after inoculation). Plant length was measured from the collet to the primary bud. Fresh total weight was measured for the whole plant 2, 8 and 16 d.a.i. Plant dry weight was measured after 16 days as dry weight (roots and stems) incubated for 3 days at 50°C.

Datura experimental setup 2

Twelve, five-week-old *D. tatula* plants were transferred from soil to a hydroponic system and plants were inoculated with bacterial strains Pp20 or Lf89. The system used was a bin containing MS medium, connected to air pump by a pipe with an air divisor, to generate bubbles in the solution. Plant parameters were measured at 1, 2 and 3 weeks a.i. Root length was measured from the collet to the tips. The number of leaves was counted and the plant length was measured from the collet to the terminal bud. Plant dry weight was measured after 3 weeks, as dry mass (roots and stems) incubated for 3 days at 50°C.
Alkaloid extraction and quantification

Metabolites from lyophilized plant material were extracted with methanol/chloroform, containing 13C sorbitol and D4-alanine as internal standards, derivatized with N-Methyl-N-(trimethylsilyl) trifluoroacetamide and analyzed by Gas Chromatography–Time of Flight-Mass Spectrometry (GC–TOF–MS) as described previously (Lisec et al. 2006). Chromatograms were evaluated using TagFinder (Luedemann et al. 2008) and alkaloids were manually identified by comparison of mass spectra and retention indices with the reference library of mass spectra and retention indices from the Golm Metabolome Database [http://gmd.mpimp-golm.mpg.de/ (Schauer et al. 2005; Kopka et al. 2005)].

Statistical analysis

All statistical analyses were performed with Excel (version 2010) and Statistica (version10). Data for plant growth experiments are presented as means ± standard deviation. Statistical significance was defined as p < .05 (Newman–Keuls test).

Results

Strains Pp20, Bt04 and Lf89 promote growth of A. thaliana in vitro

Because the three isolates Pp20, Bt04 and Lf89 were isolated from plant roots, we tested them for their capacity as PGPR strains on A. thaliana in an in vitro assay.

As parameters of growth root length, number of secondary roots (branching) and rosette diameter were measured (see Figure 1(a)). Compared to control plants without bacteria, bacterial inoculation significantly increased root lengths as well as root branching. Also rosette diameter significantly increased after inoculation.

Inoculation with some isolates changes the C/N ratio of A. thaliana shoots material

The ratio between carbon (C) and nitrogen (N) in plant material is a measure of the physiological status of plant metabolism. Plants optimize their growth according to their cellular C and N balance (C/N ratio). The C/N response regulation in plant cells plays an important role in growth and development (Coruzzi & Bush 2001). Carbon and nitrogen content and variation in A. thaliana shoots were evaluated following the inoculation with the three PGPR tested in this work. While the C/N ratios for the control plants and for plants inoculated with Lf89 show a mean value of 4.26 and 4.35, respectively, plants inoculated with Pp20 and Bt04 showed a significant downshift to 3.91 and 3.89, respectively (Figure 1(b)).

Bacterial isolates promote growth of D. stramonium and D. tatula in two different hydroponic systems

In order to investigate the performance of Pp20, Bt04 and Lf89 to promote Datura growth, we studied the effect of the inoculation in two different hydroponic systems, one for D. stramonium and the other for D. tatula as mentioned in Materials and Methods section.

Inoculation of D. stramonium plants with the bacterial strains Pp20 and Bt04 induced a small increase of plant length after 2 days (Figure 2(a)). Eight days after inoculation, Pp20 and Bt04 increased plant length by 18% and 14%, respectively; 16 days after inoculation, the length of plants inoculated with Pp20 was increased by 20%, while Bt04 increased by 15%.

For plant weights, a maximum value of 35% increase was obtained for inoculation with Bt04 after 16 days (Figure 2(a)). After drying of the 16 days samples, also the leaves and the roots dry weight showed a significant increase for plants treated with PGPR when compared to the control (Figure 2(b)).

Inoculation of D. tatula plants with strains Pp20 and Lf89 induced clear changes in plant length. A three weeks after inoculation, the highest growth promotion was obtained for Pp20-inoculated plants with an increase of 33% compared to the control plants. For Lf89, we measured an increase of 31% over the controls (Figure 2(c)). The number of leaves per plant and root length increased when compared to the controls (Figure 2(c)). We also found that Pp20 and Lf89 increased shoot and root dry weight with approximately 150% for both strains (Figure 2(d)). Root dry mass was increased by 342% with Lf89 and by 142% with Pp20 compared to the control (Figure 2(d)).

Alkaloid content of D. stramonium plants inoculated with Pp20 and Bt04

Because we demonstrated a positive influence of the inoculation on Datura plant growth, we also wanted to investigate whether there is a measurable effect on alkaloid content. We used an established metabolite profiling pipeline to quantify the relative contents of hyoscyamine, scopolamine and tropine, a precursor of hyoscyamine and atropine. We used the leaf and root material of a D. stramonium experiment to extract and quantify those alkaloids. Figure 3 shows that scopolamine contents are generally higher in leaf material, while hyoscyamine and tropine levels are higher in roots. Inoculation with strain Bt04 increased the content of scopolamine in the leaves when compared to the control, but consistent significant changes were not observed.

