Labrys okinawensis sp. nov. and *Labrys miyagiensis* sp. nov., budding bacteria isolated from rhizosphere habitats in Japan, and emended descriptions of the genus *Labrys* and *Labrys monachus*

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Three strains, MAFF 210191^T, G24103^T and G24116, assumed to be members of two novel species, were isolated from several rhizosphere habitats in different parts of Japan. Phylogenetic analysis based on 16S rRNA gene sequences revealed that the isolates formed a distinct monophyletic group together with the two known species of the genus *Labrys*, suggesting that the isolates have a close affiliation with this genus. In this study, a polyphasic approach was used to characterize and compare the three isolates with the two species of the genus *Labrys*, *Labrys monachus* and *Labrys methylaminiphilus*. All three isolates were aerobic, Gram-negative, motile and non-sporulating and they ranged in shape from spherical to short rods. The cells multiplied by budding and utilized a wide variety of monosaccharides, disaccharides and sugar alcohols as sole carbon and energy sources, but they did not utilize C₁ compounds, salicin or D-melezitose. The strains were inhibited by DL- α -alanine and glycine (both at 10 mM). The major cellular fatty acids were C_{19:0} cyclo ω 8c, C_{16:0}, C_{18:0} and C_{18:1} ω 7c. The three isolates shared < 12 % and < 11 % DNA–DNA relatedness with *L. monachus* DSM 5896^T and *L. methylaminiphilus* DSM 16812^T, respectively. The G + C content of the isolates (61–62 mol%) was also significantly lower than those of the two previously characterized species. In spite of many morphological, physiological and chemotaxonomic similarities among the three isolates, strain MAFF 210191^T could be differentiated from strains G24103^T and G24116 on the basis of 16S rRNA gene sequence divergence, DNA–DNA relatedness (< 46 %) and gelatin hydrolysis. Two novel species are therefore proposed, namely *Labrys okinawensis* sp. nov., with the type strain MAFF 210191^T (= DSM 18385^T), and *Labrys miyagiensis* sp. nov., with the type strain G24103^T (= NBRC 101365^T = NCIMB 14143^T) and also including strain G24116 (= NBRC 101366 = NCIMB 14144). Emended descriptions of the genus *Labrys* and *Labrys monachus* are also presented.

The GenBank/EMBL/DDBJ accession numbers for the nearly complete 16S rRNA gene sequences of strains MAFF 210191^T, G24103^T, G24116 and *Labrys methylaminiphilus* DSM 16812^T are AB236169–AB236172, respectively.

Tables showing substrate assimilation, cellular fatty acid profiles and DNA–DNA hybridization results for the three isolates and representative *Labrys* species are available as supplementary material in IJSEM Online.

Rhizosphere and rhizoplane bacteria, which generally represent a subset of soil micro-organisms, have various important beneficial, neutral and harmful effects on the growth and development of numerous plants (Lynch, 1990). Taxonomic studies of these micro-organisms are significant from both agricultural and ecological points of view. Assessment of the structures of rhizosphere microbial populations has long been a focus of scientific interest and numerous root-associated bacteria of both diazotrophic

and non-diazotrophic populations have been described to date; however, much remains unknown regarding their biodiversity in this important habitat. This lack of information is in part because fast-growing bacteria often outgrow the slow growers and also because many bacteria are inhibited by the high concentration of ingredients in the ordinary laboratory media used for isolation and detection (Janssen *et al.*, 2002).

Strain MAFF 210191^T, supplied by the Ministry of Agriculture, Food and Forestry of Japan, was isolated from the root-nodule of *Entada phaseoloides*, a legume found in Okinawa, Japan. In addition, in the course of investigating the bacterial population inhibited by nutrient broth but capable of growing in 100-fold-diluted nutrient broth, strains G24103^T and G24116 were isolated from a grassland soil at Sendai in Miyagi, Japan (El-Beltagy & Hattori, 1994). Preliminary investigations based on partial 16S rRNA gene sequences showed that these three strains were similar to members of the genus *Labrys* (Vasil'eva & Semenov, 1984).

