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# Transfer of *Actinomadura spadix* Nonomura and Ohara 1971 to *Actinoallomurus spadix* gen. nov., comb. nov., and description of *Actinoallomurus amamiensis* sp. nov., *Actinoallomurus caesius* sp. nov., *Actinoallomurus coprocola* sp. nov., *Actinoallomurus fulvus* sp. nov., *Actinoallomurus iriomotensis* sp. nov., *Actinoallomurus luridus* sp. nov., *Actinoallomurus purpureus* sp. nov. and *Actinoallomurus yoronensis* sp. nov.

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Ten actinomycete strains that form chains of spiral or looped spores were isolated from soil and dung samples in Japan. They contained D- and L-lysine, *meso*-diaminopimelic acid ($A_2$pm), D-glutamic acid and D- and L-alanine in the cell-wall peptidoglycan, madurose as a characteristic whole-cell sugar, MK-9($H_6$) and MK-9($H_8$) as the major isoprenoid quinones and iso-$C_{16:0}$ as the major cellular fatty acid and showed genomic DNA G+C contents of 69–74 mol%. Phylogenetic analyses based on 16S rRNA gene sequences showed that the isolated actinomycete strains consistently formed a monophyletic cluster with *Actinomadura spadix* NBRC 14099$^T$ and a separate line of descent in the phylogenetic cluster of the family *Thermomonosporaceae*. *Actinomadura spadix* NBRC 14099$^T$ also contained D- and L-lysine in addition to *meso*-$A_2$pm. This genetic and phenotypic evidence revealed that the actinomycete strains could be clearly differentiated from the other members of the family *Thermomonosporaceae* and that they warranted separate genus status. We conclude that *Actinomadura spadix* should be assigned the status of the type species of a new genus as *Actinoallomurus spadix* gen. nov., comb. nov. (type strain NBRC 14099$^T$ =ATCC 27298$^T$ =BCRC 13386$^T$ =CBS 261.72$^T$ =CIP 105479$^T$ =DSM 43459$^T$ =JCM 3146$^T$ =KCTC 9252$^T$ =NCIMB 11118$^T$ =NRRL B-16128$^T$). Further, we conclude that the ten new isolates should be assigned to the novel species *Actinoallomurus amamiensis* sp. nov. (type strain TT00-28$^T$ =NBRC 103682$^T$ =KCTC 19537$^T$), *Actinoallomurus caesius* sp. nov. (type strain A3015$^T$ =NBRC 103678$^T$ =KCTC 19535$^T$), *Actinoallomurus coprocola* sp. nov. (type strain TT04-09$^T$ =NBRC 103688$^T$ =KCTC 19542$^T$), *Actinoallomurus fulvus* sp. nov. (type strain TT99-66$^T$ =NBRC 103680$^T$ =KCTC 19536$^T$), *Actinoallomurus iriomotensis* sp. nov. (type strain TT02-47$^T$ =NBRC 103685$^T$ =KCTC 19539$^T$), *Actinoallomurus luridus* sp. nov. (type strain TT02-15$^T$ =NBRC 103683$^T$ =KCTC 19538$^T$), *Actinoallomurus purpureus* sp. nov. (type strain TTN02-30$^T$ =NBRC 103687$^T$ =KCTC 19541$^T$) and *Actinoallomurus yoronensis* sp. nov. (type strain TTN02-22$^T$ =NBRC 103686$^T$ =KCTC 19540$^T$).

Abbreviation: $A_2$pm, diaminopimelic acid.

The GenBank/EMBL/DDBJ accession numbers for the 16S rRNA gene sequences of the strains used are AB364579–AB364589, as detailed in Table 1.

A 16S rRNA gene sequence-based maximum-likelihood tree, 2D TLC of the polar lipids and SEMs of growth of strain NBRC 14099$^T$, fatty acid profiles of *Actinoallomurus* strains and details of growth of *Actinoallomurus* strains on various media are available as supplementary material with the online version of this paper.

The members of the genus *Actinomadura* are described as actinomycetes that form non-fragmenting, extensively branched substrate mycelium and aerial hyphae with arthrospores which may be straight, hooked (in the form of open loops) or irregular spirals (one to four turns) (Lechevalier & Lechevalier, 1970). The spore surfaces are either folded, irregular, smooth, spiny or warty. The cell-wall peptidoglycan contains *meso*-diaminopimelic acid ($A_2$pm), type A1$\gamma$ (Schleifer & Kandler, 1972), and whole-cell hydrolysates contain madurose. At the time of writing, the genus *Actinomadura* included about 65 species with validly published names, although about 24 of these have been reclassified. They are included within the evolutionary radiation occupied by the family *Thermomonosporaceae*. *Actinomadura spadix* Nonomura and Ohara 1971 is known as a distinct species having low 16S rRNA gene similarity with the other species of the genus; the highest 16S rRNA gene sequence similarity of the type strain, 95.9 %, is shown by the type strain of *Actinomadura alba* (Wang *et al.*, 2007). In the present study, soil isolates that were phylogenetically related to *Actinomadura spadix* were subjected to a polyphasic investigation in order to establish their taxonomic positions.

