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ORIGINAL ARTICLE

Isolation of actinomycetes from the root of the plant, *Ophiopogon japonicus*, and proposal of two new species, *Actinoallomurus liliacearum* sp. nov. and *Actinoallomurus vinaceus* sp. nov.

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Actinomycete strains K10-0485^T and K10-0528^T were isolated from the roots of *Ophiopogon japonicus* collected in Yokohama, Kanagawa Prefecture, Japan. The 16S ribosomal RNA (rRNA) gene sequences, morphological characteristics and chemotaxonomic data indicated that these strains belonged to the genus *Actinoallomurus*. Strain K10-0485^T showed high similarity of the 16S rRNA gene sequence with *A. luridus* TT02-15^T (99.1%), but the DNA–DNA hybridization relatedness values between strain K10-0485^T and *A. luridus* TT02-15^T were below 70%. Three species showed similarities of 16S rRNA gene sequences with K10-0528^T, namely *A. spadix* JCM 3146^T (98.0%), *A. purpureus* TTN02-30^T (98.0%) and *A. luridus* TT02-15^T (97.9%), but all similarity values of the 16S rRNA gene sequences were lower than the boundary value (98.7%) for distinguishing as different species. Based on phylogenetic analyses, DNA–DNA hybridization relatedness and physiological characteristics, the two isolated strains should be classified as two new species in the genus *Actinoallomurus*, and we propose the names *Actinoallomurus liliacearum* sp. nov. and *Actinoallomurus vinaceus* sp. nov. The type strain of *Actinoallomurus liliacearum* is K10-0485^T (=JCM 17938^T, BCC 49424^T, NBRC 108672^T) and that of *Actinoallomurus vinaceus* is K10-0528^T (=JCM 17939^T, BCC 49425^T, NBRC 108763^T).
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Keywords: *Actinoallomurus liliacearum*; *Actinoallomurus vinaceus*; endophytic actinomycetes; plant root

INTRODUCTION

In order to get new microbial resources that are capable of producing new metabolites, it is useful to isolate untapped actinomycete strains. We have a particular interest in non-*Streptomyces* actinomycetes as sources of new bioactive compounds. The majority of actinomycetes isolated from soil samples belong to the genus *Streptomyces*,[1] while few non-*Streptomyces* actinomycetes are typically isolated. We have isolated rare actinomycetes from plant roots, including members of a proposed novel genus, *Phytohabitans*,[2] and a novel species, *Streptosporangium oxazolinicum*,[3] which produces the new bioactive compounds known as spoxazomicins.[4] Therefore, we focused one line of our search for novel actinomycetes on plant roots.

Pozzi *et al.*[5] reported that *Actinoallomurus* strains possess the ability to produce various bioactive compounds. Many *Actinoallomurus* strains have been found among our isolates. In the process of the screening search for new bioactive compounds in these strains, two strains among our isolates, K10-0485^T and K10-0528^T, were classified as novel species of *Actinoallomurus*. At present, the genus *Actinoallomurus* comprises 12 species:[6] *Actinoallomurus acaciae*,[7] *A. amamiensis*, *A. caesius*, *A. coprocola*, *A. fulvus*, *A. iriomotensis*, *A. luridus*, *A. oryzae*,[8] *A. purpureus*, *A. radicium*,[9] *A. spadix* and *A. yoronensis*. Some type strains of these species, such as *A. acaciae*, *A. oryzae* and *A. radicium*, were isolated from plants. In this paper, we report the taxonomic characteristics of the strains K10-0485^T and K10-0528^T.

MATERIALS AND METHODS

Samples of the perennial plant *Ophiopogon japonicus*, which is native to East Asia, were collected in Kanagawa Prefecture in May 2009. In accordance with the method described by Inahashi *et al.*,[2] actinomycete strains were isolated from the roots on CMC agar (carboxymethyl cellulose 1.0%, ISP (International *Streptomyces* Project) medium 5[10] 1.7%, distilled water, pH 7.0) and water proline agar (proline 1.0%, agar 1.5%, tap water, pH 7.0). Genomic DNA from all isolates was prepared by sonication of the cell suspension[11] and 16S rRNA gene sequences were analyzed as described previously.[2] The phylogenetically closest neighbors were identified by BLAST search using DDBJ database (http://blast.ddbj.nig.ac.jp/top-j.html). Evolutionary distances[12] were estimated by SeaView version 4.2.[13] Multiple alignments with selected sequences were calculated using the ClustalW2 program. The phylogenetic tree was constructed based on the neighbor-joining method,[14] maximum-likelihood method[15] and the maximum-parsimony method.[16] Data were re-sampled with