The genus *Labrys* was first described by Vasil'eva & Semenov (1984) based on *Labrys monachus* VKM B-1479^T (= DSM 5896^T), which was isolated from silt of Lake Mustijärv in Estonia. Although the genus was placed in the *Alpha-proteobacteria* on the basis of the 16S rRNA gene sequence, its phylogenetic position at the family level remained obscure because of the unavailability of strains (Fritz *et al.*, 2004). The genus remained monospecific until recently, when Miller *et al.* (2005) reported the isolation of another strain, JLW10^T (= DSM 16812^T) from sediment of Lake Washington, Seattle, WA, USA, and proposed a second species, *Labrys methylophilus*. In the present study, the taxonomy of strains MAFF 210191^T, G24103^T and G24116 was studied and two novel species are proposed. Emended descriptions of the genus *Labrys* and *L. monachus* are also proposed.

The three isolates were cultured in modified yeast extract-mannitol broth (YMB) containing (l⁻¹, pH 7.0, 28 °C) 0.5 g yeast extract, 10 g mannitol, 0.5 g K₂HPO₄, 0.2 g NaCl, 0.2 g CaCl₂·2H₂O and 0.1 g MgSO₄·7H₂O. For long-term maintenance, cells grown in slant cultures were suspended thoroughly in 12–15 % glycerol, transferred to sterilized serum tubes and stored at –85 °C. *L. monachus* DSM 5896^T and *L. methylophilus* DSM 16812^T, obtained from the DSMZ, were used as reference strains.

Phase-contrast microscopy revealed that all three isolates were Gram-negative, non-sporulating, spherical to short rods and motile. They all occurred singly or in pairs and multiplied by budding. Cells grown at 28 °C formed visible colonies on yeast extract-mannitol agar (YMA, pH 7.0; recipe as for YMB plus 15 g agar l⁻¹) plates within 3–4 days. Colonies were 1–2 mm in diameter, circular, white to greyish, convex, opaque and viscous in consistency.

Genomic DNA was isolated as described by Ausubel *et al.* (1995). To obtain high-molecular-mass preparations for

hybridization experiments, genomic DNA was further purified by equilibrium ultracentrifugation in CsCl/ethidium bromide gradients according to the method of Hamamoto & Nakase (1995), using an ultracentrifuge (Hitachi CS-210) at 400 000 g for 16 h. The quality of DNA was verified by spectrophotometric determination of A₂₆₀/A₂₈₀, which was at least 1.8.

The 16S rRNA gene fragment was amplified as described by Normand *et al.* (1996) using Ex Taq polymerase (TaKaRa Shuzo) in a Gene Amp PCR system 9700 (PE Applied Biosystems). PCR products were purified using a QIAGEN PCR purification kit. DNA sequences were determined with the BigDye v. 3.1 Terminator Cycle Sequencing kit (PE Applied Biosystems) using an ABI PRISM 3100 Genetic Analyzer (PE Applied Biosystems). Sequence data were analysed as described by Kawasaki *et al.* (1993) using the ABI PRISM sequence analysis program and were assembled using the ABI Auto Assembler (Perkin Elmer).

The 16S rRNA gene sequences of seven soil- and plant-related bacteria, which might be relevant to taxonomic studies of members of the genus *Labrys*, were retrieved from the NCBI databases (<http://www.ncbi.nlm.nih.gov/>). The 16S rRNA gene sequence of *L. methylophilus* DSM 16812^T obtained from the database differed slightly from that determined here. The 16S rRNA gene sequence determined in the present study was used in the phylogenetic analysis. The 16S rRNA gene sequences, along with those of the three isolates, were aligned and ambiguous sites were eliminated manually prior to the construction of a phylogenetic tree by the neighbour-joining method with CLUSTAL X (Thompson *et al.*, 1997; Jeanmougin *et al.*, 1998). The robustness of individual branches was estimated by bootstrap analysis with 1000 replicates. In the phylogenetic tree, the three isolates, *L. monachus* DSM 5896^T and *L. methylophilus* DSM 16812^T formed a monophyletic group with a maximum bootstrap value, which confirms that the isolates are members of the genus *Labrys* (Fig. 1). Strains G24103^T and G24116 had identical 16S rRNA gene sequences, which showed 98.8, 98.4 and 98.5 % similarity to those of strain MAFF 210191^T, *L. monachus* DSM 5896^T and *L. methylophilus* DSM 16812^T, respectively. On the other hand, the 16S rRNA gene sequence of MAFF 210191^T respectively showed 97.5 and 98.4 % similarity to those of *L. monachus* DSM 5896^T and *L. methylophilus* DSM 16812^T.

