Hoyosella rhizosphaerae sp. nov., a halotolerant actinobacterium isolated from rhizosphere soil of *Suaeda salsa*, and emended description of the genus *Hoyosella*

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A halotolerant actinobacterium, designated J12GA03^T, was isolated from a rhizosphere soil sample of *Suaeda salsa* collected from a dried saline lake in Hebei Province, China. Cells were Gram-staining-positive, non-motile and non-spore-forming cocci. Strain J12GA03^T grew optimally at 28–37 °C, 0–3 % NaCl (w/v) and pH 6.5–7.5. It contained *meso*-diaminopimelic acid as the diagnostic diamino acid and arabinose, galactose and ribose as the diagnostic whole-cell sugars. MK-8 and MK-7 were detected as predominant menaquinones. Major fatty acids were $C_{17:1}\omega8c$, $C_{16:0}$ and $C_{17:0}$. Polar lipids were phosphatidylethanolamine, diphosphatidylglycerol, phosphatidylinositol, phosphatidylglycerol, phosphoglycolipids, glycolipids, unidentified phospholipids and additional lipids. The muramyl residue was acetyl. Mycolic acids (34–38 carbon atoms) were present. The G+C content of the genomic DNA was 55.8 mol%. It shared the highest 16S rRNA gene sequence similarities with *Amycolicicoccus subflavus* DQS3-9A1^T (98.18 %) and *Hoyosella altamirensis* OFN S31^T (97.75 %). Phylogenetic trees showed that strain J12GA03^T firmly formed a distinct monophyletic branch in the clade with *A. subflavus* DQS3-9A1^T and *H. altamirensis* DSM 45258^T. The levels of DNA–DNA relatedness with *A. subflavus* DSM 45089^T and *H. altamirensis* DSM 45258^T were 39.7±3.9 % and 35.7±3.0 %, respectively. Combining the evidence from the polyphasic taxonomic study, strain J12GA03^T represents a novel species of the genus *Hoyosella*, for which the name *Hoyosella rhizosphaerae* sp. nov. is proposed. The type strain is J12GA03^T (=DSM 101985^T=CGMCC 1.15478^T).

The suborder *Corynebacterineae* was recast into the order *Corynebacteriales* in volume five of *Bergey's Manual of Systematic Bacteriology* (Goodfellow & Jones, 2012), which encompasses 16 wall chemotype IV genera with three mycolate-less wall chemotype IV taxa including the genera *Hoyosella* (Jurado *et al.*, 2009), *Amycolicicoccus* (Wang *et al.*, 2010) and *Turicella* (Funke *et al.*, 1994). Both the genera *Hoyosella* and *Amycolicicoccus* were seen to be phylogenetically close to species of the genus *Mycobacterium*. Meanwhile, the genus *Amycolicicoccus* was found to be highly related to the genus *Hoyosella*, with 99.77 % 16S rRNA gene sequence similarity between *Amycolicicoccus subflavus* DQS3-9A1^T (Wang *et al.*, 2010) and *Hoyosella altamirensis* OFN S31^T (Jurado *et al.*, 2009). This close taxonomic relationship is also clearly shown in the EzTaxon database (http://www.ezbiocloud.net/eztaxon/hierarchy?m=browse and k=Hoyosella and d=2) in that *A. subflavus* and *H. altamirensis* were assigned to one genus, the genus *Hoyosella*, since the name *Hoyosella* has the priority based on the earlier legitimate name (Jurado *et al.*, 2009) according to the International Code of Nomenclature of Bacteria (Lapage *et al.*, 1992).

During our previous bioprospecting for actinobacteria of pharmaceutical interest in saline lakes, a halotolerant strain, designated J12GA03^T, was isolated from a rhizosphere soil sample of *Suaeda salsa* collected from Jiuliancheng Nur (41° 33′ 58.94″ N 115° 00′ 19.86″ E), a dried saline lake located in Hebei Province, China. The isolate was found to be most

The GenBank/EMBL/DDBJ accession numbers for the 16S rRNA gene sequences of strain J12GA03^T and *H. altamirensis* DSM 45258^T are KU051698 and KX146464, respectively.