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1000 bootstrap replications.[17] The values of sequence similarities with the closest strains were determined using the EzTaxon server.[18] The strains K10-0485^T and K10-0528^T, as well as *Actinoallomurus luridus* NBRC 103683^T (= TT02-15^T), were cultured for 3 weeks at 27 °C in order to observe cultural and morphological characteristics on ISP media 2, 3, 4 and 7, and HV agar[19] and YS agar (yeast extract 2.0%, starch 1.0%, agar 1.5%, pH 7.0). The color of aerial and vegetative mycelia and soluble pigments were determined using the Color Harmony Manual.[20] Morphological characteristics were observed by light microscopy and scanning electron microscopy (JSM-5600, JEOL, Tokyo, Japan). The temperature range, pH range and the NaCl tolerance for growth were determined using ISP medium 2. Utilization of carbohydrates as the sole carbon source was tested using ISP medium 9.[21] Utilization of urea was tested using the method of Gordon *et al.*[22] ISP medium 4 was used for starch hydrolysis, ISP medium 8 was used for nitrate reduction, glucose–peptone–gelatin medium (glucose 2.0%, peptone 0.5%, gelatin 20%, pH 7.0) was used for gelatin hydrolysis, 10% skim milk was used for coagulation and peptonization of milk, and skim milk agar was used for casein hydrolysis.[22] Enzyme activities were determined using the API ZYM system (BioMérieux, Lyon, France) according to the manufacturer's instructions. Biomass for the molecular systematics and chemotaxonomic studies was obtained after cultivation on a rotary shaker in yeast extract-glucose broth (yeast extract 1.0%, glucose 1.0%, pH 7.0) for 1 week at 27 °C. Isoprenoid quinones extracted as described by Collins *et al.*[23] were analyzed by LC/MS (JMS-T 100LP, JEOL) using a CAPCELL PAK C18 UG120 (Shiseido, Tokyo, Japan) with methanol/2-propanol (7:3). Purified cell wall was obtained using the method of Kawamoto *et al.*,[24] and the amino-acid composition of hydrolyzed cell walls was determined by thin layer chromatography. The *N*-acyl types of muramic acid were determined using the method of Uchida and Aida.[25] Phospholipids in cells were extracted and identified using the method of Minnikin *et al.*[26] The presence of mycolic acids was examined by thin layer chromatography in accordance with Tomiyasu.[27] Whole-cell sugar composition was analyzed according to the methods of Becker *et al.*[28] Methyl esters of cellular fatty acids were prepared by direct transmethylation with methanolic hydrochloride, and were analyzed on a GLC system (HP 6890, Hewlett Packard, Palo Alto, CA, USA). Identification and quantification of the fatty acid methyl esters, as well as numerical analysis of fatty acid profiles, were performed according to the instructions for the Microbial Identification System I with ACTIN1 method.[29] For G + C content and DNA–DNA hybridization, chromosomal DNA was prepared in accordance with the procedure of Saito and Miura.[30] DNA G + C content was determined by HPLC according to the method of Tamaoka and Komagata,[31] and DNA–DNA hybridization was performed using the photobiotin-labeling method of Ezaki *et al.*[32]

RESULTS AND DISCUSSION

Isolation of actinomycetes from plant roots

A total of 135 actinomycete strains were isolated from the roots of a single specimen of *Ophiopogon japonicus*. The 16S rRNA gene partial sequences (approximately 500 bp) showed that the isolates comprised the genera *Planotetraspora* (23%), *Acrocarpospora* (18%), *Actinoallomurus* (13%), *Streptomyces* (13%) and others (33%) (Table 1). This suggests that non-*Streptomyces* strains were isolated with high frequency. There were 17 strains of the genus *Actinoallomurus* among the isolates. Phylogenetic analysis by the almost full-length sequences showed that these strains fell into three clusters; the first cluster comprised strains K10-0467 and K10-0474, the second consisted of strains K10-0475 and K10-0528^T, and the third cluster comprised strain K10-0485^T and 12 similar strains (Figure 1). This indicates that various strains belonging to the genus *Actinoallomurus* were isolated from a single plant. Furthermore, the two clusters including K10-0485^T and K10-0528^T were not clustered with previously described species, so a taxonomic study of K10-0485^T and K10-0528^T was carried out.

