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Salinispora pacifica sp. nov., an actinomycete from marine sediments

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Abstract A polyphasic analysis was carried out to clarify the taxonomic status of four marine actinomycete strains that share a phylogenetic relationship and phenotypic characteristics with the genus *Salinispora*. These strains formed a distinct lineage within the *Salinispora* 16S rRNA and *gyrB* trees and were found to possess a range of phenotypic properties and DNA:DNA hybridization values that distinguished them from the type strains of the two validly named species in this genus, *Salinispora tropica* (CNB-440^T, ATCC BAA-916^T) and *Salinispora arenicola* (CNH-643^T, ATCC BAA-917^T). The combined genotypic and phenotypic data support this conclusion. It is proposed that the strains be designated as *Salinispora pacifica* sp. nov., the type strain of which is CNR-114^T (DSMZ YYYYT = KACC 17160^T).

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Introduction

The genus *Salinispora* is among a small but growing number of actinomycete genera that have been reported from marine sources (Han et al. 2003; Yi et al. 2004; Maldonado et al. 2005; Tian et al. 2009). Unlike other marine-derived genera described to date, members fail to grow when seawater is replaced with deionized water in the growth medium. The genus is currently composed of two species, *Salinispora arenicola* and *Salinispora tropica*, which can be distinguished from one another using a combination of chemical, genetic, and phenotypic properties (Maldonado et al. 2005). *Salinispora* strains have a cosmopolitan distribution in tropical and subtropical marine sediments (Jensen and Mafnas 2006; Freel et al. 2012) and have also been reported from a marine sponge (Kim et al. 2005). Although their occurrence in more temperate habitats has been observed when culture independent methods are applied (unpublished data), they have yet to be cultured from these environments.

Members of the genus *Salinispora* are a rich source of secondary metabolites (Fenical and Jensen 2006) including salinosporamide A, which is currently in clinical trials for the treatment of cancer (Fenical et al. 2009). The two validly named species devote a large

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percentage of their genomes to the biosynthesis of secondary metabolites (Penn et al. 2009), which are produced in species-specific patterns (Jensen et al. 2007). In addition to these two species, a new 16S rRNA phylotype, for which the name "*Salinispora pacifica*" was proposed, was cultured from marine sediments collected off Guam (Jensen and Mafnas 2006). Subsequently, additional strains belonging to this lineage were cultured from marine sediments collected around the islands of Palau (Gontang et al. 2007) and Fiji (Freel et al. 2012). This candidate species has greater phylogenetic diversity than the two validly published species and forms a sister lineage to *S. tropica* (Jensen et al. 2007). The proposed type strain produces the secondary metabolite cyanosporoside A (Oh et al. 2006), which to date has not been observed from the other two *Salinispora* spp. The present study employed a polyphasic approach to establish the taxonomic status of representative isolates within this new *Salinispora* phylotype. The resultant data show that the strains form a novel taxon, for which the name *S. pacifica* sp. nov. is proposed.

Materials and methods

The strains used in this study were cultured from marine sediment samples collected from the islands of Guam in 2002 (CNR-114^T) and Palau in 2004 (CNS-055, CNS-143, CNS-237) as previously described in Jensen et al. (2005), and Gontang et al. (2007), respectively. The strains were isolated using a heat shock method of 55 °C for 6 min followed by dilution plating onto seawater agar (Gontang et al. 2007). The strains were maintained on A1 agar (1.0 % starch, 0.4 % yeast extract, 0.2 % peptone, 1 liter seawater) and stored frozen at −80 °C in A1 supplemented with 10 % glycerol. Biomass for DNA:DNA relatedness and gene sequencing was prepared by growing the strains in A1 broth with shaking at 230 rpm for 10–14 days at 27 °C prior to harvesting by centrifugation. Genomic DNA extraction, PCR amplification, and the sequencing of 16S rRNA and *gyrB* genes were carried out as previously described (Jensen and Mafnas 2006). Sequences were checked for accurate base calling using Sequencher (version 4.5, Gene Codes Corp., Ann Arbor, MI), aligned using Clustal X (Thompson et al. 1997) and imported into MacClade (version 4.07, Sinauer Associates, Sunderland, MA) for manual alignment. Sequence similarities were calculated using various NCBI (National Center for Biotechnology Information) and BLAST (Basic Alignment Search Tool) functions. Neighbour-joining, maximum-parsimony and bootstrap analyses were performed using PAUP (version 4.0b10, Sinauer Assoc., Sunderland, MA). Sequence data have been deposited in the GenBank database (http://www.ncbi.nlm.nih.gov/Genbank/index.html) for strains CNR-114^T, CNS-237, CNS-055, and CNS-143 under the accession numbers DQ224161, DQ318246, DQ224159, and DQ92624 for 16S rRNA genes and DQ228686, JN032130, DQ22869, and DQ228693 for *gyrB* genes, respectively. DNA:DNA relatedness studies were carried out between strain CNR-114^T, *S. arenicola* CNH-643^T and *S. tropica* CNB-440^T, respectively using the identification service at the Deutsche Sammlung von Mikroorganismen und Zellkulturen (Braunschweig, Germany) as described by Kim et al. (1999).