Two supplementary figures and four supplementary tables are available with the online Supplementary Material.

related to *A. subflavus* DQS3-9A1^T and *H. altamirensis* OFN S31^T. The results of phylogenetic analyses based on 16S rRNA gene sequences revealed that strain J12GA03^T represents a novel species affiliated within the genus *Hoyosella*, and the isolate was subjected to characterization by a polyphasic taxonomic approach.

The soil sample was diluted with sterile water and spread onto selective media for actinobacteria. The colony of strain J12GA03^T appeared on Gauze's medium no. one agar (containing l^{-1}: 20.0 g soluble starch, 1.0 g KNO_3, 0.5 g K_2HPO_4, 0.24 g $MgSO_4$, 0.5 g NaCl, 0.01 g $FeSO_4$, 18.0 g agar) supplemented with cycloheximide (45.0 mg l^{-1}), nalidixic acid (25.0 mg l^{-1}) and potassium dichromate (45.0 mg l^{-1}) after incubation for 5 weeks at 28 °C and was transferred onto International *Streptomyces* Project 2 (ISP 2, yeast extract-malt extract agar; Shirling & Gottlieb, 1966) plates using the serial streaking technique until pure isolates were obtained. The purified isolates were maintained on brain heart infusion agar (BHI agar; BD) slants at 4 °C and stored as aqueous glycerol suspensions (20 %, v/v) at −80 °C.

Gram staining was performed as described by Magee *et al.* (1975). Acid-fast staining was carried out using a modified Ziehl-Neelsen test with 3 % hydrochloric acid/alcohol mixture for decoloration. Cultural characteristics of strain J12GA03^T were observed following the growth on ISP 2, BHI agar, Bennett's agar (Atlas, 1993), tryptic soy agar (TSA; BD) and artificial seawater (ASW) (Wang *et al.*, 2010) agar plates at 28 °C for 3–15 days, and all the media were supplemented with 3 % NaCl (w/v). The colony colour and production of diffusible pigments were determined with chips from the ISCC-NBS colour charts (Kelly, 1964). Cell morphology was observed by light microscopy (BH2; Olympus) after incubation on BHI agar for 3 days and by field emission scanning electron microscopy (SU-8010; HITACHI) using gold-coated dehydrated specimens after incubation in BHI broth at 35 °C for 3 days.

Growth at different temperatures (4, 10, 15, 20, 25, 28, 35, 37, 42, 45, 50 °C) and NaCl concentrations (w/v) (0, 1, 2, 3, 4, 5, 7, 8, 10, 12, 15 %) was determined on BHI agar for 3–15 days, respectively. The pH range for growth was tested in ISP 2 broth supplemented with 3 % NaCl (w/v) at 35 °C between pH 4.0 and 11.0 at intervals of 0.5 pH unit using the buffer system described by Xu *et al.* (2005). Carbon source utilization, acid production from carbohydrates and enzyme activities were determined by using Biolog GEN III MicroPlates, API 50CH strips (bioMérieux) and API ZYM strips (bioMérieux), respectively. Oxidase activity was determined by the oxidation of tetramethyl-*p*-phenylenediamine. Catalase activity was determined using 3 % H_2O_2, and bubble production was scored as a positive reaction. Hydrolysis of cellulose, urea, starch, Tween 20, Tween 40 and Tween 80 was examined using BHI agar as basal medium. Gelatin liquefaction, H_2S production and nitrate reduction were determined as described by Williams *et al.* (1983).

Susceptibilities to 22 different antibiotics was assessed by the disc diffusion method. Strain J12GA03^T and reference type strains, *H. altamirensis* DSM 45258^T and *A. subflavus* DSM 45089^T, were inoculated on Mueller–Hinton agar (Oxoid) plates using the spread plate method with the bacterial suspension (approximately 10^5 c.f.u. ml^{-1}), and then the antibiotic discs were applied. The diameters of inhibition zones (mm) were measured after 3–4 days of incubation at 35 °C.