Discussion

In our experiments, we used: (1) L. fusiformis strain, a ubiquitous Gram-positive, aerobic, rod-shaped, mesophilic and spore-forming bacterium that is commonly isolated from soil. At present, the genus *Lysinibacillus* is composed of 18 species (Cheng et al. 2015). Strains of the genus *Lysinibacillus* contain lysine and aspartate in the peptide subunit of the cell wall peptidoglycan as diagnostic amino acids rather than meso-diaminopimelic acid, which is characteristic of the genus *Bacillus* (Miwa et al. 2009). In 2015, Prabha et al. (2015) found that *L. fusiformis* strain AU01 produces extracellular protease and is able to produce an intracellular esterase. AU01 is also capable of producing a glycolipid type of biosurfactant capable of inhibiting biofilm formation by pathogenic bacteria (Pradhan et al. 2014). Also, Sgroy et al. (2009) found that *L. fusiformis* strain Ps7 showed a capacity to grow in nitrogen-free conditions and produced some plant hormones but was not able to produce siderophores, ACC deaminase or to solubilize phosphate. Park et al. (2005) showed that *L. fusiformis* PM5 and PM24 produced 100 μg ml⁻¹ IAA in defined medium, and this was considered...
as a promising potential mechanism for developing plant growth in inoculation conditions.

(2) *P. plecoglossicida* is Gram-negative non-sporulating, aerobe, rod shaped, motile by one or several polar flagella (Liu et al. 2008). Its cells are approximately 0.5 μm in width and 2.0–2.5 μm in length. Pseudomonads are well-known widespread microorganisms that have been isolated from a variety of natural sources, including soil, plants, mineral waters and clinical specimens, which are characterized by a high level of metabolic diversity (Moore et al. 1996). Metabolism is oxidative. Solid growth occurs at temperatures between 10°C and 30°C in the presence of 0–5% (w/v) NaCl. A fluorescent pigment is produced weakly on King medium B (Nishimori et al. 2000). It also has a collection

![Figure 1](image1.png)

**Figure 1.** Effect of inoculation with Pp20, Bt04 and Lf89 on plant growth and C/N ratio in the shoots of *A. thaliana*. Measurements were performed 3 weeks after inoculation. (a) Plant growth parameters are expressed here as: root length, number of small roots and rosette diameter. Each value is the mean of six replicates. (b) Values are expressed as: nitrogen content (%) and carbon content (%). Each value is the mean of three replicates. Error bars represent ±standard deviation. Letters represent homogeneous groups according to a Student–Newman–Keuls test.

![Figure 2](image2.png)

**Figure 2.** Effect of inoculation with Bt04, Pp20 and Lf89 on plant biomass of *D. stramonium* and *D. tatula* in two hydroponic systems. (a) Plants were 7 weeks old upon inoculation. Measurements were performed 2, 8 and 16 d.a.i. (days after inoculation) expressed as: plant length and total fresh weight. Each value is the mean of 10 replicates. Error bars represent ±standard deviation. (b) Leaf and root dry weight of *D. stramonium* plants after inoculation with Pp20 and Bt04. Plants were 7 weeks old upon inoculation. Values represent mean values of 10 biological replicates ±standard deviation. (c) Effect of inoculation with Pp20 and Lf89 on plant biomass of *D. tatula*. Measurements were performed at three time points (after 1, 2 and 3 weeks), expressed as: plant length, leaf number and root length. Each value is the mean of four replicates ±standard deviation. (d) Shoot and root dry weight of *D. tatula* plants after inoculation with Pp20 and Lf89. Values represent mean values of four biological replicates ±standard deviation. Letters represent homogeneous groups according to a Student–Newman–Keuls test.
of genes predicting adhesion proteins, detoxifying compounds, volatile components and enzymes such as cellulase, phytase and deaminase (Martínez-García et al. 2015). Previously described by Nishimori et al. (2000), it has a respiratory but not a fermentative metabolism and exhibits biofilm formation capacity. Jha et al. (2009) found that this genus can be used as biofertilizers as well as biocontrol agents because of the innate potential of phosphate solubilization, production of siderophores, IAA, protease, cellulase and HCN. Dharni et al. (2014) found that the P. plecoglossicida PsF610 increased the dry biomass of rose-scented geranium (Pelargonium graveolens cv. bourbon) shoots by 38%, roots by 40%, essential oil yield 39% and chlorophyll by 28%, respectively, over uninoculated controls.

(3) The genus Bacillus is a large and heterogeneous collection of a Gram-positive aerobic or facultatively anaerobic, rod-shaped, endospore-forming bacteria that are widely distributed in the environment. They tolerate various environmental conditions. The majority is mesophilic, with an optimal temperature of 30–45°C; however, some are thermophiles, growing at even 65°C, or psychrophiles, able to grow at 0°C. They are found to grow over a range of pH from 2 to 11. In general, Bacilli are capable of using simple organic compounds, such as amino acids, sugars, organic acids or carbohydrates, as well as various other substances. The Bacilli include species of industrial, biotechnological and environmental interest, as well as clinically important strains. In terms of metabolic properties, they present a diverse group, as they can degrade various substrates and produce many molecules, including lipopeptide biosurfactants, compounds composed of cyclic peptides linked to various fatty acids. Since this group of biosurfactants exhibits, besides the surface active properties, antibacterial, antifungal, antiviral and antitumor activities, they are commonly used in agriculture, food production, chemistry, cosmetics and pharmaceutics (Pacwa-Plociniczak et al. 2015).