Table 1 Classification of isolates by 16S rRNA gene sequences

Genus	*Number of isolates*	*Ratio (%)*
Planotetraspora	31	23
Acrocarpospora	24	18
Actinoallomurus	17	13
Streptomyces	17	13
Nonomuraea	14	10
Sphaerisporangium	14	10
Micromonospora	7	5
Microbispora	3	2
Polymorphospora	3	2
Amycolatopsis	2	1
Unidentified	3	2
Total	135	100

Taxonomic study of strains K10-0485^T and K10-0528^T

Morphological, cultural and physiological characteristics. Strain K10-0485^T grew well on ISP media 2 and 3, and on YS agar. Spores on aerial mycelia were only produced on HV agar after 3 weeks at 27 °C. Vegetative mycelia were branched but not fragmented. The colony color of the strain was white to pale yellow, and soluble pigment was not produced (Figure 2, Table 2). The temperature and pH range for growth were 20–40 °C and pH 5–8, with optimal growth at 28–36 °C and pH 5–7. The strain did not grow on 3% NaCl medium. Casein was degraded. Starch was hydrolyzed. Gelatin was weakly liquefied. Milk was not peptonized or coagulated. Nitrate was not reduced to nitrite. Melanin was not produced. The strain utilized L-arabinose, D-fructose, D-galactose, D-glucose, *myo*-inositol, L-rhamnose, D-sucrose, D-xylose and cellulose, but did not utilize dulcitol, maltose, D-mannitol, melibiose, raffinose or D-sorbitol. Using the API ZYM system, *N*-acetyl-β-glucosaminidase, acid phosphatase, cystine allylamidase, α-chymotrypsin, esterase (C4), α-glucosidase, β-glucosidase, β-glucuronidase, leucine allylamidase, α-mannosidase, naphthol-AS-BI-phosphohydrolase and valine allylamidase were positive, while alkaline phosphatase, esterase lipase (C8), α-galactosidase, β-galactosidase and trypsin were weakly positive, and α-fucosidase and lipase (C14) were negative (Table 3).

Strain K10-0528^T grew well on ISP media 2 and 3, and on YS agar. Aerial mycelia were only produced on ISP medium 2 for 6 weeks at 27 °C, but spores were not observed. Vegetative mycelia were branched but not fragmented. The colony color of the strain was white to gray–purple and purple soluble pigment was produced (Table 2). The temperature and pH range for growth were 15–39 °C and pH 5–8, with optimal growth at 23–39 °C and pH 5–6. The strain did not grow on 2% NaCl medium. Casein was degraded. Starch was hydrolyzed. Gelatin was not liquefied. Milk was not peptonized or coagulated. Nitrate was not reduced to nitrite. Melanin was not produced. The strain utilized L-rhamnose and D-sucrose, but did not utilize L-arabinose, dulcitol, D-fructose, D-galactose, D-glucose, *myo*-inositol, maltose, D-mannitol, melibiose, raffinose, D-sorbitol, D-xylose or cellulose. Using the API ZYM system *N*-acetyl-β-glucosaminidase, acid phosphatase, alkaline phosphatase, cystine allylamidase, α-fucosidase, β-galactosidase, α-glucosidase, β-glucosidase, leucine allylamidase, naphthol-AS-BI-phosphohydrolase and valine allylamidase were positive, esterase (C4), esterase lipase (C8), α-galactosidase and α-mannosidase were weakly positive, and α-chymotrypsin, β-glucuronidase lipase (C14) and trypsin were negative (Table 3).