Strain J12GA03^T grew well on BHI agar, TSA and ISP 2, and moderately on Bennett's agar and ASW agar. The cells of strain J12GA03^T formed circular, raised and slightly dry colonies. Moderate or light yellowish-pink colonies occurred on BHI agar, ISP 2 and Bennett's agar, and pinkish-white colonies on TSA and ASW agar. The colour of the colonies was darker or lighter as the culture temperature became lower or higher, respectively. No diffusible pigment was observed on any media tested. The cells were Gram-staining-positive, acid-fast-staining-negative, non-motile, non-spore-forming and non-flagellum-forming cocci. Cells were arrayed singly, in pairs, in tetrads or in clusters. Cells were spherical and 0.7–0.9 μm in diameter, and binary fission was observed by scanning electron microscopy (Fig. 1). Growth occurred at the temperature of 20–42 °C (optimum, 28–37 °C), pH 5.5–9.5 (optimum pH 6.5–7.5) and in the presence of 0–8 % NaCl (w/v) (optimum, 0–3 %). Details of the physiological characteristics of strain J12GA03^T are given in Table 1 and in the species description. Results of screening of antibiotic susceptibilities for strain J12GA03^T,

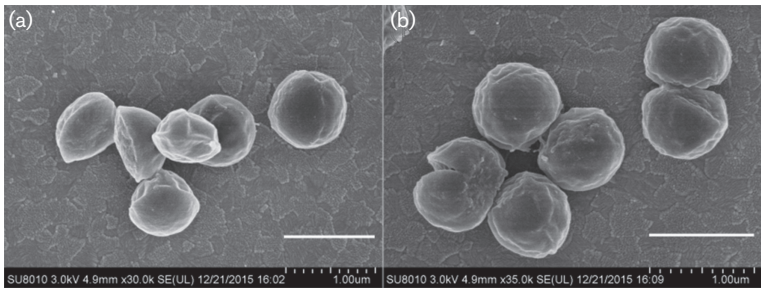

Fig. 1. Scanning electron micrographs of cells of strain J12GA03^T grown in BHI broth for 3 days at 35 °C; (a) ×30 000 magnification, (b) ×35 000 magnification. Bars, 1 μm.

Table 1. Differential characteristics of strain J12GA03^T and reference type strains

Strains: 1, J12GA03^T; 2, *A. subflavus* DSM 45089^T; 3, *H. altamirensis* DSM 45258^T. Data for all the strains were obtained in this study. +, Positive reaction; (+), weakly positive reaction; − , negative reaction. S, susceptible to antibiotic; R, resistant to antibiotic. All the strains were positive for catalase, nitrate reduction, urease activity and hydrolysis of Tween 40. All the strains were negative for oxidase activity, H_2S production, gelatin liquefaction, and hydrolysis of Tween 20 and cellulose. In the Biolog GEN III microplates, acetic acid, *N*-acetyl-D-glucosamine, *N*-acetyl-β-D-mannosamine, D-arabitol, cellobiose, citric acid, dextrin, D-fructose, L-fucose, D-fucose, D-fructose 6-phosphate, D-galactose, gentiobiose, α-D-glucose, D-glucose 6-phosphate, glycerol, *myo*-inositol (weakly), α-lactose (weakly), malic acid, maltose, D-mannitol (weakly), D-mannose, melibiose, pectin, propionic acid, L-rhamnose, D-sorbitol (weakly), trehalose and turanose could be utilized as carbon sources by all the strains, but inosine and salicin could not be utilized. In the API ZYM strips, all the strains were positive for acid phosphatase, alkaline phosphatase, cystine arylamidase (weakly), esterase (C 4), esterase lipase (C 8), α-glucosidase, leucine arylamidase, lipase (C 14), valine arylamidase (weakly) and naphthol-AS-BI-phosphohydrolase, and negative for trypsin, α-chymotrypsin, α-fucosidase, α-galactosidase, β-galactosidase, β-glucuronidase, *N*-acetyl-β-glucosaminidase and α-mannosidase.