The ability of the isolates to utilize various carbon compounds as sole carbon and energy sources was tested in YMB basal medium (YMB with 1/10 yeast extract and without mannitol). Stock solution of each substrate was added to the basal medium at a final concentration of 0.2 % (w/v). The mixture was adjusted to pH 7.0, filter-sterilized (0.45 µm; Millipore), inoculated with washed cells, incubated at 28 °C and observed for growth for 2 weeks. Growth was confirmed by both observation with the naked eye and spectrophotometry (OD₆₀₀). The ability to utilize nitrogen sources was investigated in a similar manner in

Fig. 1. Neighbour-joining phylogenetic tree based on 16S rRNA gene sequences indicating the position of strains MAFF 210191^T, G24103^T and G24116 with relevant soil- and plant-associated bacteria. Bar, 5 substitutions per 100 nucleotide positions.

yeast extract-free basal liquid medium (as described above) at a final concentration of 0.1 % (w/v) for each tested substrate. The ability to utilize C₁ compounds was tested on mono-, di- or trimethylamine-supplemented minimal medium (Harder *et al.*, 1973; Miller *et al.*, 2005). Data regarding carbon and nitrogen source utilization are summarized partly in Table 1 and partly in Supplementary Table S1 (available in IJSEM Online). Sensitivity to antibiotics was determined by streaking the cells in triplicate on YMA plates containing the following antibiotics: erythromycin (10 µg ml⁻¹), ampicillin (50 µg ml⁻¹), penicillin (100 µg ml⁻¹), kanamycin (10 µg ml⁻¹), chloramphenicol (50 µg ml⁻¹) and tetracycline (10 µg ml⁻¹). Growth was assessed after 2 weeks of incubation. With the exception of erythromycin, the three isolates and *L. monachus* DSM 5896^T were sensitive to all of the tested antibiotics. The three isolates could be differentiated from *L. monachus* DSM 5896^T by the assimilation of sucrose, D-cellobiose and dulcitol as sole carbon and energy sources and utilization of L-methionine, L-proline, L-serine, L-tyrosine and L-phenylalanine as sole nitrogen sources. The three isolates could be distinguished from *L. methylaminiphilus* DSM 16812^T by the following characteristics: they were not able to utilize C₁ compounds, they could not assimilate DL- α -alanine or glycine as sole nitrogen source, they were unable to grow on Luria-Bertani agar (LB), tryptic soy agar (Difco) or tryptone-glucose-yeast extract agar [TGY, pH 7.0, containing (l⁻¹) 5 g tryptone, 1 g glucose, 2.5 g yeast extract and 15 g agar], they were inhibited by 10 mM DL- α -alanine and glycine and they were sensitive to ampicillin, penicillin, kanamycin, chloramphenicol and tetracycline.

Growth temperatures, pH range and tolerance of NaCl were tested in triplicate in YMB tubes. Citrate utilization, starch and gelatin hydrolysis and 3-ketolactose production from lactose oxidation were tested according to Smibert & Krieg (1994). For cellular fatty acid analysis, all strains were grown on modified YMB for 4 days at 28 °C. The fatty acids were analysed with the Sherlock MIDI (Microbial Identification) system (Sasser, 1990; Tighe *et al.*, 2000). Analyses were based on the conversion of fatty acids to fatty acid methyl esters by mild acidic methanolysis. The methyl ester derivatives thus produced were detected by GLC, followed by data analysis with Sherlock MIS software. Quinone analysis was performed as described by Yamada *et al.* (1969). The cellular fatty acid data of the three isolates are summarized and

compared with those of the two reference strains in Supplementary Table S2. The cellular fatty acid profiles of the three isolates were largely consistent with those of *L. monachus* DSM 5896^T and *L. methylaminiphilus* DSM 16812^T (Fritz *et al.*, 2004; Miller *et al.*, 2005). In contrast to *L. monachus* DSM 5896^T, the amount of C_{18:0} 3-OH was larger than that of C_{16:0} 3-OH in the three isolates. On the other hand, neither of the above hydroxy acids was found in *L. methylaminiphilus* DSM 16812^T and only a trace amount of C_{17:0} 3-OH (0.40 %) was detected. However, the clear dominance of C_{19:0} cyclo ω 8c fatty acid (> 53 %) in all of the strains reported so far is unique, and is potentially a marker for the genus *Labrys*.

