# Yania halotolerans gen. nov., sp. nov., a novel member of the suborder *Micrococcineae* from saline soil in China

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A novel coccoid, halotolerant actinobacterium, designated strain YIM 70085<sup>T</sup>, was isolated from a soil sample that was collected in Xinjiang Province, China, and characterized by using a polyphasic approach. Optimum growth temperature was 28 °C and growth occurred optimally in culture media that contained 10 % KCl. The peptidoglycan type was A4 $\alpha$ , L-Lys-Gly-L-Glu. Whole-cell sugars consisted of xylose, mannose and galactose. Phospholipids were diphosphatidylglycerol, phosphatidylglycerol, one unknown phospholipid, one unknown glycolipid and traces of phosphatidylinositol. Menaquinones were MK-8 (83%), MK-7 (12%) and MK-9 (15%). Predominant fatty acids were i-C<sub>15:0</sub> (44·29%), ai-C<sub>15:0</sub> (35·60%) and ai-C<sub>17:0</sub> (9·74%). The DNA G+C content was 53·5 mol%. Phylogenetic analysis based on 16S rRNA gene sequences showed that strain YIM 70085<sup>T</sup> occupies a branch that is distinct from, although very close to, the family *Micrococcaceae* in the suborder *Micrococcineae*. Based on its phenotypic characteristics, phylogenetic position (as determined by 16S rRNA gene sequence analysis) and 16S rDNA signature nucleotide data, it is concluded that the isolate represents a novel member of the suborder *Micrococcineae*, for which the name *Yania halotolerans* gen. nov., sp. nov. is proposed. The type strain is YIM 70085<sup>T</sup> (=CCTCC AA001023<sup>T</sup>=DSM 15476<sup>T</sup>).

Phylogenetic analysis based on 16S rRNA gene sequence data is currently one of the most effective methods for delineation of bacteria. For descriptions of actinomycete genera or species, organisms should also be characterized with respect to a rich spectrum of chemotaxonomic properties, such as profiles of fatty acids, polar lipids, peptidoglycan amino acids, isoprenoid quinones and, less frequently, polyamines (Stackebrandt & Schumann, 2000). For classification at higher taxonomic ranks of the classis *Actinobacteria*, i.e. families or suborders, 16S rDNA signature nucleotide data should also be used (Stackebrandt *et al.*, 1997; Stackebrandt & Schumann, 2000). Both a separate phylogenetic position and the emergence of novel signature patterns are indicative of a novel higher taxon (Stackebrandt & Schumann, 2000).

Until now, 13 families with validly published names have been described in the suborder *Micrococcineae*, including the family *Micrococcaceae* Pribram 1929 emend. Stackebrandt *et al.* 1997, the family *Brevibacteriaceae* Breed 1953 emend. Stackebrandt et al. 1997 and other families of Gram-positive cocci and rods. In this study, we report the results of polyphasic identification of a halotolerant actinobacterial strain, designated strain YIM 70085<sup>T</sup>, that was isolated from a saline soil sample in Xinjiang Province, west China. The strain grew optimally in media that contained 10% KCl, NaCl or MgCl<sub>2</sub>.6H<sub>2</sub>O. Based on the results of chemotaxonomic analyses, this strain is distinct from members of all actinomycete families in the suborder Micrococcineae. 16S rDNA sequence comparison revealed that strain YIM 70085<sup>T</sup> represents a distinct lineage within the suborder *Micrococcineae*, exhibiting < 94.3 % similarity to its phylogenetic neighbours. In addition, its 16S rDNA signature nucleotides are different from those of members of other families in the suborder Micrococcineae. Our results indicate that the isolate should be placed as a novel member of the suborder Micrococcineae: Yania halotolerans gen. nov., sp. nov. However, as the novel genus and species is represented only by a single strain, affiliation to a novel higher taxon should await phylogenetic analyses of further isolates of this genus. Strain YIM 70085<sup>T</sup> has been deposited in the China Center for Type Culture Collection as strain CCTCC AA001023<sup>T</sup> and in the Deutsche Sammlung von Mikroorganismen und Zellkulturen as DSM 15476<sup>T</sup>.