During the course of an ecological study of actinomycetes conducted in Japan, ten actinomycete strains were isolated from soil and dung samples collected from various places (Table 1). All the strains except TT04-09$^T$ were isolated by the yeast extract-SDS method (Hayakawa & Nonomura, 1989) using humic acid-vitamin (HV) agar medium (Hayakawa & Nonomura, 1987). Strain TT04-09$^T$ was obtained from cow dung by incubation on water agar containing 50 mg cycloheximide $l^{-1}$ for 1 month at room temperature. PCR amplification, sequencing of 16S rRNA genes and phylogenetic analyses were performed as described previously (Tamura & Hatano, 2001). Sequences were aligned using CLUSTAL_X version 1.83 (Thompson *et al.*, 1997) and the alignment was corrected manually. Alignment gaps and ambiguous bases were removed prior to the phylogenetic analyses by using BioEdit version 7.0.5.3 (Hall, 1999). Neighbour-joining (Saitou & Nei, 1987) phylogenetic trees were constructed using CLUSTAL_X according to Kimura's two-parameter model (Kimura, 1980). Maximum-parsimony (Kluge & Farris, 1969) and maximum-likelihood (Felsenstein, 1981) analyses were performed using the PAUP software version 4.0b10 (Swofford, 2002). Bootstrap values (Felsenstein, 1985) based on 1000 replications were used to evaluate the confidence limits on phylogenies. The 16S rRNA gene sequence analysis revealed that the isolated strains formed a monophyletic cluster with *Actinomadura spadix* NBRC 14099$^T$ in which the branch was supported by a bootstrap value of 100 % (Fig. 1 and Supplementary Fig. S1, available in IJSEM Online). Binary 16S rRNA gene sequence similarities among the isolates and *Actinomadura spadix* NBRC 14099$^T$ were not less than 96.7 %. Strains TT02-47$^T$, TT02-22, and TT99-68 shared high 16S rRNA gene sequence similarities, ranging from 99.8 to 100 %. All of the isolates and *Actinomadura spadix* NBRC 14099$^T$ showed similarity of 96.2 % or less with other *Actinomadura* strains, such as the type strains of *Actinomadura nitritigenes* (96.2 %), *Actinomadura echinospora* (96.0 %), *Actinomadura alba* (95.9 %) and *Actinomadura fulvescens* (95.8 %).

For chemotaxonomic analyses, freeze-dried cells were obtained from cultures grown in yeast extract-glucose

**Table 1.** Actinomycete strains used in this study

All isolation locations are in Japan.

| Strain | Source, locality and year of isolation | 16S rRNA gene sequence accession number |
|---|---|---|
| *Actinoallomurus* (*Actinomadura*) *spadix* comb. nov. NBRC 14099$^T$ | Soil, Yamanashi (Nonomura & Ohara, 1971) | AB364581 |
| *Actinoallomurus amamiensis* sp. nov. TT00-28$^T$ | Grove soil, *Citrus tankan*, Amami Island, Kagoshima, 2000 | AB364583 |
| *Actinoallomurus caesius* sp. nov. A3015$^T$ | Mangrove forest soil, Iriomote Island, Okinawa, 1998 | AB364589 |
| *Actinoallomurus coprocola* sp. nov. TT04-09$^T$ | Cow dung, Futtsu, Chiba, 2003 | AB364579 |
| *Actinoallomurus fulvus* sp. nov. TT99-66$^T$ | Sugar-cane field soil, Iriomote Island, Okinawa, 1999 | AB364582 |
| *Actinoallomurus iriomotensis* sp. nov. | | |
| TT02-47$^T$ | Meadow soil, Iriomote Island, Okinawa, 2001 | AB364586 |
| TT02-22 | As above | AB364587 |
| TT99-68 | Pineapple field soil, Iriomote Island, Okinawa, 1999 | AB364580 |
| *Actinoallomurus luridus* sp. nov. TT02-15$^T$ | Meadow soil, Iriomote Island, Okinawa, 2001 | AB364585 |
| *Actinoallomurus purpureus* sp. nov. TTN02-30$^T$ | Forest soil, Yoro valley, Chiba, 2002 | AB364588 |
| *Actinoallomurus yoronensis* sp. nov. TTN02-22$^T$ | Forest soil, Yoro valley, Chiba, 2002 | AB364584 |