Characteristic	1	2	3
Isolation source	Soil from a saline lake	Saline soil contaminated with crude oil	Biofilm in a cave
Isolation country	China	China	Spain
NaCl (w/v) concentration for growth	0–8 %	0–10 %	0–10 %
Hydrolysis of:			
Starch	−	(+)	+
Tween 80	−	(+)	(+)
Carbon sources utilization			
Sucrose	−	+	+
Stachyose	−	+	+
Raffinose	−	(+)	+
Acid produced from:			
D-Ribose, trehalose	+	−	−
Maltose, melezitose	−	(+)	+
Aesculin, xylitol, salicin	−	+	+
Sucrose, D-glucose	−	+	+
Turanose, D-arabitol	−	+	+
D-Arabinose, cellobiose	−	−	(+)
L-Arabinose	−	+	−
D-Tagatose	+	(+)	−
β-Glucosidase activity	−	+	+
Antibiotic susceptibilities			
Sulfamethoxazole (300 μg)	S	R	R
Sulfamethoxazole/ trimethoprim (23.75/1.25 μg)	S	R	R
Glycolipids (GLs)	GL1, GL2	GL1, GL2, GL3	GL1, GL2, GL3
Major menaquinones	MK-8 (62.6 %), MK-7 (36.3 %)	MK-8 (85.1 %), MK-7 (12.8 %)	MK-8 (86.4 %), MK-7 (11.3 %)
Major cellular fatty acids (>10 %)	$C_{17:1}\omega8c$, $C_{16:0}$, $C_{17:0}$	$C_{16:0}$, $C_{18:1}\omega9c$, $C_{17:1}\omega8c$, $C_{16:1}\omega7c$/ $C_{16:1}\omega6c$	$C_{16:0}$, $C_{17:1}\omega8c$, $C_{18:1}\omega9c$, $C_{16:1}\omega7c$/ $C_{16:1}\omega6c$
DNA G+C content (mol %)	55.8	58.1	59.8

H. altamirensis DSM 45258^T and *A. subflavus* DSM 45089^T are given in Table S1, available in the online Supplementary Material.

Biomass of strain J12GA03^T and two reference type strains (*H. altamirensis* DSM 45258^T and *A. subflavus* DSM 45089^T) for chemotaxonomic studies except fatty acid analysis was obtained after incubation in BHI broth supplemented with 3 % NaCl (w/v) at 35 °C for 3 days on a rotary shaker and harvested by centrifugation, then washed three times with distilled water. Diaminopimelic acid isomers and the whole-cell sugars were prepared and determined by TLC on aluminum cellulose sheets (Merck) as described by Staneck & Roberts (1974) and Hasegawa *et al.* (1983), respectively. The acyl type of the muramyl residue in peptidoglycan was determined by the glycolate test as reported by Uchida *et al.* (1999). Polar lipids were extracted, examined by two-dimensional TLC on silica gel 60 F_{254} plates (Merck) and identified using the procedures of Minnikin

et al. (1984). The solvent systems of the first and the second dimensions were chloroform/methanol/water (64 : 27 : 5, by vol.) and chloroform/methanol/acetic acid/water (80 : 18 : 12 : 5, by vol.), respectively. Menaquinone components were extracted as reported by Collins *et al.* (1977), then separated and determined by HPLC coupled with a single quadrupole mass spectrometer as described by Guo *et al.* (2015). For the analysis of cellular fatty acids, cells of strain J12GA03^T and the two reference type strains were harvested after cultivation on tryptic soy agar (BD) at 35 °C for 4 days. Cellular fatty acids were extracted according to the standard protocol of Sasser (1990), and the analyses of fatty acid methyl esters were performed according to the instructions of the Sherlock Microbial Identification System (MIDI) with MIDI Sherlock version 6.2 and library TSBA 6.21 employed. The analyses of mycolic acids for strain J12GA03^T and the two reference type strains were performed by the Identification Services, DSMZ, Braunschweig, Germany.