Chemotaxonomy. Strains K10-0485^T and K10-0528^T both contained *meso*-diaminopimelic acid, lysine, alanine and glutamic acid in the cell-wall peptidoglycan, and galactose, glucose, madurose, mannose and ribose were detected as whole-cell sugars. The *N*-acyl type of

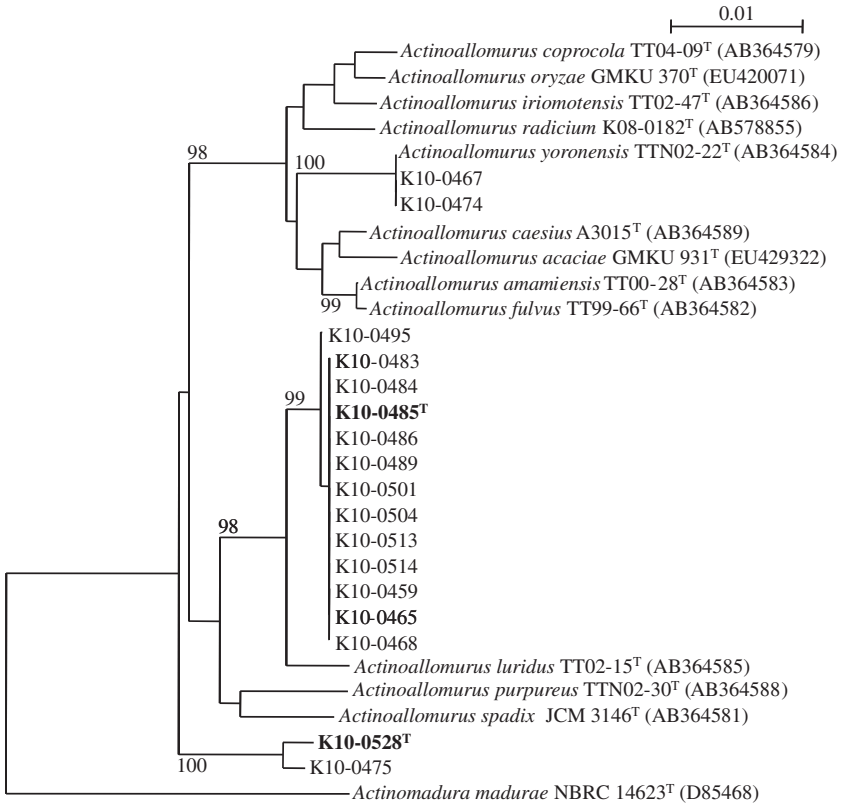

Figure 1 Neighbor-joining tree, based on 16S rRNA gene sequences, showing relationship between isolates from plant roots and members of the genus *Actinoallomurus*. Only bootstrap values above 70% (percentages of 1000 replications) are indicated. Bar, 0.01 nucleotide substitutions per site.

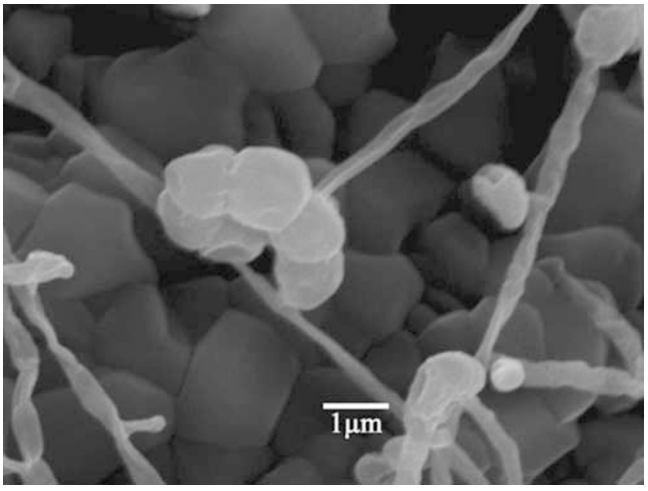

Figure 2 Scanning electron micrograph of strain K10-0485^T grown on HV agar for 3 weeks at 27 °C.

muramic acid was acetyl. Phosphatidylglycerol and diphosphatidylglycerol were detected, but phosphatidylethanolamine, phosphatidylcholine and phosphatidylinositol were not found. Mycolic acids were not detected. The major menaquinones were MK-9 (H_8) (72%), MK-9 (H_6) (25%) for strain K10-0485^T, and MK-9 (H_6) (60%) and MK-9 (H_8) (40%) for strain K10-0528^T. The major cellular fatty acids were iso-$C_{16:0}$ (41.3%), 10-methyl $C_{17:0}$ (17.2%) and anteiso-$C_{17:0}$ (14.6%) for K10-0485^T, and iso-$C_{16:0}$ (30.9%), anteiso-$C_{17:0}$ (18.5%) and ω9c $C_{17:1}$ (15.4%) for K10-0528^T (Table 4). The G+C content of the genomic DNA was 72 mol% in both strains. These chemotaxonomic properties of strains K10-0485^T and K10-0528^T, which contain both *meso*-diaminopimelic acid and lysine as diamino acids in the cell-wall peptidoglycan, madurose and galactose as whole-cell sugars, and MK-9 (H_8) and MK-9 (H_6) as the predominant menaquinones, indicated that they are members of the genus *Actinoallomurus*.