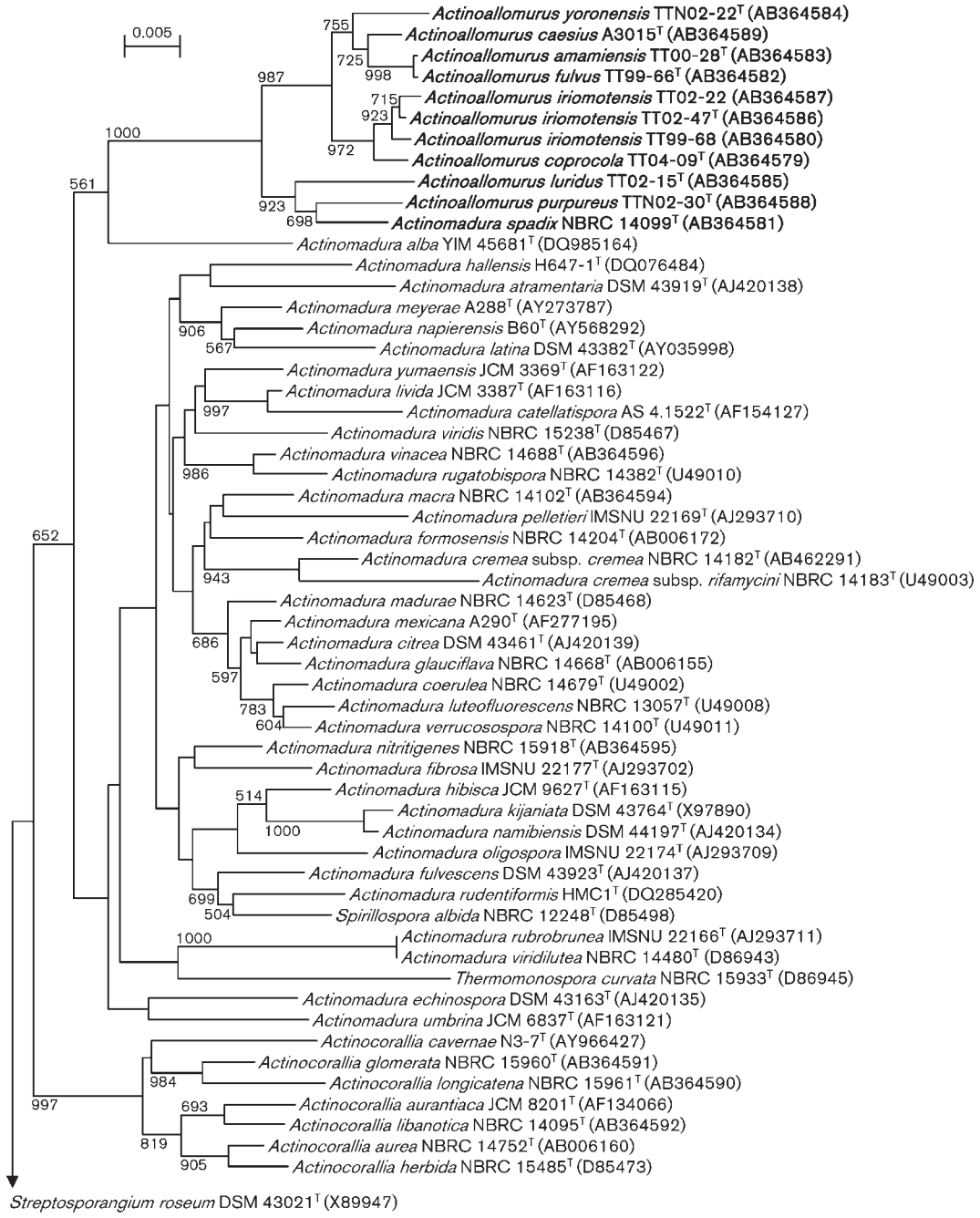


**Fig. 1.** Phylogenetic tree derived from the 16S rRNA gene sequences of *Actinoallomurus* species and related actinomycetes belonging to the family *Thermomonosporaceae*. The tree was constructed by using the neighbour-joining method (Saitou & Nei, 1987). The sequence of *Streptosporangium roseum* DSM 43021$^T$ was used as the outgroup. Bar, 0.005 $K_{nuc}$ in nucleotide sequences. Numbers on branches indicate confidence limits that were estimated by bootstrap analysis with 1000 replicates; only values >50 % are provided. A maximum-likelihood tree is available as Supplementary Fig. S1.

broth on a rotary shaker at 28 °C for 5 days. Analyses of whole-cell sugar patterns, cell-wall amino acids, menaquinones, the acyl type of the peptidoglycan, mycolic acids and DNA base composition were performed as described previously (Tamura *et al.*, 1994). Amino acid isomers of the cell-wall peptidoglycan and their composition were determined and analysed by liquid chromatography/mass spectrometry according to the method described by Nozawa *et al.* (2007). Cellular fatty acid methyl esters were prepared and analysed according to the protocol of the MIDI Sherlock Microbial Identification System (Sasser, 1990; MIDI, 2002). MK-9($H_6$) (17–73 %) and MK-9($H_8$)

(18–83 %) were the predominant menaquinones present in these strains. In addition, MK-9($H_4$) and MK-9($H_2$) (19 and 16 %, respectively) were present in strain A3015$^T$ and MK-9($H_4$) was present in strains TT02-15$^T$ and TT04-09$^T$ (12 and 13 %, respectively). Madurose, galactose and glucose were present as whole-cell sugars. The cell-wall peptidoglycan of *Actinomadura spadix* NBRC 14099$^T$ and the ten isolates contained *meso*-$A_2$pm, D- and L-lysine, D- and L-alanine and D-glutamic acid. Neither D- nor L-lysine could be detected in the cell-wall peptidoglycan of *Actinomadura madurae* NBRC 14623$^T$ or *Actinomadura viridis* NBRC 15238$^T$ under the same conditions. The major fatty acids present in these strains were iso-$C_{16:0}$ and anteiso-$C_{17:0}$. Strain A3015$^T$ contained less anteiso-$C_{17:0}$ than 10-methyl $C_{17:0}$, which was present in a large amount. The cellular fatty acid compositions of *Actinomadura spadix* NBRC 14099$^T$ and the new isolates are shown in Supplementary Table S1. With regard to phospholipids, phosphatidylglycerol was detected; however, phosphatidylethanolamine, phosphatidylcholine and ninhydrin-positive unidentified phospholipids were not detected (phospholipid type PI *sensu* Lechevalier *et al.* 1977) (Supplementary Fig. S2). The DNA G+C contents of *Actinomadura spadix* NBRC 14099$^T$ and the new isolates ranged from 69.0 to 74.2 mol%.