For the N₂ fixation experiment, jellified Winogradsky's N₂-free medium (Hashidoko *et al.*, 2002; Tchan & New, 1984) was designed. A mineral mixture was prepared containing (g l⁻¹): KH₂PO₄, 50; MgSO₄·7H₂O, 25; NaCl, 25; FeSO₄·7H₂O, 1; Na₂MoO₄·2H₂O, 1; and MnSO₄·4H₂O, 1. The mixture was adjusted to pH 7.2 with solid NaOH and added to (5 ml l⁻¹) a solution of sugar (10 g mannitol l⁻¹) and powdery CaCO₃ (0.1 g l⁻¹), adjusted to pH 7.0 with 2 M H₂SO₄ and filtered (0.45 µm; Millipore). The resulting filtrate was mixed with gellan gum (0.3 %, w/v), dissolved by heating, dispensed into tubes, sterilized and cooled appropriately to make soft gels. Cells were washed, suspended in sterile water, inoculated into the tubes and vortexed before incubation at 28 °C. Results were recorded after 2 weeks of incubation. To search for *nifH* (encoding the iron protein of nitrogenase) and *nodA* (encoding the N-acyl transferase, a key nod factor in nodulation) genes, Southern hybridization was performed as described by Sambrook *et al.* (1989). Although the strains showed clear growth in Winogradsky's N₂-free mineral medium, attempts to amplify or hybridize with the *nifH* and *nodA* genes failed with the specific primers and probes used (Zehr & McReynolds, 1989). These findings are consistent with results obtained with *L. monachus* DSM 5896^T and *L. methylaminiphilus* DSM 16812^T, which were isolated from lake sediment; both were reported to be unable to fix nitrogen. In contrast, strain MAFF 210191^T was isolated from a root nodule of the legume *Entada phaseoloides* and strains G24103^T and G24116 were isolated from a grassland soil, which is suggestive of symbiotic or associative nitrogen-fixing capability. Therefore, our failure to detect the *nif* or *nod* genes might be explained by the presence of distantly related N₂-fixing gene machineries that

Table 1. Differential characteristics of strains MAFF 210191^T, G24103^T and G24116 and the two representatives of the genus *Labrys*

Strains: 1, *L. okinawensis* sp. nov. MAFF 210191^T (this study); 2, *L. miyagiensis* sp. nov. strains G24103^T and G24116 (this study); 3, *L. monachus* DSM 5896^T (data from this study unless indicated); 4, *L. methylaminiphilus* DSM 16812^T (data from Miller *et al.*, 2005). –, Not detectable; V, variable; W, weak; NR, not reported.

Characteristic	1	2	3	4
Source of isolation	Root module	Soil	Lake sediment ^{a*}	Lake sediment
Cell shape	Spherical to short rods	Spherical to short rods	Flat, triangular with radial symmetry	Rods
Motility	Motile	Motile	Non-motile	Non-motile
Growth pH:				
Range	4.0–9.0	4.0–9.0	4.0–9.0	4.0–9.5†
Optimum	6.0–7.5	6.0–7.5	6.0–7.5	5.0–7.0
Temperature range for growth (°C)	15–32	15–32	20–50 ^b	10–35
Utilization as sole carbon and energy source:				
Sucrose	+	+	–	+
D-Cellobiose	+	+	–	NR
Dulcitol	+	+	–	NR
Utilization of C ₁ compounds:				
Methylamine	–	–	W ^a	+
Dimethylamine	–	–	NR	+
Trimethylamine	–	–	– ^a	+
Utilization as sole nitrogen source:				
DL- α -Alanine	–	–	–	+
Glycine	–	–	–	+
L-Glutamic acid	–	–	–	+
L-Tryptophan	+	+	+	–
L-Arginine	+	V	–	+
Catalase	W	W	+	+
Hydrolysis of gelatin	+	–	–	W
G + C content (mol%)	62.3	61.0–61.4	65.0	65.7

*Data from other studies as follows: *a*, Vasil'eva & Semenov (1984); *b*, Miller *et al.* (2005).