Phylogenetic analysis. Phylogenetic analysis, based on the 16S rRNA gene sequences, indicated that the two strains belong to the genus *Actinoallomurus*, and that strain K10-0485^T is closest to *A. luridus* TT02-15^T, while strain K10-0528^T is closest to *A. spadix* JCM 3146^T, *A. purpureus* TTN02-30^T and *A. luridus* TT02-15^T (Figure 3).

The organisms showing the highest similarity values for strain K10-0485^T were *A. luridus* TT02-15^T (99.1%), *A. purpureus* TTN02-30^T (98.3%) and *A. spadix* JCM 3146^T (97.7%). The organisms showing the highest similarity values to strain K10-0528^T were strains K10-0485^T (98.1%), *A. purpureus* TTN02-30^T (98.0%), *A. spadix* JCM 3146^T (98.0%) and *A. luridus* TT02-15^T (97.9%). As the highest similarity value to strain K10-0528 ^{T} was 98.1%, much lower than the boundary value of 98.7%[33] for distinguishing organisms of different species, this strain should be recognized as a separate species. The DDBJ accession numbers of the 16S rRNA gene sequences of strains K10-0485^T and K10-0528^T are AB668306 and AB668307, respectively.

DNA–DNA hybridization. DNA–DNA relatedness values between strain K10-0485^T and *A. luridus* TT02-15^T were 53–63%, below the recommended DNA–DNA relatedness cutoff point for species delineation of 70%.[34]

Table 2 Cultural characteristics of strains K10-0485^T, K10-0528^T and *A. luridus* TT02-15^T

Medium	A. luridus *TT02-15^T*	*K10-0485^T*	*K10-0528^T*
Growth on ISP 2			
Growth	Good	Good	Good
Growth color	Pale yellow (1 1/2ca) to Pearl pink (3ca)	Pearl pink (3ca)	Dusty mauve (8ge)
Reverse color	Colonial yellow (2ga)	Bamboo (2gc)	Rasberry (9pe)
Aerial mycelium	None	None	None
Soluble pigment	None	None	Orchid (10ic)
Growth on ISP 3			
Growth	Moderate	Good	Good
Growth color	White (a) to Pale yellow (1ca)	White (a)	White (a) to Gray (12pm)
Reverse color	Light wheat (2ea)	White (a)	White (a) to Gray (12li)
Aerial mycelium	None	None	None
Soluble pigment	None	None	Pale pink (8ca)
Growth on ISP 4			
Growth	Moderate	Moderate	Moderate
Growth color	White (a) to Light ivory (2ca)	White (a)	Pale pink (6ca)
Reverse color	Light ivory (2ca)	White (a)	Pale brown (4gc)
Aerial mycelium	None	None	None
Soluble pigment	None	None	Pale pink (7ca)
Growth on ISP 7			
Growth	Moderate	Moderate	Moderate
Growth color	White (a)	White (a)	White (a)
Reverse color	White (a)	White (a)	White (a)
Aerial mycelium	None	None	None
Soluble pigment	None	None	None
Growth on YS agar			
Growth	Good	Good	Good
Growth color	White (a)	Pale yellow (1ca)	Orchid gray (9ig)
Reverse color	White (a)	White (a)	Ash rose (7 1/2ie)
Aerial mycelium	None	None	None
Soluble pigment	None	None	Rose (8ic)

Table 3 Differential physiological characteristics of strains K10-0485^T, K10-0528^T and *A. luridus* TT02-15^T