The whole-cell hydrolysates of strain J12GA03^T contained *meso*-diaminopimelic as the diagnostic diamino acid, and contained arabinose, galactose and ribose as the diagnostic whole-cell sugars (Fig. S1). The acyl type of muramyl residues in the peptidoglycan was acetylated. Strain J12GA03^T showed a polar lipid profile (Fig. S2) typical for the genus *Hoyosella*. Polar lipids comprised diphosphatidylglycerol, phosphatidylethanolamine, phosphatidylglycerol, phosphatidylinositol, two phosphoglycolipids (PGL1, PGL2), two glycolipids (GL1, GL2), two unidentified phospholipids (PL1, PL2) and several unidentified lipids. The menaquinone profile of strain J12GA03^T was composed of saturated menaquinones MK-8 (62.6 %), MK-7 (36.3 %) and MK-9 (1.1 %), which supported the affiliation within the genus *Hoyosella* and was similar to those of the two reference type strains containing MK-8 as the predominant menaquinone (Table S2). The fatty acid profile was composed predominantly of mono-unsaturated and saturated straight-chain fatty acids, and the major fatty acids were $C_{17:1}\omega8c$ (32.44 %), $C_{16:0}$ (22.15 %) and $C_{17:0}$ (14.24 %). The cellular fatty acid profiles of strain J12GA03^T and the two reference type strains are given in Table S3. Mycolic acids were detected in strain J12GA03^T, *H. altamirensis* DSM 45258^{T}and *A. subflavus* DSM 45089^T, and the overall size of the mycolic acids (number of carbons) for strain J12GA03^T was 34–38. Details of mycolic acid profiles of the three strains tested are shown in Table S4.

The extraction of genomic DNA from strain J12GA03^T and PCR amplification of the 16S rRNA gene were performed as described by Li *et al.* (2007). The purified PCR products were cloned using a pEASY-T1 Cloning kit (TransGen Biotech) following the manufacturer's instructions, and sequenced by Life Sciences Solutions Group, Thermo Fisher Scientific. The EzTaxon-e server (http://eztaxon-e.ezbiocloud.net/; Kim *et al.*, 2012) was employed to calculate the 16S rRNA gene sequence similarities between strain J12GA03^T and its near phylogenetic neighbours. Multiple alignments with the corresponding sequences obtained from the GenBank/EMBL/DDBJ databases were carried out using BioEdit (version 7.0.9.0) (Hall, 1999). The software package MEGA version 5.0 (Tamura *et al.*, 2011) was applied to reconstruct the phylogenetic trees with the neighbour-joining (Saitou & Nei, 1987), maximum-likelihood (Felsenstein, 1981) and maximum-parsimony (Fitch, 1971) algorithms. Evolutionary distance matrices was evaluated according to Kimura's two-parameter model (Kimura, 1980, 1983) for the neighbour-joining and maximum-likelihood algorithms, and close-neighbour interchange (search level=2, random addition=100) for the maximum-parsimony algorithm. The topologies of the phylogenetic trees were assessed by bootstrap analysis (Felsenstein, 1985) based on 1000 replications.

For the tests of genomic DNA G+C content and level of DNA–DNA relatedness, the genomic DNA of strain J12GA03^T and reference type strains were prepared according to the method described by Marmur (1961). The genomic DNA G+C content was determined using the thermal characterization of denaturation (Marmur & Doty, 1962) with *H. altamirensis* DSM 45258^T and *A. subflavus* DSM 45089^T as the references. DNA–DNA hybridizations were performed with *H. altamirensis* DSM 45258^T and *A. subflavus* DSM 45089^T by the thermal denaturation and renaturation method of De Ley *et al.* (1970) using a PharmaSpec UV/VIS spectrophotometer (UV-2550; Shimadzu) equipped with a Peltier-thermostatted multicell changer and a temperature controller (S-1700; Shimadzu) with *in-situ* temperature probe. The DNA–DNA hybridizations were conducted twice in each case with three replicates.