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The GenBank/EMBL/DDBJ accession number for the 16S rDNA sequence of strain YIM  $70085^{T}$  is AY228479.

## Micro-organisms and culture conditions

Strain YIM 70085<sup>T</sup> was isolated by using the dilution plating method from a saline soil sample from Xinjiang Province, west China. The medium used for selective isolation was glycerol/asparagine agar (ISP 5 medium; Shirling & Gottlieb, 1966) that was supplemented with 15% KCl (w/v) and incubated at 28 °C for about 2 weeks. The strain was maintained on potato agar or ISP 5 agar slants that contained 10% KCl (w/v) at 4°C and as glycerol suspensions (20%, v/v) at -20°C. Biomass for chemical and molecular systematic studies was obtained by cultivation in flasks of modified ISP 5 medium (10% KCl, w/v; pH 7·0) that were shaken (at about 150 r.p.m.) at 28 °C for 1 week.

## Morphological observations

Morphological properties were examined by light microscopy with a model BH-2 microscope (Olympus) and by transmission electron microscope with a model H-800 transmission electron microscope (Hitachi). Colour determination was done with colour chips from the ISCC-NBS Color Charts Standard Samples no. 2106 (Kelly, 1964). Morphological features were observed on glycerol/asparagine agar medium that was supplemented with 10 % (w/v) KCl.

Morphological observation of a 24–48 h culture of strain YIM 70085<sup>T</sup>, grown on glycerol/asparagine agar (with 10% KCl), revealed that cells were non-spore-forming, were coccoid or oval, were about  $0.4-0.7 \mu m$  in diameter and occurred singly or in clusters (Fig. 1). Colonies reached maximum size (2 mm in diameter) after 1 week incubation at 28 °C. They were yellow, circular, lubricious and opaque.



**Fig. 1.** Transmission electron micrograph of strain YIM 70085<sup>T</sup> grown on glycerol/asparagine agar (ISP 5 medium) with 10% KCl (w/v) for 2 days at 28 °C. Bar, 0.5  $\mu$ m.

# Metabolic properties

Gram-staining was carried out on 24–48 h cultures. Catalase activity was determined by production of bubbles after the addition of a drop of  $3 \% H_2O_2$ . All physiological and biochemical tests were performed at 28 °C. Carbon source utilization tests, sugar fermentation tests and qualitative enzyme tests were carried out in microtitre plates, according to Kämpfer *et al.* (1991). Some physiological properties were tested by using the API Coryne system and an API ID32 E test kit (bioMérieux).

Strain YIM 70085<sup>T</sup> was Gram-positive and aerobic; it utilized almost all carbon sources tested, but acid was produced only from glucose, maltose, sucrose and fructose. Its physiological and biochemical properties are given in detail in the species description.

## **Chemotaxonomic properties**

Sugar analysis of whole-cell hydrolysates followed procedures that were described by Stanek & Roberts (1974). Purified peptidoglycan preparations were obtained by the method of Schleifer & Kandler (1972). Amino acids and peptides in cell-wall hydrolysates were analysed by twodimensional ascending TLC on cellulose plates, using the solvent systems of Schleifer & Kandler (1972). The N-terminal amino acid of the interpeptide bridge was determined by dinitrophenylation, as described by Schleifer (1985). Molar ratios of amino acids were determined by GC and GC-mass spectrometry of N-heptafluorobutyryl amino acid isobutyl esters (MacKenzie, 1987). Analysis of enantiomers of peptidoglycan amino acids was performed by GC of N-pentafluoropropionyl amino acid isopropyl esters (Frank et al., 1980) on an L-Chirasil-Val column (Macherey-Nagel) as described by Groth et al. (1997).

Polar lipids were extracted, examined by two-dimensional TLC and identified by using published procedures (Minnikin *et al.*, 1979; Collins & Jones, 1980). Menaquinones were isolated by using the method of Collins *et al.* (1977) and analysed by HPLC (Groth *et al.*, 1997). Analysis of the whole-cell fatty acid pattern followed the MIDI system (Microbial ID) (Kroppenstedt, 1985; Meier *et al.*, 1993).