Morphological characteristics were observed by scanning electron microscopy as described previously (Tamura *et al.*, 1994). *Actinomadura spadix* NBRC 14099$^T$ and the isolates formed spore chains of up to 15 spores, which were hooked (in the form of open loops), looped or spiral on the tips of the branched aerial mycelium (Supplementary Fig. S3). The spiral spore chain occasionally resembled a sporangium (pseudosporangium). The spores were spherical, oval and/or rod-shaped with a smooth surface, 0.5–1.2 × 0.5–2.0 μm. Motile cells were not observed.

Cultural and physiological characteristics were examined as described previously (Gordon *et al.*, 1974; Seino *et al.*, 1985) as well as by using API kits (bioMérieux). Detailed results of these analyses are provided in Supplementary Table S2 and Table 2 and in the descriptions of the species. Strain TTN02-30$^T$ formed purple colonies on ISP-4, ISP-7 and yeast extract-starch agar. The mass colour of the other strains ranged from yellow to reddish brown. Growth and physiological characteristics differed among all the strains except TT02-47$^T$, TT02-22 and TT99-68, which showed similar characteristics. The isolates and *Actinomadura spadix* NBRC 14099$^T$ grew optimally at pH 5.5–6.0.

The microplate hybridization method developed by Ezaki *et al.* (1988, 1989) was applied to determine DNA–DNA relatedness. The DNA–DNA relatedness among strains TT02-47$^T$, TT02-22 and TT99-68 ranged from 61 to 90 %. In contrast, the relatedness among *Actinomadura spadix* NBRC 14099$^T$ and strains TTN02-30$^T$, TT02-15$^T$, TT02-47$^T$, TT04-09$^T$, TTN02-22$^T$, A3015$^T$, TT00-28$^T$ and TT99-66$^T$ ranged from 6 to 55 %.

The phylogenetic data and chemotaxonomic characteristics, especially the presence of D- and L-lysine and *meso*-$A_2$pm in the cell-wall peptidoglycan, clearly distinguished *Actinomadura spadix* NBRC 14099$^T$ and the isolates at the genus level from other previously known *Actinomadura* species and members of *Thermomonosporaceae*. Therefore, a new genus, *Actinoallomurus* gen. nov., is proposed for *Actinomadura spadix* and the new isolates.

Based on DNA–DNA relatedness and certain phenotypic properties, nine species of the genus *Actinoallomurus* are described; discriminatory properties of these species are listed in Supplementary Table S2 and Table 2. The type species of the genus is *Actinoallomurus spadix* gen. nov., comb. nov. Strains TT02-47$^T$, TT02-22 and TT99-68 were identified as members of a single species, *Actinoallomurus iriomotensis* sp. nov. The other species of the genus *Actinoallomurus* are *Actinoallomurus amamiensis* sp. nov., *Actinoallomurus caesius* sp. nov., *Actinoallomurus coprocola* sp. nov., *Actinoallomurus fulvus* sp. nov., *Actinoallomurus luridus* sp. nov., *Actinoallomurus purpureus* sp. nov. and *Actinoallomurus yoronensis* sp. nov.

Although the presence of lysine in the cell-wall peptidoglycan is characteristic of the new genus, the structure of the peptidoglycan, including the position of lysine, has not been determined. In phylogenetic trees constructed on the basis of both the neighbour-joining and maximum-likelihood methods, *Actinomadura alba* was placed phylogenetically outside the cluster composed of the genera *Actinomadura* and *Actinocorallia*. Therefore, further taxonomic study is required in order to clarify the taxonomy of the genus *Actinomadura*.

## Description of *Actinoallomurus* gen. nov.

*Actinoallomurus* (Ac.ti.no.al.lo.mu′rus. Gr. n. *actis*, *actinos* ray, used to refer to actinomycetes; Gr. adj. *allos* different; L. masc. n. *murus* wall; N.L. masc. n. *Actinoallomurus* actinomycetes with a different wall).