Comparative analyses with the nearly full-length (1483 bp) 16S rRNA gene sequence revealed that strain J12GA03^T shared the highest 16S rRNA gene sequence similarities with *A. subflavus* DQS3-9A1^T (98.18 %) and *H. altamirensis* OFN S31^T (97.75 %), followed by *Mycobacterium fallax* (95.60 %), *Mycobacterium sphagni* (95.17 %), *Mycobacterium goodii* (95.12 %) and *Mycobacterium anyangense* (95.10 %). Meanwhile, a nearly full-length (1483 bp) 16S rRNA gene sequence of *H. altamirensis* DSM 45258^T was obtained, which was identical to the sequence (FJ179485, 1333 bp) of *H. altamirensis* OFN S31^T reported by Jurado *et al.* (2009). The longer 16S rRNA gene sequence of *H. altamirensis* DSM 45258^T was applied in the reconstruction of phylogenetic trees. Three phylogenetic trees (Fig. 2) clearly displayed that strain J12GA03^T, *H. altamirensis* DSM 45258^T and *A. subflavus* DQS3-9A1^T clustered in a clade close to the cluster of the species of the genus *Mycobacterium*, and strain J12GA03^T formed a distinct monophyletic branch in the clade. The results of phylogenetic analysis indicated that the isolate is a novel member affiliated with the genus *Hoyosella*.

The G+C content of the genomic DNA of strain J12GA03^T was 55.8 mol%. The levels of DNA–DNA relatedness between strain J12GA03^T and *A. subflavus* DSM 45089^T, and between strain J12GA03^T and *H. altamirensis* DSM 45258^T were determined to be 39.7±3.9 % and 35.7±3.0 % (mean±SD), respectively, obviously below the 70 % cut-off value considered to be the threshold for