Enantiomeric analysis of peptidoglycan amino acids revealed equal amounts of L- and D-Glu. As D-Glu occupies position 2 of the peptide subunit, it was concluded that the N-terminal glutamic acid residue of the interpeptide bridge is of the L-configuration. Therefore, the peptidoglycan type of strain YIM 70085<sup>T</sup> was A4 $\alpha$ , L-Lys–Gly–L-Glu. Whole-cell sugars consisted of xylose, mannose and galactose. Phospholipids were diphosphatidylglycerol, phosphatidylglycerol, one unknown phospholipid (which was close to phosphatidylcholine, but its reaction with Dragendorff reagent was negative), one unknown glycolipid and traces of phosphatidylinositol. Menaquinones were MK-8 (83%), MK-7 (12%) and MK-9 (15%). Cellular fatty acid results are indicated in the species description and Table 1.

#### **Table 1.** Differential chemotaxonomic characteristics of strain YIM 70085<sup>T</sup> and related taxa

Taxa: 1, *Yania* gen. nov. (YIM 70085<sup>T</sup>); 2, *Brevibacteriaceae* (*Brevibacterium*); 3, *Micrococcus*; 4, *Arthrobacter*; 5, *Kocuria*; 6, *Nesterenkonia*; 7, *Renibacterium*; 8, *Rothia*; 9, *Citricoccus*. Abbreviations:  $A_2pm$ , diaminopimelic acid; MCA<sub>var</sub>, variable monocarboxylic amino acid in the interpeptide bridge; DMDG, dimannosyldiacylglycerol; DPG, diphosphatidylglycerol; GL, unidentified glycolipid; PE, phosphatidylethanolamine; PG, phosphatidylglycerol; PI, phosphatidylinositol; PIM, phosphatidylinositol mannosides; PL, unidentified phospholipid. Abbreviations for menaquinones are exemplified by MK-8(H<sub>2</sub>), a partially saturated menaquinone with one of eight isoprene units hydrogenated. Abbreviations for fatty acids are exemplified by ai-C<sub>15:0</sub>, 12-methyltetradecanoic acid; i-C<sub>15:0</sub>, 13-methyltetradecanoic acid; C<sub>16:0</sub>, hexadecanoic acid.

Characteristic	1	2*	Micrococcaceae								
			3†	<b>4</b> †	5†	<b>6</b> †	<b>7</b> †	<b>8</b> †	<b>9</b> ‡		
Diamino acid	L-Lys	<i>meso</i> -A <sub>2</sub> pm	L-Lys	L-Lys	L-Lys	L-Lys	L-Lys	L-Lys	L-Lys		
Interpeptide bridge	Gly–L-Glu	None	Peptide subunit or D-Asp	MCA <sub>var</sub>	L-Ala <sub>3-4</sub>	Gly–L-Glu	L-Ala–Gly	L-Ala <sub>3</sub>	Gly–L-Glu		
Predominant menaquinone(s)	7, 8, 9	8(H <sub>2</sub> )	8, 8(H <sub>2</sub> )	9(H <sub>2</sub> )/8, 9	7(H <sub>2</sub> ), 8(H <sub>2</sub> )	8, 9	9, 10	7	7(H <sub>2</sub> )/8(H <sub>2</sub> )/9(H <sub>2</sub> )		
Polar lipids	DPG, PG,	DPG, PG,	DPG, PI, PG,	DPG, PG,	DPG, PG,	DPG, PI,	DPG, GL	DPG, PG	DPG, PG,		
	PI, PL, GL	DMDG	PL, GL	PI, DMDG	(PI, L, GL)	PG, PL, GL			PI, PL, GL		
Major fatty acids	i-C <sub>15:0</sub> ,	ai-C <sub>15:0</sub> ,	i-C <sub>15:0</sub> ,	i-C <sub>15:0</sub> ,	ai-C <sub>15:0</sub> ,	ai-C <sub>15:0</sub> ,	ai-C <sub>15:0</sub> ,	ai-C <sub>15:0</sub> ,	ai-C <sub>15:0</sub> , ai-C <sub>17:0</sub> ,		
	ai-C <sub>15:0</sub> ,	ai-C <sub>17:0</sub>	ai-C <sub>15:0</sub>	ai-C <sub>15:0</sub> ,	ai-C <sub>17:0</sub> ,	ai-C <sub>17:0</sub> ,	ai-C <sub>17:0</sub>	ai-C <sub>17:0</sub> ,	i-C <sub>16:0</sub> , i-C <sub>15:0</sub>		
	ai-C <sub>17:0</sub>			i-C <sub>16:0</sub>	i-C <sub>16:0</sub>	i-C <sub>16:0</sub>		C <sub>16:0</sub>			
DNA G+C content (mol%)	53–54	60–67	69–76	56–69	66–75	70–72	52–54	49–53	68		