The purpose of the study was to evaluate the growth-promoting effects of three strains of bacterial PGPR, P. plecoglossicida Pp20, Bacillus spec. Bt04 and L. fusiformis Lf89 on the growth of three plant species (A. thaliana, D. stramonium and D. tatula) in two different culture media (in vitro and hydroponics).

In the first assay with A. thaliana plants, significant improvement of root length, number of lateral roots, rosette diameter and dry root weight was recorded due to inoculation with Pp20, Bt04 and Lf89. Ryu et al. (2003) established a standardized in vitro PGPR test (I-plate system) for A. thaliana. This initial study assessed the effects on Arabidopsis growth promotion by B. subtilis GB03 and Bacillus amyloliquefaciens IN937a and showed a significant improvement in the growth of plants exposed to these bacteria, compared to controls (Ryu et al. 2003). Root length and shoot number were the best indicators of the response of Arabidopsis to inoculation with our strains.

Use of PGPRs as stimulants of seed germination in medicinal and aromatic species can provide more uniformity in germination, seedling emergence and other growth stages in particular flowering, which is a critical time to achieve more bioactive secondary metabolites (Ghorbanpour et al. 2015).

However, although growth promotion upon inoculation with our PGPR strains was demonstrated for two Datura species, we could not find major changes in the alkaloid content per gram dry weight in the roots. Since plants grew bigger, the alkaloid production on a per plant basis is therefore expected to increase and may specifically change shoot composition in the Bt04 interactions.

**Conclusion**

Three rhizobacterial strains were isolated from the Sahara (South) and the North of Algeria and were identified as P. plecoglossicida, Bacillus sp. and L. fusiformis strains by 16S rRNA gene sequence analysis. All strains increased growth of A. thaliana and two Datura species. Our studies showed that the utilization of beneficial PGPR isolates has great potential to stimulate the growth and development of plants (Datura and Arabidopsis) under different culture conditions. Additionally, an interesting shift in C/N ratio of plant material was observed for some strains. However, alkaloid production in D. stramonium showed only little differences after PGPR inoculation.

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**Disclosure statement**

No potential conflict of interest was reported by the authors.
References

Bertrand H, Nalin R, Bally R, Cleyet-Marel JC. 2001. Isolation and identification of the most efficient plant growth-promoting bacteria associated with canola (Brassica napus). Biol Fertil Soils. 33:152–156.

Bhatia S, Dubey RC, Maheshwari DK. 2005. Enhancement of plant growth and suppression of collar rot of sunflower caused by Sclerotium rolfsii through fluorescent Pseudomonas. Indian Phytopathol. 58:17–24.

Bhattacharyya P, Jha DK. 2015. Enhanced growth and suppression of collar rot of sunflower caused by Sclerotium rolfsii through fluorescent Pseudomonas. Indian Phytopathol. 58:17–24.

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References

Bertrand H, Nalin R, Bally R, Cleyet-Marel JC. 2001. Isolation and identification of the most efficient plant growth-promoting bacteria associated with canola (Brassica napus). Biol Fertil Soils. 33:152–156.

Bhatia S, Dubey RC, Maheshwari DK. 2005. Enhancement of plant growth and suppression of collar rot of sunflower caused by Sclerotium rolfsii through fluorescent Pseudomonas. Indian Phytopathol. 58:17–24.

Bhattacharyya P, Jha DK. 2015. Enhanced growth and suppression of collar rot of sunflower caused by Sclerotium rolfsii through fluorescent Pseudomonas. Indian Phytopathol. 58:17–24.

Bhattacharyya P, Jha DK. 2015. Enhanced growth and suppression of collar rot of sunflower caused by Sclerotium rolfsii through fluorescent Pseudomonas. Indian Phytopathol. 58:17–24.

Bhattacharyya P, Jha DK. 2015. Enhanced growth and suppression of collar rot of sunflower caused by Sclerotium rolfsii through fluorescent Pseudomonas. Indian Phytopathol. 58:17–24.

Bhattacharyya P, Jha DK. 2015. Enhanced growth and suppression of collar rot of sunflower caused by Sclerotium rolfsii through fluorescent Pseudomonas. Indian Phytopathol. 58:17–24.

Bhattacharyya P, Jha DK. 2015. Enhanced growth and suppression of collar rot of sunflower caused by Sclerotium rolfsii through fluorescent Pseudomonas. Indian Phytopathol. 58:17–24.

Bhattacharyya P, Jha DK. 2015. Enhanced growth and suppression of collar rot of sunflower caused by Sclerotium rolfsii through fluorescent Pseudomonas. Indian Phytopathol. 58:17–24.