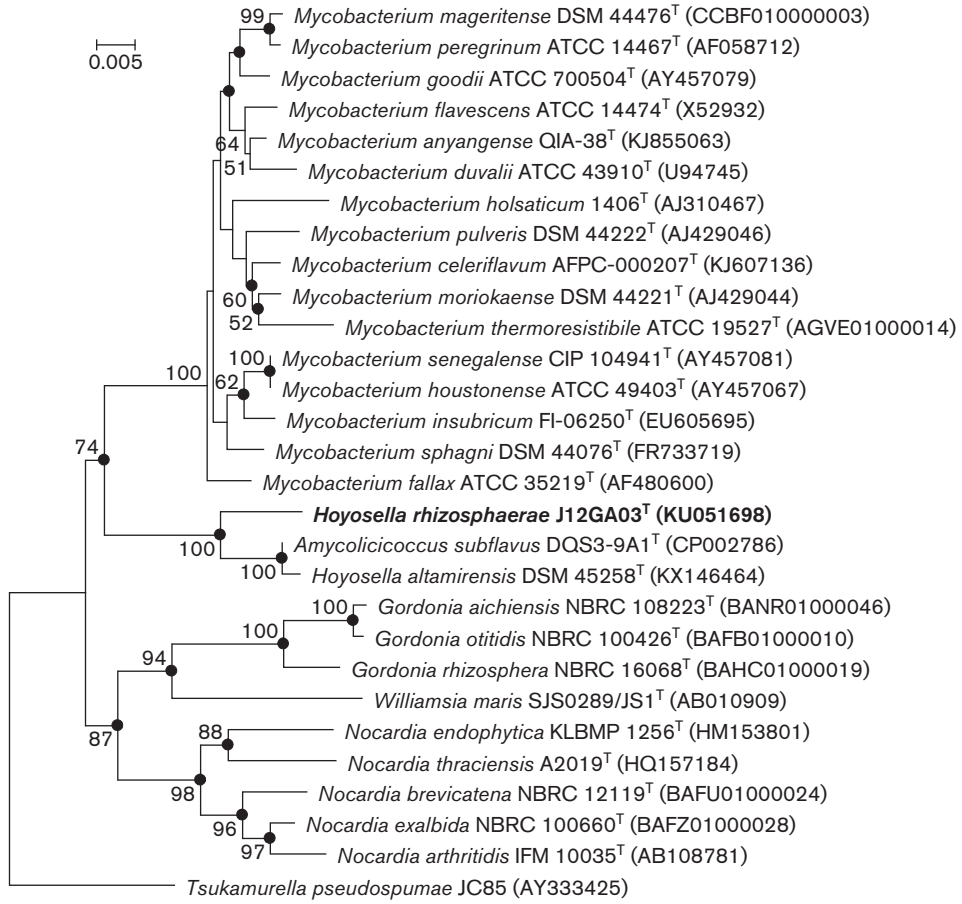

Fig. 2. Neighbour-joining tree showing the phylogenetic relationships between strain J12GA03^T and representatives of the order *Corynebacteriales* based on 16S rRNA gene sequences The sequence of *Tsukamurella pseudospumae* JC85 was used as the outgroup. Filled circles at nodes indicate corresponding branches that were also recovered by using the maximum-likelihood and maximum-parsimony algorithms. Bootstrap values >50 % based on 1000 replications are shown at branch nodes. Bar, 0.005 substitutions per nucleotide position.

the definition of genomic species (Wayne *et al.*, 1987). These values of DNA–DNA relatedness supported the affiliation of J12GA03^T as a representative of a novel species of the genus *Hoyosella*.

Affiliation of strain J12GA03^T to the genus *Hoyosella* was supported by the common chemotaxonomic characteristics including phosphatidylethanolamine, phosphatidylinositol, diphosphatidylglycerol and phosphatidylglycerol as the main phospholipids, MK-8 as predominant menaquinone, $C_{16:0}$ and $C_{17:1}\omega8c$ as major fatty acids, acetylated muramyl residue and C_{34}–C_{38} mycolic acids shared by *A. subflavus* DSM 45089^T and *H. altamirensis* DSM 45258^T (Fig. S2, Tables S2–S4). Meanwhile, strain J12GA03^T could be distinguished from *H. altamirensis* DSM 45258^T and *A. subflavus* DSM 45089^T in some chemotaxonomic characteristics. For instance, $C_{17:0}$ was detected as a major fatty acid for strain J12GA03^T, while $C_{18:1}\omega9c$ and summed feature 3 ($C_{16:1}\omega7c$/$C_{16:1}\omega6c$) were identified as major fatty acids for the two reference type strains, *A. subflavus* DSM 45089^T and *H. altamirensis* DSM 45258^T; a glycolipid (GL3) detected in *A. subflavus* DSM 45089^T and *H. altamirensis* DSM 45258^T was not found in strain J12GA03^T. Phenotypic characteristics that differentiate strain J12GA03^T from *H. altamirensis* DSM 45258^T and *A. subflavus* DSM 45089^{T}are summarized in Table 1.

Combining the evidence of phylogenetic analysis, phenotypic characteristics, chemotaxonomic data and DNA–DNA relatedness, strain J12GA03^T represents a novel species of the genus *Hoyosella*, for which the name *Hoyosella rhizosphaerae* sp. nov. is proposed.

Emended description of the genus *Hoyosella* Jurado *et al.* 2009

The genus description is as given by Jurado *et al.* (2009) with the following change. Mycolic acids (34–38 carbon atoms) are present.

Description of *Hoyosella rhizosphaerae* sp. nov.

Hoyosella rhizosphaerae (rhi.zo.sphae′rae. N.L. gen. n. *rhizosphaerae* of the rhizosphere).