\*Data taken from Collins (2001).

†Data taken from Stackebrandt & Schumann (2000).

‡Data taken from Altenburger et al. (2002).

#### **Molecular analyses**

Extraction of genomic DNA and amplification of 16S rDNA were done as described by Cui *et al.* (2001). Multiple alignments of sequences from a broad selection of actinobacteria and calculations of sequence similarity levels were carried out by using CLUSTAL X (Thompson *et al.*, 1997). A phylogenetic tree was constructed by using the neighbourjoining method of Saitou & Nei (1987) from  $K_{nuc}$  values (Kimura, 1980, 1983). The topology of the phylogenetic tree was evaluated by the bootstrap resampling method of Felsenstein (1985) with 1000 replicates.

Chromosomal DNA for determination of base composition was prepared by following the method of Marmur (1961). The G+C content of the DNA was determined to be 53.5 mol% by using the thermal denaturation method of Marmur & Doty (1962).

Almost-complete 16S rDNA sequence data of strain YIM 70085<sup>T</sup> (1503 bp) were determined. BLAST search results of strain YIM 70085<sup>T</sup> came from GenBank/EMBL/DDBJ and the Protein Data Bank. Reference sequences that contained unidentified and unpublished sequences were not included (Fig. 2). After elimination of all sites for which nucleotides were not determined in any sequences, 1461 nt was compared. A tree depicting the phylogenetic relationships of strain YIM  $70085^{T}$  and its closest relatives is shown in Fig. 2. Strain YIM  $70085^{T}$  formed a distinct phylogenetic lineage between the families Micrococcaceae and Brevibacteriaceae in the suborder Micrococcineae, but was related much more closely to the Micrococcaceae. Highest sequence similarity values were found with sequences of species of the family Micrococcaceae, such as Arthrobacter cumminsii (X93354) (94.31%) Arthrobacter albus (AJ243421) (94·38%), Kocuria rosea (Y11330) (93.72%) and Kocuria polaris (AJ278868) (93.65%).

### **Taxonomic conclusion**

The results of 16S rDNA sequence comparisons clearly demonstrate that strain YIM 70085<sup>T</sup> is a member of the suborder *Micrococcineae*. Strain YIM 70085<sup>T</sup> has many unique 16S rDNA signature nucleotides, compared to other families of the suborder *Micrococcineae*, such as 140–223 (A–G), 142–221 (C–A), 615–625 (G–U), 839–874 (A–A) and 1134–1140 (A–U) (see Table 2). Thus, strain YIM 70085<sup>T</sup> may represent a novel family of the suborder *Micrococcineae*, based on the viewpoint of Stackebrandt *et al.* (1997).