Non-acid-fast and strictly aerobic actinomycetes that show extensive branching and non-fragmenting substrate hyphae. Spore chains develop on the tips of the aerial mycelium. Spores form oval to short rods (0.5–1.2 μm wide and 0.7–2.0 μm long). Spores are non-motile. The surface of colonies is powdery. Strains show good growth on most test media; however, they show extremely poor sporulation on these media. The optimum temperature for growth is generally 25–30 °C. The cell wall contains D-glutamic acid, D- and L-alanine, D- and L-lysine and *meso*-$A_2$pm. Whole-cell hydrolysates contain madurose and galactose. Cellular fatty acids comprise iso-branched, anteiso-branched, 10-methylated branched saturated and unsaturated fatty acids; iso-$C_{16:0}$ is the major component. MK-9($H_6$) and MK-9($H_8$) are the predominant menaquinones. Phosphatidylglycerol and diphosphatidylglycerol are detected; phosphatidylethanolamine, phosphatidylcholine and ninhydrin-positive unidentified phospholipids are not detected (phospholipid pattern type PI). Mycolic acids are not detected. The acyl type of the muramic acid is *N*-acetyl. The G+C content of DNA is 69–74 mol%. Unique

**Table 2.** Differential characteristics of type strains of the genus *Actinoallomurus* gen. nov.

Strains: 1, *Actinoallomurus spadix* NBRC 14099$^T$; 2, *Actinoallomurus amamiensis* TT00-28$^T$; 3, *Actinoallomurus caesius* A3015$^T$; 4, *Actinoallomurus coprocola* TT04-09$^T$; 5, *Actinoallomurus fulvus* TT99-66$^T$; 6, *Actinoallomurus iriomotensis* TT02-47$^T$; 7, *Actinoallomurus luridus* TT02-15$^T$; 8, *Actinoallomurus purpureus* TTN02-30$^T$; 9, *Actinoallomurus yoronensis* TTN02-22$^T$. +, Positive; −, negative; v, variable; w, weakly positive.

| Characteristic | 1 | 2 | 3 | 4 | 5 | 6 | 7 | 8 | 9 |
|---|---|---|---|---|---|---|---|---|---|
| Nitrate reduction | + | − | − | − | − | v | − | − | − |
| Pyrazinamidase | + | + | − | + | + | + | + | + | + |
| Pyrrolidonyl arylamidase | − | − | − | − | + | − | + | − | − |
| Urea hydrolysis | − | − | − | + | − | − | − | − | + |
| Catalase | + | + | + | + | + | + | − | + | + |
| Alkaline phosphatase | + | − | + | v | + | + | v | − | v |
| Valine arylamidase | + | − | − | − | − | − | − | − | − |
| Acid phosphatase | + | + | + | + | + | + | + | − | − |
| Phosphohydrolase | + | + | + | − | + | + | + | w | + |
| β-Galactosidase | v | − | − | v | − | − | − | v | v |
| β-Glucuronidase | − | − | v | v | v | v | + | − | + |
| α-Glucosidase | + | − | − | v | v | v | v | v | v |
| β-Glucosidase | + | − | − | − | − | − | − | − | − |
| *N*-Acetyl-β-glucosaminidase | v | v | v | v | + | + | + | v | + |
| Utilization of: | | | | | | | | | |
| Glycerol | + | + | + | − | + | + | + | + | + |
| Erythritol | + | − | − | − | + | + | − | − | + |
| D-Arabinose | + | + | − | + | + | + | − | − | + |
| L-Arabinose | + | + | − | − | + | + | + | − | + |
| L-Xylose | + | + | − | + | + | + | − | − | + |
| Methyl β-D-xylopyranoside | + | + | − | + | + | v | + | − | − |
| L-Sorbose | − | − | − | − | + | v | − | − | + |
| L-Rhamnose | + | + | + | + | + | + | + | − | + |
| Dulcitol | + | + | − | + | + | + | − | − | + |
| Inositol | + | + | − | + | + | + | − | − | + |
| D-Sorbitol | + | + | − | + | + | v | − | − | + |
| Methyl α-D-mannopyranoside | + | + | − | + | + | + | + | + | + |
| Methyl α-D-glucopyranoside | + | − | − | − | + | + | − | + | − |
| Amygdalin | + | + | − | + | + | + | + | + | + |
| Arbutin | + | + | − | + | + | v | + | + | + |
| Salicin | + | + | − | + | + | v | + | + | + |
| Cellobiose | + | + | − | + | + | + | + | + | + |
| Maltose | + | + | w | + | + | + | + | + | + |
| Lactose | + | + | − | + | + | + | + | + | + |
| Melibiose | + | + | − | + | + | + | + | − | + |
| Sucrose | + | + | − | + | + | + | + | + | + |
| Inulin | + | + | − | + | + | v | − | − | + |
| Melezitose | + | + | − | − | + | v | + | + | + |
| Starch | + | + | − | + | + | + | + | + | + |
| Glycogen | + | + | − | + | + | + | + | + | + |
| Xylitol | + | + | − | − | + | v | − | − | + |
| Turanose | + | + | − | + | + | + | + | + | + |
| D-Lyxose | + | + | − | − | + | + | + | − | + |
| D-Tagatose | + | + | − | − | + | + | + | − | + |
| D-Fucose | + | − | − | − | + | v | − | − | + |
| L-Fucose | + | + | − | − | + | + | + | + | + |
| D-Arabitol | + | + | − | + | + | v | − | − | + |
| L-Arabitol | + | + | − | − | + | v | − | − | + |
| 2-Ketogluconate | + | + | + | + | + | + | + | − | + |
| 5-Ketogluconate | + | + | − | − | + | v | + | − | + |

nucleotide signatures are present at the following positions of the 16S rRNA gene (corresponding to the numbering of the *Escherichia coli* 16S rRNA gene; Brosius *et al.*, 1978): 128 (T), 141 : 222 (G–C), 415 (A), 442 : 492 (C–G), 445 : 488 (C–G), 446 : 489 (C–G), 493 (G), 602 : 636 (C–G), 610 (A), 614 : 626 (G–C), 615 : 625 (A–T), 630 (C), 657 : 749 (G–C), 658 : 748 (T–A), 659 : 747 (A–T), 854 (T), 990(T), 1159 (T) and 1262 (A). The genus belongs to the family *Thermomonosporaceae*. The type species is *Actinoallomurus spadix*.