Cultural, morphological and staining properties

The strains were examined for cultural and morphological features following growth for 3 weeks at 28 °C on glycerol-asparagine agar (ISP [International *Streptomyces* Project] medium 5), oatmeal agar (ISP medium 3), inorganic salts-starch agar (ISP medium 4), peptone-yeast extract agar (ISP medium 1) and yeast extract-malt extract agar (ISP medium 2) (Shirling and Gottlieb, 1966) prepared using 75 % seawater prepared using Instant Ocean synthetic sea salt (Aquarium Systems Sarrebourg, France). The spore arrangement and spore surface ornamentation of isolate CNR-114^T were observed by examining gold coated, dehydrated specimens prepared from an ISP medium 2 agar plate using a scanning electron microscope (Cambridge Stereoscan 240) and the procedure described by O'Donnell et al. (1993). Gram reaction and Ziehl-Neelsen preparations were examined by light microscopy following growth on ISP 2 medium agar as in an earlier study (Maldonado et al. 2005).

Phenotypic properties

The strains were examined for their ability to hydrolyse aesculin, allantoin and urea, to reduce nitrate, to degrade adenine, DNA, elastin, guanine, hypoxanthine, starch, Tween 60, uric acid, xanthine and xylan, and for their capacity to use sole nitrogen sources

using the media and methods described by Williams et al. (1983). They were tested for the ability to use a range of sole carbon compounds using the basal medium of Stevenson (1967), for tolerance to temperature, pH and sodium chloride using ISP medium 4 agar and for their sensitivity to antibiotics using glucose-yeast extract agar (Gordon and Mihn 1962) as the basal medium. All media were prepared using 75 % seawater and tests examined weekly for up to 4 weeks, as appropriate; all of the tests were incubated at 28 °C, apart from the temperature tests. Constituent enzyme activities of the strains were determined using API ZYM strips (BioMerieux) according to the manufacturer's instructions; a standard inoculum equivalent to 2.5 on the McFarland scale (Murray et al. 1999) was used to inoculate the strips. Assimilation of a broad range of substrates was determined using Biolog GP2 plates. To this end, a mixture of black spores and mycelial fragments of each of the strains was obtained by centrifugation of modified Bennett's broths (Jones 1949) prepared using 75 % seawater, following incubation at 28 °C for 14 days; the resultant biomass was washed in sterile distilled water, centrifuged and then suspended in Biolog inoculating fluid to give a standard suspension equivalent to McFarland scale 5. Aliquots of the resultant suspension (150 µl) were pipetted into the wells of the microplates which were incubated at 28 °C for up to 7 days when the plates were read visually to detect the pink colour changes which indicated positive results.

Chemotaxonomy

Biomass for all but one of the chemotaxonomic procedures was harvested by centrifugation, from strains grown for 5 days at 28 °C in shake flasks of M1 broth, washed twice in distilled water and freeze-dried. Strains CNS-237, CNR-114^T and DSM 44818^T were examined using standard methods to determine the diagnostic isomers of diaminopimelic acid (A_2pm; Staneck and Roberts 1974), isoprenoid quinones (Collins 1994), muramic acid type (Uchida et al. 1999), mycolic acids (Minnikin et al. 1975), polar lipids (Minnikin et al. 1984) and whole-organism sugars (Hasegawa et al. 1983). Biomass for the fatty acid analyses on all of the strains was harvested from shake flasks of Trypticase Soy broth (Difco), prepared with 75 % seawater, and incubated at 28 °C for 14 days. Fatty acids extracted from the resultant preparations were methylated and analysed by gas liquid chromatography using the standard Sherlock Microbial Identification (MIDI) system and the ACTINO 5 database (Sasser 1990).