From the phylogenetic tree, strain YIM 70085<sup>T</sup> formed a separate phylogenetic lineage between the families *Micro-coccaceae* and *Brevibacteriaceae*, but was related much more closely to the former. Sequence similarity levels between strain YIM 70085<sup>T</sup> and type strains of the suborder *Micro-coccineae* were <94.3%. In addition, some signature nucleotides of the 16S rDNA sequence of strain YIM 70085<sup>T</sup> were not included in those of the families *Micro-coccaceae* and *Brevibacteriaceae* (Stackebrandt *et al.*, 1997; Stackebrandt & Schumann, 2000). The 16S rDNA signature nucleotide data of strain YIM 70085<sup>T</sup> are shown in Table 2. The results of comparison of 16S rDNA signature nucleotides between the two families and strain YIM 70085<sup>T</sup> are indicated in bold type (see Table 2).

Additionally, there are significant differences in chemotaxonomic characteristics between strain YIM 70085<sup>T</sup> and the phylogenetically most closely related genera of the families *Micrococcaceae* and *Brevibacteriaceae* (see Table 1). The type genus of the family *Brevibacteriaceae*, *Brevibacterium*, obviously differs from strain YIM 70085<sup>T</sup> in displaying *meso*-A<sub>2</sub>pm as the diagnostic diamino acid of the peptidoglycan and partially saturated menaquinones. Members of the genera *Micrococcus, Arthrobacter* and



**Fig. 2.** Phylogenetic dendrogram obtained by distance matrix analysis of 16S rDNA sequences, showing the position of strain YIM 70085<sup>T</sup> among its phylogenetic neighbours. Numbers on branch nodes are bootstrap values (percentages of 1000 resamplings; only values >50 % are given). The sequence of *Streptomyces megasporus* DSM 41476<sup>T</sup> (GenBank accession no. Z68100) was used as root. Bar, 1% sequence divergence.

0.01

#### Table 2. Patterns of selected 16S rDNA signature nucleotides that define families of the suborder Micrococcineae

Families: 1, Dermacoccaceae; 2, Rarobacteraceae; 3, Sanguibacteraceae; 4, Bogoriellaceae; 5, Dermatophilaceae; 6, Cellulomonadaceae; 7, Intrasporangiaceae; 8, Dermabacteraceae; 9, Jonesiaceae; 10, Microbacteriaceae; 11, Promicromonosporaceae; 12, Brevibacteriaceae; 13, Micrococcaceae; 14, Yania gen. nov. Data for Yania gen. nov. are from this study; data for all other taxa are from Stackebrandt & Schumann (2000). R, Purine; Y, pyrimidine; V, variable. Residues in small capitals are present in some, but not all, strains. The results of comparison of signature nucleotides between the families *Brevibacteriaceae* and *Micrococcaceae* and strain YIM 70085<sup>T</sup> are indicated in bold type.