In addition to those given in the genus description, the morphological, chemotaxonomic and general characteristics of each species are as follows.

## Description of *Actinoallomurus spadix* (Nonomura and Ohara 1971) comb. nov.

*Actinoallomurus spadix* (spa′dix. L. masc. adj. *spadix* chestnut-brown).

Basonym: *Actinomadura spadix* Nonomura and Ohara 1971.

The description is based on data provided by Nonomura & Ohara (1971). The colour of the aerial mycelium is white. The tips of the aerial mycelium divide into oval to rod-shaped, irregular-sized spores (0.5–1.0 × 0.7–1.5 μm) at maturity. The mass colour of the substrate mycelium is yellowish brown to purplish red. Spore chains form spirals or loops. Spores are smooth. The substrate mycelium is long, well-developed and branched. Grows at 20–45 °C, at pH 5.0–7.0 and at up to 2 % NaCl. Glycerol, L-arabinose, D-ribose, D-xylose, D-adonitol, methyl β-D-xylopyranoside, D-galactose, D-glucose, D-fructose, D-mannose, L-rhamnose, D-mannitol, methyl α-D-mannopyranoside, methyl α-D-glucopyranoside, *N*-acetylglucosamine, amygdalin, cellobiose, maltose, lactose, melibiose, sucrose, trehalose, melezitose, raffinose, glycogen, xylitol, turanose, D-lyxose, L-fucose and gluconate are utilized as sole carbon sources. Tests for nitrate reduction, aesculin hydrolysis, gelatin hydrolysis and catalase, pyrazinamidase, alkaline phosphatase, β-galactosidase, α-glucosidase, leucine arylamidase, valine arylamidase, acid phosphatase, phosphohydrolase, β-glucosidase and β-mannosidase activities are positive. Major cellular fatty acids are iso-$C_{16:0}$, anteiso-$C_{17:0}$ and iso-$C_{18:0}$. The G + C content of the DNA of the type strain is 74 mol%.

The type strain is NBRC 14099$^T$ (=ATCC 27298$^T$ =BCRC 13386$^T$ =CBS 261.72$^T$ =CIP 105479$^T$ =DSM 43459$^T$ =JCM 3146$^T$ =KCTC 9252$^T$ =NCIMB 11118$^T$ =NRRL B-16128$^T$).

## Description of *Actinoallomurus amamiensis* sp. nov.

*Actinoallomurus amamiensis* (a.ma′mi.en′sis. N.L. masc. adj. *amamiensis* pertaining to Amami Island, Japan, where the organism was originally isolated).

The colour of the aerial mycelium is white. The tips of the aerial mycelium divide into oval to rod-shaped, irregular-sized spores (0.5–1.0 × 0.7–1.5 μm) at maturity. The mass colour of the substrate mycelium is pale yellow to reddish brown. Spore chains form loops or spirals. Spores are smooth. The substrate mycelium is long, well-developed and branched. Grows at 20–37 °C, at pH 5.0–6.0 and at up to 2 % NaCl. Glycerol, D- and L-arabinose, D-ribose, D-xylose, D-adonitol, D-galactose, D-glucose, D-fructose, D-mannose, L-rhamnose, D-mannitol, methyl α-D-mannopyranoside, *N*-acetylglucosamine, salicin, cellobiose, maltose, lactose, melibiose, sucrose, trehalose, raffinose, starch, glycogen, gentiobiose, turanose, L-fucose and gluconate are utilized as sole carbon sources. Tests for aesculin hydrolysis, gelatin hydrolysis and catalase, pyrazinamidase, leucine arylamidase, acid phosphatase and phosphohydrolase activities are positive. Major cellular fatty acids are iso-$C_{16:0}$ and anteiso-$C_{17:0}$. The G + C content of the DNA of the type strain is 72 mol%.

The type strain is TT00-28$^T$ (=NBRC 103682$^T$ =KCTC 19537$^T$), which was isolated from soil of a citrus grove on Amami Island, Kagoshima, Japan.

## Description of *Actinoallomurus caesius* sp. nov.

*Actinoallomurus caesius* (cae′si.us. L. masc. adj. *caesius* grey–blue).