The G + C values of the DNA of isolates CNR-114^T and CNS-055 were derived from nearly complete genome sequences generated by the Joint Genome Institute (http://genome.jgi.doe.gov/) using the Illumina platform.

Results and discussion

The two validly named species within the genus *Salinispora* have distinct biogeographical distributions in marine sediments, with *S. arenicola* being recovered from global collection sites while *S. tropica*, to date, has only been recovered from Caribbean locations (Jensen and Mafnas 2006; Freel et al. 2012). The present investigation included strains CNR-114^T, CNS-055, CNS-143, and CNS-237, which were recovered from marine sediments collected from tropical Pacific Ocean locations off the islands of Palau and Guam. These strains form a distinct phylogenetic lineage that is a sister to *S. tropica*. Based on DNA:DNA hybridization values, they were proposed to represent a new species (Jensen and Mafnas 2006). Strains within this lineage have been recovered from sites throughout the Pacific Ocean, the Red Sea, and the Sea of Cortez (Becerril-Espinosa et al. 2012; Freel et al. 2012) and display considerable phylogenetic diversity relative to the two validly named species (Freel et al. 2012). These strains originate from collection sites that also yielded *S. arenicola* but not *S. tropica*, providing further evidence for geographical isolation between *S. tropica* and the strains described in this paper.

Phylogenetic analyses

The 16S rRNA gene sequences for strains CNR-114^T, CNS-055, CNS-143, and CNS-237 were aligned to those for *Escherichia coli* (accession number J01695) and the type strains for *S. tropica* (CNB-440^T, ATCC BAA-916^T) and *S. arenicola* (CNH-643^T, ATTC BAA-917^T). This alignment, with common gaps removed, revealed that strains CNR-114^T and CNS-143 are clonal at the 16S rRNA gene level, while strains CNS-055 and CNS-237 represent sequence variants that have been designated as phylotypes "A" and "B", respectively. Of the total 1,479 nucleotide positions

examined, the three sequence types have four polymorphic positions. When considering all 16S rRNA gene sequence types observed to date for this lineage at least 13 variable nucleotide positions have been documented (Freel et al. 2012), which is considerably greater than that observed for *S. tropica* (no polymorphic sites) or *S. arenicola* (seven polymorphic sites). There are three clade-specific signature nucleotides associated with the four new strains. These consist of C, A, and A at positions 221, 234, and 264 in the alignment, while in both *S. tropica* and *S. arenicola* the respective nucleotides are T, G, and G (Freel et al. 2012). Strain CNR-114^T shares all but 11 of 1,479 nucleotide positions (99.3 % 16S rRNA gene sequence identity) with the *S. arenicola* type strain and all but six nucleotides (99.6 % 16S sequence identity) with the *S. tropica* type strain, while the *S. arenicola* and *S. tropica* type strains share 99.5 % sequence identity.