	A. luridus *TT02-15^T*	*K10-0485^T*	*K10-0528^T*
NaCl tolerance (%)	1	2	1
Peptonization of milk	+	−	−
Utilization of:			
L-arabinose	−	+	−
D-xylose	−	+	−
D-glucose	+	+	−
D-fructose	+	+	−
L-rhamnose	−	+	+
myo-Inositol	−	+	−
D-galactose	−	+	−
Cellulose	−	+	−
Enzyme activity of:			
Alkaline phosphatase	±	±	+
Esterase (C4)	+	+	±
Valine allyl amidase	±	+	+
Cystine allyl amidase	±	+	+
Trypsin	−	±	−
α-Chymotrypsin	−	+	−
α-Galactosidase	−	±	±
β-Galactosidase	−	±	+
β-Glucuronidase	±	+	−
β-Glucosidase	−	+	+
α-Mannosidase	−	+	±
α-Fucosidase	−	−	+

Abbreviations: −, negative; +, positive; ±, weakly positive.

Table 4 Cellular fatty acid compositions (%) of strains K10-0485^T, K10-0528^T and *A. luridus* TT02-15^T

	A. luridus *TT02-15^T*[a]	*K10-0485^T*	*K10-0528^T*
iso-$C_{16:1}$ iso G	7.1	3.3	7.0
iso-$C_{16:0}$	40.4	41.3	30.9
anteiso-$C_{17:1}$	5.6	1.5	4.3
anteiso-$C_{17:0}$	23.9	14.6	18.5
ω9c $C_{17:1}$	–	2.6	15.4
$C_{17:0}$	–	1.3	1.6
10-Methyl $C_{17:0}$	8.5	17.2	10.4
iso-$C_{18:0}$	2.9	2.9	0.6
ω9c $C_{18:1}$	–	1.2	2.8
$C_{18:0}$	–	3.1	1.4
10-Methyl iso-$C_{18:0}$	3.8	–	–
10-Methyl $C_{18:0}$	1.7	3.2	1.8
$C_{19:0}$	–	1.2	0.6
Unknown ECL[b] 16.048	6.0	1.0	–
Unknown ECL 19.368	–	1.8	–
Unknown 18.756/19:1	–	2.0	2.6

Fatty acids less than 1% in all strains are omitted.
–, not detected.
[a]Data are taken from Tamura *et al.*[6]
[b]Equivalent chain length.

CONCLUSION

We isolated 135 actinomycete strains from the roots of the Japanese plant *Ophiopogon japonicus*. The 16S rRNA gene partial sequences indicated that non-*Streptomyces* strains were isolated at high rate (87.4%) containing 17 *Actinoallomurus* strains, which possess the ability to produce various bioactive compounds (Table 1). Although the 17 strains were isolated from a sole plant root, they were separated phylogenetically into three groups based on the almost full-length 16S rRNA gene sequences (Figure 1). Two strains, K10-0485^T and K10-0528^T, showed low similarities with nearest previously described species, indicating the need for further taxonomic studies.

The morphological and chemotaxonomic properties of strains K10-0485^T and K10-0528^T supported the notion that these strains belong to the genus *Actinoallomurus*. DNA–DNA relatedness values between strain K10-0485^T and the closest species (*A. luridus* TT02-15^T) were below the critical value of 70%. Furthermore, strain K10-0485^T can be distinguished from related species based on cultural and physiological characteristics, as shown in Tables 2 and 3. The 16S rRNA gene sequence similarities between strain K10-0528^T and possibly related

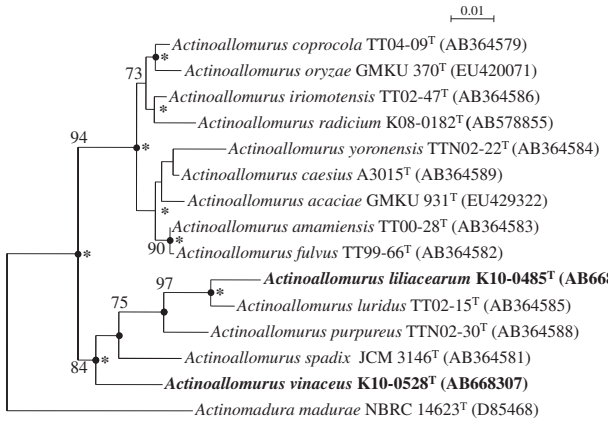

Figure 3 Neighbor-joining tree, based on 16S rRNA gene sequences, showing relationship between strains K10-0485^T, K10-0528^T and members of the genus *Actinoallomurus*. Only bootstrap values above 70% (percentages of 1000 replications) are indicated. Solid circle branches also recovered in the maximum-likelihood tree; asterisks show branches also recovered in the maximum-parsimony tree; Bar, 0.01 nucleotide substitutions per site.

strains were low (less than 98.1%). These observations support the contention that strains K10-0485^T and K10-0528^T represent two novel species in the genus *Actinoallomurus*, for which the names *Actinoallomurus liliacearum* sp. nov. and *Actinoallomurus vinaceus* sp. nov., respectively, are proposed.