The colour of the aerial mycelium is white. The tips of the aerial mycelium divide into oval to rod-shaped, irregular-sized spores (0.8–1.0 × 0.8–1.5 μm) at maturity. The mass colour of the substrate mycelium is pale yellow to brown or yellowish pink. Spore chains form loops or spirals. Spores are smooth. The substrate mycelium is long, well-developed and branched. Grows at 20–37 °C, at pH 5.0–7.0 and at up to 2 % NaCl. Glycerol, D-ribose, D-adonitol, D-galactose, D-glucose, D-fructose, D-mannose, L-rhamnose, D-mannitol, *N*-acetyl-glucosamine, aesculin ferric citrate, trehalose, raffinose, gentiobiose, gluconate and 2-ketogluconate are utilized as sole carbon sources. Tests for aesculin hydrolysis, gelatin hydrolysis and catalase, alkaline phosphatase, leucine arylamidase, acid phosphatase and phosphohydrolase activities are positive. Major cellular fatty acids are iso-$C_{16:0}$ and 10-methyl $C_{17:0}$. The G + C content of the DNA of the type strain is 71 mol%.

The type strain is A3015$^T$ (=NBRC 103678$^T$ =KCTC 19535$^T$), which was isolated from mangrove forest soil on Iriomote Island, Okinawa, Japan.

## Description of *Actinoallomurus coprocola* sp. nov.

*Actinoallomurus coprocola* (cop.ro′co.la. Gr. n. *kopros* faeces; L. suffix *-cola* inhabitant of; N.L. masc. n. *coprocola* inhabitant of faeces).

The colour of the aerial mycelium is white. The tips of the aerial mycelium divide into oval to rod-shaped, irregular-sized spores (0.5–1.0 × 0.7–1.5 μm) at maturity. The mass colour of the substrate mycelium is pale yellow to purplish red. Spore chains form compact loops or spirals. Spores are

wrinkled to smooth. The substrate mycelium is long, well-developed and branched. Grows at 20–37 °C, at pH 5.0–7.0 and at up to 3 % NaCl. D-Arabinose, D-ribose, D-galactose, D-glucose, D-fructose, D-mannitol, methyl α-D-mannopyranoside, *N*-acetylglucosamine, cellobiose, maltose, lactose, melibiose, sucrose, trehalose, inulin, raffinose, starch, glycogen, gentiobiose, turanose and D-arabitol are utilized as sole carbon sources. Tests for aesculin hydrolysis, urea hydrolysis, gelatin hydrolysis and catalase, pyrazinamidase, leucine arylamidase and acid phosphatase activities are positive. Major cellular fatty acids are iso-$C_{16:0}$ and anteiso-$C_{17:0}$. The G+C content of the DNA of the type strain is 74 mol%.

The type strain is TT04-09$^T$ (=NBRC 103688$^T$ =KCTC 19542$^T$), which was isolated from cow dung at Futtsu, Chiba, Japan.

### Description of *Actinoallomurus fulvus* sp. nov.

*Actinoallomurus fulvus* (ful′vus. L. masc. adj. *fulvus* yellowish brown).

The colour of the aerial mycelium is white. The tips of the aerial mycelium divide into oval to rod-shaped, irregular-sized spores (0.7–1.2 × 0.8–1.5 μm) at maturity. The mass colour of the substrate mycelium is pale yellow to reddish brown. Spore chains form loops or spirals. Spores are smooth. The substrate mycelium is long, well-developed and branched. Grows at 20–37 °C, at pH 5.0–7.0 and at up to 2 % NaCl. Glycerol, D- and L-arabinose, D-ribose, D-xylose, D-adonitol, D-galactose, D-glucose, D-fructose, D-mannose, L-rhamnose, inositol, methyl α-D-mannopyranoside, *N*-acetylglucosamine, amygdalin, salicin, cellobiose, maltose, lactose, melibiose, sucrose, trehalose, inulin, raffinose, starch, glycogen, turanose, D-lyxose, L-fucose and gluconate are utilized as sole carbon sources. Tests for aesculin hydrolysis, gelatin hydrolysis and catalase, pyrazinamidase, pyrrolidonyl arylamidase, alkaline phosphatase, leucine arylamidase, acid phosphatase, phosphohydrolase and *N*-acetyl-β-glucosaminidase activities are positive. Major cellular fatty acids are iso-$C_{16:0}$ and anteiso-$C_{17:0}$. The G+C content of the DNA of the type strain is 72 mol%.

The type strain is TT99-66$^T$ (=NBRC 103680$^T$ =KCTC 19536$^T$), which was isolated from soil of a sugar-cane field on Iriomote Island, Okinawa, Japan.

### Description of *Actinoallomurus iriomotensis* sp. nov.

*Actinoallomurus iriomotensis* (i′ri.o.mo.ten′sis. N.L. masc. adj. *iriomotensis* pertaining to Iriomote Island, Japan, where the organism was originally isolated).