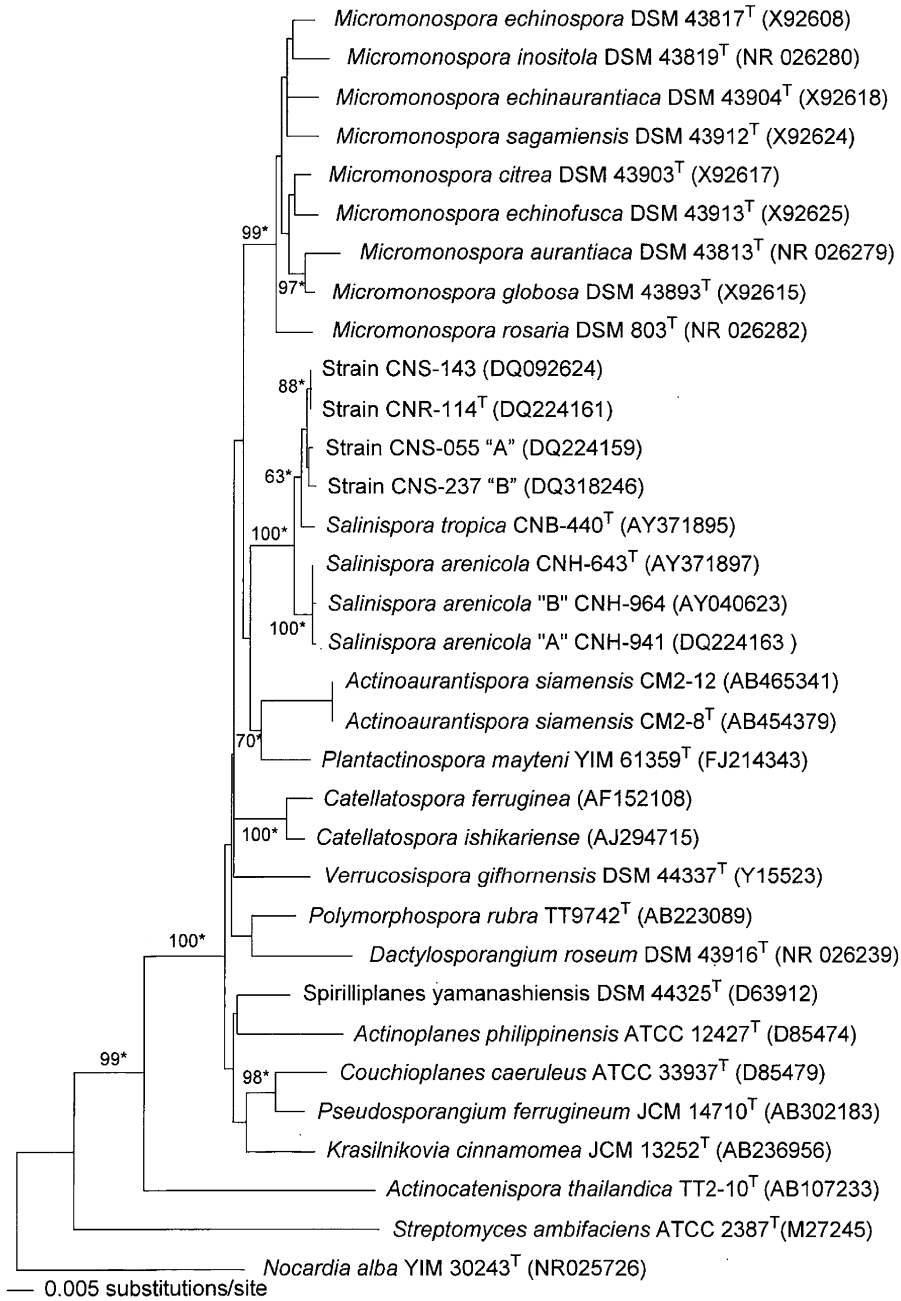

Fig. 1 Neighbour-joining phylogenetic tree (Saitou and Nei 1987) created from 33 nearly complete (1,359 nucleotides) 16S rRNA gene sequences showing relationships between strains CNS-143, CNR-114^T, CNS-055, and CNS-237 and representatives of the family *Micromonosporaceae*. *Asterisks* indicate branches of the tree also found using the unweighted pair group method with arithmetic means (Sneath and Sokal 1973), maximum-likelihood (Felsenstein 1981), and maximum-parsimony (Kluge and Farris 1969) methods. Numbers at nodes indicate the level of bootstrap support (%) based on a neighbour-joining analysis of 1,000 re-sampled datasets; only values above 55 % are shown

A phylogenetic tree based on 16S rRNA gene sequences clearly places strains CNR-114^T, CNS-055, CNS-143, and CNS-237 within the genus *Salinispora* (Fig. 1). Interestingly, the recently described genera *Actinoaurantispora* (Thawai et al. 2009) and *Plantactinospora* (Qin et al. 2009), neither of which were derived from marine sources, form a sister lineage with the genus *Salinispora* (Fig. 1) replacing *Micromonospora* as the most closely related taxon and filling in the taxonomic space between these genera. However, the poor bootstrap support for the clade that includes the genera *Salinispora*, *Actinoaurantispora,* and *Plantactinospora* suggests that these relationships may change as additional sequence data become available. Although the clade formed by the four new strains was separated from *S. tropica*, the weak bootstrap support at this branch point warranted additional studies. For this reason, a second analysis using the *gyrB* housekeeping gene was performed. This locus is known to provide good resolution within members of the family *Micromonosporaceae* (Kasai et al. 2000). The *gyrB* gene tree (Fig. 2) shows strong support for the phylogenetic separation of *S. tropica* (CNB-440) and the four new sequences using multiple treeing methods and thus supports the sister relationship of these two lineages to *S. arenicola*.