Aerobic, halotolerant, Gram-staining-positive actinobacterium that forms no hyphae. No flagellum or spore is produced. Cells are spherical with a diameter of 0.7–0.9 μm and occur singly, in pairs, in tetrads or in clusters. Colonies are circular, raised and slightly dry, and the colours of the colonies are moderate or light yellowish-pink or pinkish-white. Cells grow well on BHI agar, TSA and ISP 2, and moderately on Bennett's agar and ASW agar. No diffusible pigment is observed. Growth occurs at 20–42 °C (optimum 28–37 °C), pH 5.5–9.5 (optimum pH 6.5–7.5) and in the presence of 0–8 % (w/v) NaCl (optimum 0–3 %). Hydrolysis of Tween 40, nitrate reduction, catalase and urease activities are positive. Hydrolysis of starch, cellulose, Tween 20 and Tween 80, oxidase activity, and H_2S production and gelatin liquefaction are negative. Acid phosphatase, alkaline phosphatase, cystine arylamidase (weakly), esterase (C 4), esterase lipase (C 8), α-glucosidase, leucine arylamidase, lipase (C 14), valine arylamidase (weakly) and naphthol-AS-BI-phosphohydrolase activities are positive. Trypsin, α-chymotrypsin, α-fucosidase, α-galactosidase, β-galactosidase, β-glucosidase, β-glucuronidase, *N*-acetyl-β-glucosaminidase and α-mannosidase are negative. Acetic acid, *N*-acetyl-D-glucosamine, *N*-acetyl-β-D-mannosamine, D-arabitol, cellobiose, citric acid, dextrin, D-fructose, D-fructose 6-phosphate, L-fucose, D-fucose, D-galactose, gentiobiose, α-D-glucose, D-glucose 6-phosphate, glycerol, *myo*-inositol (weakly), α-lactose (weakly), malic acid, maltose, D-mannitol (weakly), D-mannose, melibiose, pectin, propionic acid, L-rhamnose, D-sorbitol (weakly), trehalose and turanose can be utilized as carbon sources; but inosine, raffinose, salicin, stachyose and sucrose can not be utilized. Acids can be produced from D-glucose, glycerol, potassium 5-keto-gluconate, D-ribose, D-tagatose and trehalose. Susceptible to penicillin, ampicillin, amoxicillin, amoxicillin/clavulanic acid, cefalotin, cefotaxime, ceftazidime, cefepime, ceftriaxone, meropenem, amikacin, ciprofloxacin, sulfamethoxazole, sulfamethoxazole/trimethoprim, clarithromycin, chloramphenicol, gentamicin, erythromycin, vancomycin, rifampicin and ticarcillin/clavulanic acid, and resistant to clindamycin. Cells contain *meso*-diaminopimelic acid as the diagnostic diamino acid, and arabinose, galactose and ribose as the diagnostic whole-cell sugars. Polar lipids contain phosphatidylethanolamine, diphosphatidylglycerol, phosphatidylinositol, phosphatidylglycerol, phosphoglycolipids, glycolipids, unidentified phospholipids and additional lipids. The predominant menaquinones are MK-8 and MK-7. Major fatty acids are $C_{17:1}\omega8c$, $C_{16:0}$ and $C_{17:0}$. The muramyl residue is acetyl type. Mycolic acids (34–38 carbon atoms) are present.

The type strain, J12GA03^T (=DSM 101985^T=CGMCC 1.15478^T), was isolated from a rhizosphere soil sample of *Suaeda salsa* collected in Jiuliancheng Nur, a dried saline lake located in Hebei Province, China. The G+C content of the genomic DNA of the type strain is 55.8 mol%.

Acknowledgements

This research was partly supported by Science and Technology Research Program for Colleges and Universities in Hebei Province (grant no. QN2014078), IMB grant (grant no. IMBF201407), the Natural Sciences Foundation of China (NSFC, grant no. 81373308).

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