†Growth at pH 10.0 was observed in this study.

remained undetected because of non-optimal reaction conditions or due to the presence of a novel system yet to be identified.

The G + C contents were determined by HPLC (Hitachi LaChrom L-7100) separation as described by Tamaoka & Komagata (1984). The G + C contents were 62.3, 61.4 and 61.0 mol% for MAFF 210191^T, G24103^T and G24116, respectively. The G + C content of the reference strain *L. monachus* DSM 5896^T was 65.0 mol%, which is lower than that previously reported (67.9 mol%) by Vasil'eva & Semenov (1984), who used the DNA melting-point method for the determination of G + C content. The G + C contents of the three isolates (61–62 mol%) were noticeably lower than those of both *L. monachus* DSM 5896^T (65.0 mol%) and *L. methylaminiphilus* DSM 16812^T (65.7 mol%).

DNA–DNA hybridization was carried out according to the photobiotin microplate method as described by Ezaki *et al.*

(1989) (see Supplementary Table S3). Hybridizations were performed on immunoplates (Nunc) at 51 °C in a 2 × SSC buffer containing 50 % (v/v) formamide. The three isolates possessed low DNA–DNA relatedness to both *L. monachus* DSM 5896^T (<12 %) and *L. methylaminiphilus* DSM 16812^T (<11 %). The DNA–DNA relatedness between G24103^T and G24116 was >91 %, whereas their relatedness to MAFF 210191^T was <39 % and <46 %, respectively, thus justifying the placement of strains G24103^T and G24116 into one species and strain MAFF 210191^T into a separate species (Wayne *et al.*, 1987). This classification is also supported by findings that the 16S rRNA gene sequences of G24103^T and G24116 are identical and 98.8 % similar to that of MAFF 210191^T. Moreover, isolate MAFF 21091^T has a slightly higher G + C content (62.3 mol%) than G24103^T (61.4 mol%) and G24116 (61.0 mol%). In addition, isolate MAFF 210191^T is able to liquefy gelatin and can use L-glutamine and L-aspartic acid as sole nitrogen sources; all of these properties differentiate it physiologically from the other two isolates.

Therefore, two novel species are proposed with the names *Labrys okinawensis* sp. nov. for isolate MAFF 210191^T and *Labrys miyagiensis* sp. nov. for isolates G24103^T and G24116; these names reflect the respective sites of their first isolation.

The genus *Labrys* was established on the basis of a single isolate (Vasil'eva & Semenov, 1984). A second species of this genus, also based on a single isolate, has been proposed recently (Miller *et al.*, 2005). Here, two further species are proposed on the basis of the study of three isolates. The description of the genus, which was formulated according to the characters of the first isolate, is no longer sufficient to encompass the reported variations in cell shape, motility, G+C content, methylotrophy etc. among its members. Moreover, in the current description of the genus, there is no information regarding its cellular fatty acid profile, which is an important taxonomic marker for the genus *Labrys*. Hence, some emendations of the description of the genus *Labrys* are required; emendations compiling the findings of all five isolates reported so far are proposed.

Similarly, the present description of *L. monachus*, used as a reference species in this study, can be emended, as well as enriched, with the addition of certain significant species-level characteristics such as G+C content, growth inhibition by certain amino acids, cellular fatty acid profile and other features revealed in the course of the present investigations. Therefore, an emended description of *L. monachus* is also proposed.

Emended description of the genus *Labrys*

Characteristics of the genus are as described by Vasil'eva & Semenov (1984), except that cells can be rod-shaped and may or may not possess triangular radial symmetry, may have short prosthecae, can be motile or non-motile and may be facultative methylotrophs; furthermore, the G+C content of the DNA varies from 61.0 to 66.0 mol% and the predominant cellular fatty acids are C_{19:0} cyclo ω 8c, C_{16:0}, C_{18:0} and C_{18:1} ω 7c. The major ubiquinone is Q-10. The genus belongs to the *Alphaproteobacteria*.