DNA:DNA hybridization experiments were carried out between the strain CNR-114^T and the type strains of *S. tropica* CNB440^T and *S. arenicola* CNH-643^T. The mean hybridization values calculated from two replicate experiments for CNR-114^T with the type strains of *S. tropica* and *S. arenicola* were 38.6 % and 53.3 %, respectively. Both of these values are below the 70 % cut-off point recommended for the assignment of bacterial strains to the same genomic species (Wayne et al. 1987).

Chemotaxonomic, cultural and morphological properties

The isolates exhibited cultural and morphological properties typical of members of the genus *Salinispora*

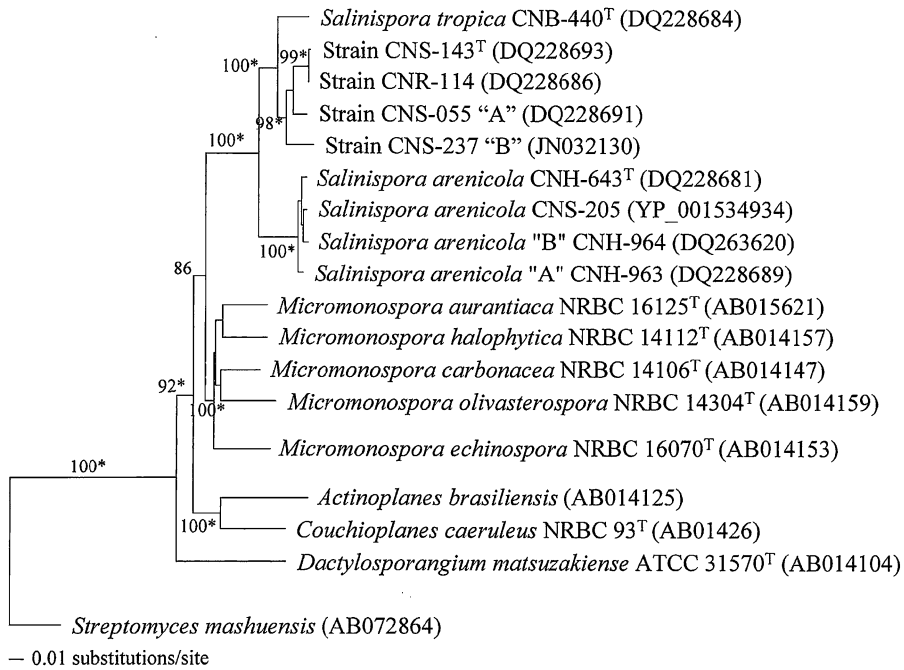

Fig. 2 Neighbour-joining phylogenetic tree (Saitou and Nei 1987) created from 18 *gyrB* nucleotide sequences (714 nucleotides) showing relationships between strains CNS-143, CNR-114^T, CNS-055, and CNS-237 and representatives of some genera classified in the family *Micromonosporaceae*. *Asterisks* indicate branches of the tree also found using the unweighted pair-group method with arithmetic means (Sneath and Sokal 1973), maximum-likelihood (Felsenstein 1981), and maximum-parsimony (Kluge and Farris 1969) methods. Numbers at nodes indicate the level of bootstrap support (%) based on a neighbour-joining analysis of 1,000 re-sampled datasets; only values above 55 % are shown

(Maldonado et al. 2005; Jensen et al. 2012). All of the isolates were observed to form extensively branched substrate hyphae, lack aerial hyphae and failed to grow when seawater was replaced with deionized water in a complex growth medium. The strains were found to grow well on most of the ISP media (Table 1) forming light to dark orange colonies, which on further incubation became dark due to the formation of spores. Isolate CNR 114^T was determined to form round, smooth surfaced spores (0.6 × 1.0 µm diameter) singly and in clusters on ISP 2 agar (Fig. 3).