Position	1	2	3	4	5	6	7	8	9	10	11	12	13	14
41-401	G–C	G–C	G–C	G–C	G–C	G–C	G–C	G–C	G–C	G–C	G–C	U–A	G–C	G–C
45-396	U–G	U–G	U–G	U–G	U–G	U–G	U–G	U–G	U–G	U–G	U–G	U–G	U–G	U–G
69–99	R–U	A–U	G–U	A–U	A–U	G–U	G–U	A–U	A–U	R–U	G–U	C–U	A–U	C–U
144-178	C–G	U–G	C–G	U–G	U–A	C–G	C–G	U–G	U–G	C–G	U–G	U–G	C–G	C–G
140-223	C–G, G–C	C–G	G–C	G–U	C–G	C–G	G–Y	C–G	U–U	G–Y	C–G	G–C	R–U	A–G
142-221	C–G	C–G	C–G	C–G	C–G	C–G	C–G	C–G	C–G	U–A	C–G	U–A	C–G	C–A
248-276	C–G	C–G	C–G	C–G	C–G	C–G	C–G	U–G	C–G	C–G	C–G	C-C	C–G	U–G
258-268	G–C	G–C	A–U	G–C	A–U	G–C	A–U	A–U	G–C	A–U	G–C	G–C	A–U	G–C
293-304	G–Y	G–C	G–C	G–U	G–U	G–C	G–C	G–C	G–C	G–U	G–C	G–C	G–U	G–U
379-384	C–G	C–G	C–G	C–G	C–G	C–G	C–G	C–G	G–U	C–G	C–G	G–C	C–G	C–G
407-435	A–U, G–C	A–U	A–U	A–U	A–U	C–G	A–U	G–Y	G–C	A–U	A–U	C–G	A–U	A–U
502-543	A–U	G–C	G–C	G–C	A–U	G–c	A–U	R–Y	G–C	R–Y	G–C	A–U	R–Y	A–U
586-755	C–G	U–G	C–G	C–G	C–G	C–G	Y–R	U–A	C–G	C–G	C–G	U–A	C-G	C–G
589–650	U–A	U–A	U–A	C–G	U–G	U–A	U–A	U–G	U–G	U–A	U–A	U–A	C–G	U–G
591-648	U–A	U–A	U–A	U–A	U–A	U–A	U–A	U–A	U–A	U–A	U–A	G–U	U–A	U–A
610	R	А	U	А	А	А	А	А	U	V	U	Α	G	G
602–636	C–G	G–U	G–U	C–G	C–G	C–G	C–G	C–G	U–G	C–G	G–U	C–G	C-G	C–G
612–628	Y–G	U–A	U–A	C–G	C–G	C–G	C–G	Y–G	C–G	C–G	C–G	G-C	C–G	U–A
615-625	G–C	C–G	A–U	G–C	G–C	A–U	R–Y	A–U	A–U	A–U	U–A	A–U	G–C	G–U
616–624	G–Y	G–C	U–G	G–C	G–U	C–G	G-Y	G–C						
660–745	G–C	G–C	G–C	G–C	G–C	G–C	G–C	G–C	G–C	G–C	U–A	A–U	v	A–U
668–738	A–U	A–U	U–A	A–U	A–U	U–A	A–U	A–U	U–A	A–U	A–U	A–U	A–U	A–U
670–736	A–U	A–U	A–U	A–U	A–U	A–U	A–U	A–U	A–U	A–U	A–U	U–A	A–U	A–U
839-847	U–A	U–A	U–A	C–G	C–G	C–G	U–A	R–U	U–A	G–U	C–G	A–U	A–U	A–A
863	U	А	U	U	U	А	U	U	U	U	U	U	U	U
1133–1141	A–U	G–C	A–U	A–U	A–U	G–C	A–U	A–U	A–U	A–U	G–C	A–U	A–U	A–U
1134-1140	C–G	G–C	C–G	C–G	C–G	G–C	C–G	C–G	C–G	C–G	G–C	C–G	C-G	A–U
1244-1293	C–G	C–G	C–G	C–G	C–G	C–G	C–G	C–G	C–G	C–G	C–G	U–A	C-G	C–G
1254-1283	G–C	G–C	G–C	G–C	U–A	G–C	G–C	G–C	G–C	G–C	G–C	A–C	G–C	G–C
1263-1272	A–U	A–U	A–U	A–U	A–U	A–U	A–U	A–U	A–U	A–U	A–U	C–G	A–U	A–U
1310-1327	G–C	G–C	G–C	G–C	G–C	G–C	G–C	G–C	G–C	A–U	G–C	U–A	R–Y	G–C
1414-1486	C–G	C–G	C–G	C–G	C–G	U–G	C–G	C–G	C–G	U–A	C–G	C-G	C–G	C–G

*Kocuria* in the family *Micrococcaceae* show highest 16S rDNA sequence similarity to strain YIM 70085<sup>T</sup>, but they can be differentiated easily, e.g. by partially saturated menaquinones, higher DNA G+C contents and different interpeptide bridges of the peptidoglycan. Peptidoglycan type L-Lys–Gly–L-Glu was found previously in the type strain of *Nesterenkonia halobia*. However, the combination of this peptidoglycan type with the menaquinone profile and polar lipid and fatty acid patterns of strain YIM 70085<sup>T</sup>, as well as its G+C content, are unique among members of the suborder *Micrococcineae*.