The colour of the aerial mycelium is white. The tips of the aerial mycelium divide into oval to rod-shaped, irregular-sized spores (0.5–0.7 × 0.6–1.2 μm) at maturity. The mass colour of the substrate mycelium is yellow to orange. Spore chains form compact hooks or spirals. Spores are wrinkled to smooth. The substrate mycelium is long, well-developed and branched. Grows at 20–37 °C, at pH 5.0–6.0 and at up to 2 % NaCl. Glycerol, D- and L-arabinose, D-ribose, D-xylose, D-adonitol, D-galactose, D-glucose, D-fructose, D-mannose, D-mannitol, *N*-acetylglucosamine, cellobiose, lactose, melibiose, raffinose, starch, glycogen, gentiobiose and L-fucose are utilized as sole carbon sources. Tests for aesculin hydrolysis, gelatin hydrolysis and catalase, pyrazinamidase, alkaline phosphatase, leucine arylamidase, acid phosphatase, phosphohydrolase and *N*-acetyl-β-glucosaminidase activities are positive. Major cellular fatty acids are iso-$C_{16:0}$ and anteiso-$C_{17:0}$. The G+C content of the DNA of the three known strains is 71–73 mol%.

The type strain is TT02-47$^T$ (=NBRC 103685$^T$ =KCTC 19539$^T$), which was isolated from meadow soil on Iriomote Island, Okinawa, Japan. Strains TT99-68 (=NBRC 103681) and TT02-22 (=NBRC 103684), isolated from soil on the same island, are also strains of this species.

### Description of *Actinoallomurus luridus* sp. nov.

*Actinoallomurus luridus* (lu′ri.dus. L. masc. adj. *luridus* pale yellow).

The colour of the aerial mycelium is white. The tips of the aerial mycelium divide into oval to rod-shaped, irregular-sized spores (0.5–1.0 × 0.7–1.5 μm) at maturity. The mass colour of the substrate mycelium is pale yellow to yellowish brown. Spore chains form loops or spirals. Spores are wrinkled to smooth. The substrate mycelium is long, well-developed and branched. Grows at 20–45 °C, at pH 5.0–7.0 and at up to 2 % NaCl. D-Ribose and D-xylose are utilized as sole carbon sources. Tests for aesculin hydrolysis, gelatin hydrolysis and pyrazinamidase, pyrrolidonyl arylamidase, leucine arylamidase, acid phosphatase, phosphohydrolase, β-glucuronidase and *N*-acetyl-β-glucosaminidase activities are positive. Major cellular fatty acids are iso-$C_{16:0}$ and anteiso-$C_{17:0}$. The G+C content of the DNA of the type strain is 72 mol%.

The type strain is TT02-15$^T$ (=NBRC 103683$^T$ =KCTC 19538$^T$), which was isolated from meadow soil on Iriomote Island, Okinawa, Japan.

### Description of *Actinoallomurus purpureus* sp. nov.

*Actinoallomurus purpureus* (pur.pu′re.us. L. masc. adj. *purpureus* purple coloured).

The colour of the aerial mycelium is white. The tips of the aerial mycelium divide into oval to rod-shaped, irregular-sized spores (0.5–1.0 × 0.7–1.5 μm) at maturity. The mass colour of the substrate mycelium is pale yellow to reddish purple. Spore chains form loops or spirals. Spores are smooth. The substrate mycelium is long, well-developed and branched. Grows at 15–30 °C, at pH 5.0–7.0 and at up to 5 % NaCl. D-Galactose, D-glucose, D-fructose, D-mannose, *N*-acetylglucosamine, cellobiose, maltose, sucrose, trehalose, starch, glycogen, gentiobiose, turanose, L-fucose and gluconate are utilized as sole carbon sources. Tests for aesculin hydrolysis, gelatin hydrolysis and

catalase, pyrazinamidase, β-galactosidase, α-glucosidase and *N*-acetylglucosaminidase activities are positive. Major cellular fatty acids are iso-$C_{16:0}$ and anteiso-$C_{17:0}$. The G+C content of the DNA of the type strain is 70 mol%.

The type strain is TTN02-30$^T$ (=NBRC 103687$^T$ =KCTC 19541$^T$), which was isolated from forest soil in the Yoro valley, Chiba, Japan.

### Description of *Actinoallomurus yoronensis* sp. nov.

*Actinoallomurus yoronensis* (yo.ro.nen′sis. N.L. masc. adj. *yoronensis* pertaining to the Yoro valley, Japan, where the organism was originally isolated).

The colour of the aerial mycelium is white. The tips of the aerial mycelium divide into oval to rod-shaped, irregular-sized spores (0.5–0.8 × 0.7–1.5 μm) at maturity. The mass colour of the substrate mycelium is yellow to reddish brown. Spore chains form loops or spirals. Spores are smooth. The substrate mycelium is long, well-developed and branched. Grows at 15–37 °C, at pH 5.0–6.0 and at up to 1% NaCl. Glycerol, D-glucose, D-ribose, D-xylose, D-mannitol, maltose and sucrose are utilized as sole carbon sources. Tests for aesculin hydrolysis, urea hydrolysis, gelatin hydrolysis and catalase, pyrazinamidase, leucine arylamidase, phosphohydrolase, β-glucuronidase and *N*-acetyl-β-glucosaminidase activities are positive. Major cellular fatty acids are iso-$C_{16:0}$ and anteiso-$C_{17:0}$. The G+C content of the DNA of the type strain is 69 mol%.

The type strain is TTN02-22$^T$ (=NBRC 103686$^T$ =KCTC 19540$^T$), which was isolated from forest soil in the Yoro valley, Chiba, Japan.

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