The strains were found to contain *meso*-A_2pm, arabinose, galactose and xylose as major whole-organism hydrolysates; N-glycolated muramic acid; tetrahydrogenated menaquinones with nine isoprene units as the predominant isoprenologue; complex mixtures of saturated, *iso*- and *anteiso*- fatty acids with iso-$C_{16:0}$ as the major component (Table 2); diphosphatidylglycerol, phosphatidylethanolamine, phosphatidylmethylethanolamine, phosphatidylglycerol, phosphastidylinositol and phosphatidylinositol mannosides as major polar lipids (Fig. 4); but lack mycolic acids. This chemotaxonomic profile is consistent with the classification of the strains in the genus *Salinispora* (Maldonado et al. 2005; Jensen et al. 2012). In general, all of the strains showed similar fatty acid profiles following growth in Trypticase Soy broth, though all but one of the isolates gave markedly lower proportions of the major component iso-$C_{16:0}$ and higher proportions of $C_{17:1}\omega8c$. The G + C content of the DNA of isolates CNR-114^T and CNS-055 were determined to be 69.70 and 70.13 mol%, respectively, values that fall within the range (70–73 mol%) originally reported for the genus *Salinispora* (Maldonado et al. 2005).

Phenotypic tests

It can be seen from Table 3 that the isolates are distinguished from the type strains of *S. arenicola* and *S. tropica* by a number of phenotypic properties, notably

Table 1 Growth and cultural characteristics of the *Salinispora* strains after 2 weeks at 28 °C

Characteristics	*S. pacifica*				*S. arenicola*	*S. tropica*
	CNS-055	CNR-114^T	CNS-143	CNS-237	DSM 44819^T CNH-643^T	DSM 44418^T CNB-440^T
Glucose-yeast extract agar (ISP medium 2)						
Growth	+	+++	+++	+++	+++	+++
Substrate mycelium	Light orange	Orange	Orange	Orange	Orange	Orange
Glycerol-asparagine agar (ISP medium 5)						
Growth	++	+++	+++	+++	+++	+++
Substrate mycelium	Light orange	Dark orange	Dark orange	Light orange	Orange	Orange
Inorganic salts-starch agar (ISP medium 4)						
Growth	++	++	+++	+	++	++
Substrate mycelium	Light orange	Orange	Dark orange	Orange	Light orange	Light orange
Oatmeal agar (ISP medium 3)						
Growth	+	++	++	+++	+++	+++
Substrate mycelium	Light orange	Orange	Orange	Orange	Dark orange	Orange
Peptone-yeast extract-iron agar (ISP medium 6)						
Growth	–	+	–	–	+++	–
Substrate mycelium	N/A	Orange	N/A	N/A	Orange	N/A
Tryptone-yeast extract agar (ISP medium 1)						
Growth	+++	+++	+++	+++	+++	+++
Substrate mycelium	Light orange	Orange	Orange	Orange	Dark orange	Orange
Tyrosine agar (ISP medium 7)						
Growth	+++	+++	++	++	+++	+++
Substrate mycelium	Orange	Orange	Orange	Orange	Orange	Orange

Key: +++, abundant growth; ++, moderate growth; +, poor growth; -, no growth

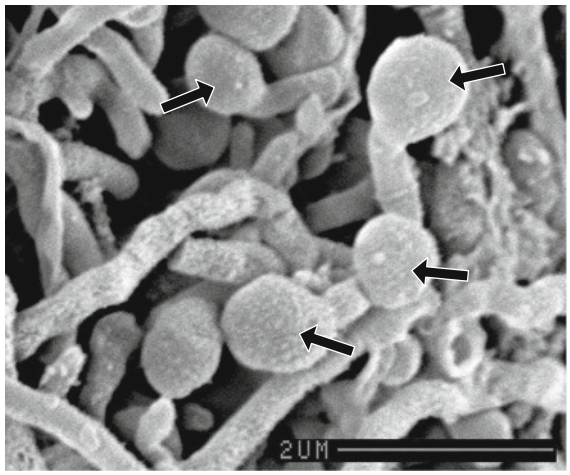

Fig. 3 Scanning electron micrograph of *Salinispora pacifica* CNR-114^T growing on ISP medium 2 supplemented with 75 % seawater after incubation at 28 °C for 3 weeks. *Bar*, 2 μm