Thus, based on these results, which show that strain YIM 70085<sup>T</sup> differs from all established genera of the suborder *Micrococcineae* in rDNA signature nucleotides, 16S rDNA

sequence and phenotypic characteristics, a novel genus and species, *Yania halotolerans* gen. nov., sp. nov., are proposed to accommodate isolate YIM 70085<sup>T</sup>. However, as the novel genus and species are represented only by a single strain, affiliation to a novel higher taxon should await phylogenetic analyses of further isolates of this genus.

#### Description of Yania gen. nov.

*Yania* [Ya'ni.a. N.L. fem. n. *Yania* named after Sun-Chu Yan (1912–1994), a Chinese microbiologist who devoted his life to the study of actinomycete taxonomy and antibiotics].

Cells are non-motile, aerobic, Gram-positive, non-sporeforming, coccoid or oval and about  $0.4-0.7 \mu m$  in diameter and occur singly or in clusters. Oxidase-negative and catalase-positive. Cell-wall peptidoglycan type is A4 $\alpha$ , L-Lys–Gly–L-Glu. Whole-cell sugars are xylose, mannose and galactose. Phospholipids are diphosphatidylglycerol, phosphatidylglycerol, one unknown phospholipid, one unknown glycolipid and traces of phosphatidylinositol. Predominant menaquinones are MK-8, MK-7 and MK-9. Major cellular fatty acids are i-C<sub>15:0</sub>, ai-C<sub>15:0</sub> and ai-C<sub>17:0</sub>. DNA G+C content is 53–54 mol%. The type species is *Yania halotolerans*.

#### Description of Yania halotolerans sp. nov.

Yania halotolerans (ha.lo.to'le.rans. Gr. n. hals salt; L. pres. part. tolerans tolerating; N.L. part. adj. halotolerans referring to the organism's ability to tolerate high salt concentrations).

Morphological, chemotaxonomic and general characteristics are as described for the genus. Colonies reach maximum size (2 mm in diameter) after 1 week incubation at 28 °C. They are yellow, circular, lubricious and opaque. Almost all carbon sources tested are utilized, including glucose, galactose, arabinose, starch, cellobiose, lactose, mannose, mannitol, fructose, sucrose, maltose and xylose, but acid is produced only from glucose, maltose, sucrose and fructose. Positive for milk peptonization and urease, but negative for milk coagulation, nitrate reduction, gelatin liquefaction, growth on cellulose and production of H<sub>2</sub>S and melanin. Starch and Tweens 20, 40 and 80 are not hydrolysed. Cells do not form indole; methyl red and Voges-Proskauer tests are negative. Temperature range for growth is 10-40 °C, with an optimum temperature of 28-30 °C. Growth pH is optimal at 7.0-8.0. Concentration ranges of NaCl, KCl and MgCl<sub>2</sub>.6H<sub>2</sub>O for growth are 0-25, 0-20 and 0–15 %, respectively. Cell-wall peptidoglycan type is A4 $\alpha$ , L-Lys-Gly-L-Glu. Whole-cell sugars consist of glucose, arabinose, xylose and ribose. Phospholipids are diphosphatidylglycerol, phosphatidylglycerol, one unknown phospholipid, one unknown glycolipid and traces of phosphatidylinositol. Menaquinones are MK-8 (83%), MK-7 (12%) and MK-9 (15%). Cellular fatty acids are  $i-C_{15:0}$  (44·29%),  $ai-C_{15:0}$  (35·60%),  $ai-C_{17:0}$  (9·74%),  $C_{15:0}$  (0.28%),  $C_{16:0}$  (0.87%), i- $C_{14:0}$  (1.55%), i- $C_{16:0}$ (3.35%), i-C<sub>17:0</sub> (2.76%), ai-C<sub>19:0</sub> (0.42%), C<sub>16:1</sub> $\omega7c$ (0.48%) and  $i-C_{15.1}$  (0.65%). DNA G+C content is 53.5 mol%.

The type strain is YIM  $70085^{T}$  (=CCTCC AA001023<sup>T</sup> = DSM 15476<sup>T</sup>). Isolated from saline soil that was collected in Xinjiang Province, west China.

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