Description of *Actinoallomurus liliacearum* sp. nov.

Actinoallomurus liliacearum (li.li.a.ce.a'rum. N.L. pl. n. Liliaceae, a scientific family name; referring to the isolation of the strain from the roots of Liliaceae plant, *Ophiopogon japonicas*).

The color of colonies is white to gray. Vegetative mycelia are branched and not fragmented, and the color of aerial mycelia is white. Spore chains form spirals. Spores are smooth. Growth occurs at 20–40 °C and pH 5–8, with no growth in the presence of 3% NaCl. Casein and cellulose are degraded. Starch is hydrolyzed, but urea is not. Gelatin is weakly liquefied. Milk is not peptonized or coagulated. Nitrate is not reduced to nitrite. Melanin is not produced. L-arabinose, D-fructose, D-galactose, D-glucose, *myo*-inositol, L-rhamnose, D-sucrose and D-xylose are utilized, but dulcitol, maltose, D-mannitol, melibiose, raffinose and D-sorbitol are not. According to the API ZYM system, *N*-acetyl-β-glucosaminidase, acid phosphatase, cystine allylamidase, α-chymotrypsin, esterase (C4), α-glucosidase, β-glucosidase, β-glucuronidase, leucine allylamidase, α-mannosidase, naphthol-AS-BI-phosphohydrolase and valine allylamidase are positive, alkaline phosphatase, esterase lipase (C8), α-galactosidase, β-galactosidase and trypsin are weakly positive, α-fucosidase and lipase (C14) are negative. Major fatty acids are iso-$C_{16:0}$, 10-methyl $C_{17:0}$ and anteiso-$C_{17:0}$. The G + C content of the genomic DNA of type strain is 72 mol%. The species description is based on a single strain. The type strain is K10-0485^T (= JCM 17938^T, BCC 49424^T, NBRC 108762^T), which was isolated from the root of *Ophiopogon japonicus* in Yokohama, Kanagawa, Japan.

Description of *Actinoallomurus vinaceus* sp. nov.

Actinoallomurus vinaceu (vi.na'ceus. L. masc. adj. *vinaceus* grape, referring to the color of the colonies).

The color of colonies is gray to purple. Vegetative mycelia are branched and not fragmented, while the color of aerial mycelia is white. Growth occurs at 15–39 °C and pH 5–8, with no growth in the presence of 2% NaCl. Casein is degraded, but cellulose is not. Starch is hydrolyzed, but urea is not. Gelatin is not liquefied. Milk is not peptonized or coagulated. Nitrate is not reduced to nitrite. Melanin is not produced. L-rhamnose and D-sucrose are utilized, but L-arabinose, dulcitol, D-fructose, D-galactose, D-glucose, *myo*-inositol, maltose, D-mannitol, melibiose, raffinose, D-sorbitol and D-xylose are not. According to the API ZYM system, *N*-acetyl-β-glucosaminidase, acid phosphatase, alkaline phosphatase, cystine allylamidase,α-fucosidase, β-galactosidase, α-glucosidase, β-glucosidase, leucine allylamidase, naphthol-AS-BI-phosphohydrolase and valine allylamidase are positive, esterase (C4), esterase lipase (C8), α-galactosidase and α-mannosidase are weakly positive, α-chymotrypsin, β-glucuronidase lipase (C14) and trypsin are negative. Major fatty acids are iso-$C_{16:0}$, anteiso-$C_{17:0}$ and ω9c $C_{17:1}$. The G + C content of the genomic DNA of type strain is 72 mol%. The species description is based on a single strain. The type strain is K10-0528^T (= JCM 17939^T, BCC 49425^T, NBRC 108763^T), which was isolated from a root of *Ophiopogon japonicus* in Yokohama, Kanagawa, Japan.

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