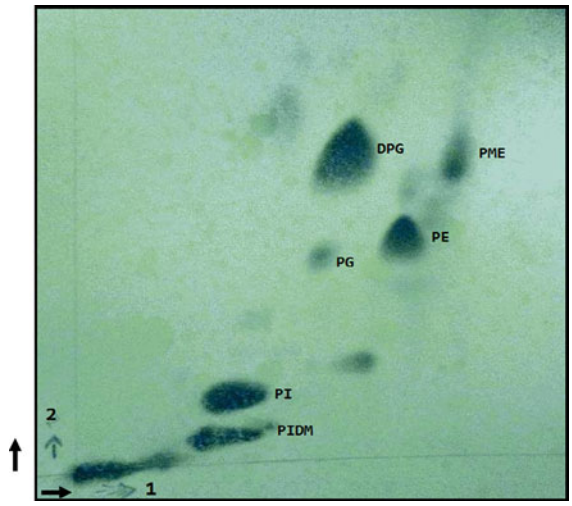

Fig. 4 Two-dimensional-chromatography of polar lipids of *Salinispora pacifica* CNR-114^T stained with molybdophosphoric acid spray (Sigma). Chloroform: methanol: water (32.5: 12.5 : 2.0 v/v) was used in the first direction and chloroform: acetic acid: methanol: water (40: 7.5: 6.2 v/v) in the second direction. *DPG* diphosphatidylglycerol, *PE* phosphatidylethanolamine, *PG* phosphatidylglycerol, *PI* phosphatidylinositol, *PME* phophatidylmethylethanolamine, *DM* phosphatidylinositol dimannosides

by their inability to assimilate diverse compounds as sole carbon sources for growth. In contrast, all of the strains had many phenotypic features in common. They all produced acid and alkaline phosphatase, leucine arylamidase and naphthol-AS-BI-phosphohydrolase (as

Table 2 Major and minor fatty acids of the *Salinispora* strains[a]

Characteristics	*S. pacifica*				*S. arenicola*	*S.tropica*
	CNS 055	CNR 114^T	CNS 143	CNS 237	DSM 44819^T	DSM 44418^T
iso-$C_{14:0}$	2.9	3.7	1.7	1.2	1.6	1.3
iso-$C_{15:0}$	9.7	14.2	17.5	7.7	8.5	6.4
anteiso-$C_{15:0}$	2.6	3.9	2.5	1.1	1.6	1.6
$C_{16:0}$	6.0	3.4	1.6	1.9	0.6	0.7
iso-$C_{16:1}$ H	–	1.5	2.7	3.5	6.5	9.6
iso-$C_{16:0}$	22.5	27.0	30.5	43.4	46.1	49.8
$C_{17:0}$	13.9	8.0	3.5	0.9	0.7	0.8
iso-$C_{17:0}$	2.3	2.6	3.6	4.4	1.8	0.9
anteiso-$C_{17:0}$	2.6	3.5	3.5	3.1	4.9	4.2
$C_{17:0}$ 10-methyl	5.3	3.1	4.9	5.4	5.3	4.6
$C_{17:1}$ ω*8c*	16.6	16.8	9.9	3.7	4.4	4.1
$C_{18:0}$	3.8	2.1	0.8	2.5	0.6	0.5
$C_{18:0}$ 10-methyl	–	–	1.5	6.0	2.7	1.7
Summed feature 6[b]	3.9	1.8	0.9	–	0.5	–
Summed feature 9[b]	–	2.5	8.3	7.3	8.9	8.8

[a] Peaks that accounted for less than 2 % of the total fatty acid composition in strains are not included in the Table

[b] Summed feature represent two or three fatty acids that could not be separated using the MIDI system. Summed feature 6 consisted of $C_{19:1}$ ω11c and / or $C_{19:1}$ ω9c and summed feature 9 of co-methyl $C_{16:0}$ and / or iso-$C_{17:1}$ ω9c

Table 3 Phenotypic properties that separate the *Salinispora* isolates from the type strains of *S. arenicola* and *S. tropica*

Characteristics	Isolates	*S. arenicola* DSM 44419^T	*S. tropica* DSM 44418^T
API ZYM test:			
α-chemotrypsin	−	−	+
Growth at 10 °C	−	+	+
Growth on sole carbon sources at 1%, w/v:			
Erythritol	−	+	−
Fucose, melezitose	−	−	+
Lactose	−	+	+
Assimilation (Biolog GP 2) of:			
Arbutin	+	−	−
Dextrin, D-fructose, gentibiose, glycerol, α-ketoglutaric acid, D-mannitol, methyl pyruvate, pyruvic acid, Tween 80	−	+	−
Sole nitrogen sources:			
L-alanine	−	+	−
L-glutamic acid	+	−	−
Susceptibility to (μg ml^{-1})			
Gentamicin (5)	−	+	+
Penicillin (5)	−	+	+
Rifampin (20)	−	+	−

Key: +, positive; -, negative

determined by API ZYM tests); hydrolysed allantoin, reduced nitrate; degraded DNA, starch and Tween 60; used L-alanine, L-arginine, L-methionine and L-valine as sole nitrogen sources, and were sensitive (μg/ml) to chloramphenicol (4), novobiocin (5), rifampin (20), and oxytetracycline (8). None of the organisms produced cystine arylamidase, α-fucosidase, α- or β- galactosidase, α- glucosidase, β- glucuronidase, lipase (C14), trypsin or valine arylamidase (AP1 ZYM tests); hydrolysed aesculin or urea; degraded adenine, elastin, guanine, hypoxanthine, uric acid, xanthine or xylan; grew on D-arabinose, D-arabitol, dextrin, dulcitol, mannitol, ribose, L-sorbose, turanose or xylose as sole carbon sources (all at 1%, w/v); used L-aspartic acid as a sole nitrogen source or were sensitive (μg/ml) to ampicillin (8), gentamicin (5), novobiocin (25), cephaloridine (4) and streptomycin (25 μg). The isolates showed different responses to several tests, notably the ability to produce esterase (C4), esterase lipase (C8), β-glucosidase, *N*-acetyl-β-glucosamidase and α-mannosidase (API ZYM tests) and to use L-histidine as the sole nitrogen source.

Conclusions

The strains analyzed here form a distinct and well-supported phylogenetic lineage within the genus *Salinispora*. There is no evidence from the present study, or from prior culture based (Freel et al. 2012) or culture independent studies (Mincer et al. 2005) that strains within this lineage overlap in their geographic distribution with the sister lineage *S. tropica*. These observations suggest that geographic isolation (allopatry) has played a role in the divergence of these two closely related lineages. It is evident from the genotypic and phenotypic data that the isolates can be distinguished readily from the type strains of *S. arenicola* and *S. tropica*. It is, therefore, proposed that the isolates be recognized as a new species of the genus *Salinispora*, *S. pacifica* with isolate CNR-114^T as the type strain.

Description of *Salinispora pacifica* sp. nov

Salinispora pacifica (pa.cif.ica L. fem. adj. *pacifica* of the pacific).

The description is based on data from this and an earlier study (Oh et al. 2006).

Aerobic, Gram-positive, non-acid-fast actinomycete, which form a substrate mycelium that carries round, smooth-surfaced spores singly and in clusters. Strains fail to grow when seawater is replaced with deionized water in complex growth media. Good

growth occurs on ISP media prepared using 75 % seawater. Grows between 20 and 30 °C, optimally around 28 °C and from pH 7.0–9.0. Additional phenotypic properties are cited in the text and in Table 2. The chemotaxonomic properties are typical of the genus. The G + C content of the DNA falls within the range 67.70–70.13 mol%. The type strain produces cyanosporoside A.

The type strain, CNR-114^T (DSMZ# = KACC 17160^T), was isolated from a marine sediment sample collected at a depth of 82 m from the Pacific Ocean off the island of Guam on 19 January 2002. The GenBank accession numbers for the 16S rRNA and *gyrB* sequences of strains CNR-114^T, CNS-237, CNS-055, and CNS-143 are DQ224161, DQ318246, DQ224159, DQ92624 and DQ228686, JN032130, DQ228691, DQ228693, respectively.

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