Emended description of *Labrys monachus*

The characteristics of the species are as described by Vasil'eva & Semenov (1984). In addition, the species does not grow on NA, LB, TSB or TGY media. Glycine and DL- α -alanine (10 mM) inhibit growth. Citrate is not utilized, vitamins are not required, starch is not hydrolysed, gelatin is not liquefied and 3-ketolactose is not produced from lactose oxidation. Oxidase- and catalase-positive. Tolerates erythromycin (10 μ g ml⁻¹), but is sensitive to ampicillin (50 μ g ml⁻¹), penicillin G (100 μ g ml⁻¹), kanamycin (10 μ g ml⁻¹), chloramphenicol (50 μ g ml⁻¹) and tetracycline (10 μ g ml⁻¹). The major cellular fatty acids are C_{19:0} cyclo ω 8c, C_{16:0}, C_{18:0} and C_{18:1} ω 7c. The major quinone is ubiquinone Q-10. The type strain is DSM 5896^T (= VKM B-1479^T). The DNA G+C content of the type strain is 65.0 mol% (HPLC).

Description of *Labrys okinawensis* sp. nov.

Labrys okinawensis (o.ki.na.wen'sis. N.L. masc. adj. *okinawensis* referring to Okinawa, the province in Japan where the bacterium was first isolated).

Grows slowly and forms white to greyish, semi-translucent, round, raised, convex and smooth colonies on YMA within 2–3 days. Cells are aerobic, Gram-negative, non-sporulating, spherical to short rods. Cells multiply by budding and produce large amounts of extracellular mucilage. Does not grow on nutrient agar (NA), LB, tryptone soya broth (TSB; Oxoid) or TGY media. Uses D-glucose, sucrose, fructose, D-maltose, D-galactose, D-trehalose, L-rhamnose, D-sorbitol, D-xylose, dulcitol, D-arabinose, adonitol, xylitol, *meso*-erythritol, inositol, D-cellobiose and D-mannitol but not D-melezitose or salicin as sole carbon and energy sources. Uses L-threonine, L-methionine, L-proline, L-serine, L-histidine, L-lysine, L-valine and L-tryptophan but not DL- α -alanine, glycine, L-isoleucine, L-glutamic acid, L-cysteine, L-leucine or L-asparagine as sole nitrogen sources. Glycine and DL- α -alanine (10 mM) inhibit growth. Growth is observed at 15–32 °C, but not at 5 or 37 °C. Grows at pH 4.0–9.0. Growth is seen in YMB medium containing up to 0.3 % NaCl. Liquefies gelatin. Does not utilize citrate, require vitamins, hydrolyse starch or produce 3-ketolactose from lactose oxidation. Oxidase- and catalase-positive. Tolerates erythromycin (10 μ g ml⁻¹), but is sensitive to ampicillin (50 μ g ml⁻¹), penicillin G (100 μ g ml⁻¹), kanamycin (10 μ g ml⁻¹), chloramphenicol (50 μ g ml⁻¹) and tetracycline (10 μ g ml⁻¹). The major cellular fatty acids are C_{19:0} cyclo ω 8c, C_{16:0}, C_{18:1} ω 7c and C_{18:0}. The major quinone is ubiquinone Q-10.

The type strain is MAFF 210191^T (= DSM 18385^T), which was isolated from a root nodule of *Entada phaseoloides* on Okinawa, Japan. The DNA G+C content of this strain is 62.3 mol%.

Description of *Labrys miyagiensis* sp. nov.

Labrys miyagiensis (mi.ya.gi.en'sis. N.L. masc. adj. *miyagiensis* referring to Miyagi, the prefecture in Japan where the bacterium was first isolated).

Grows slowly and forms white to greyish, semi-translucent, round, raised, convex and smooth colonies on YMA within 2–3 days. Cells are aerobic, Gram-negative, non-sporulating and spherical to short rods. Cells multiply by budding and produce large amounts of extracellular mucilage. Does not grow on NA, LB, TSB or TGY medium. Uses D-glucose, sucrose, fructose, D-maltose, D-galactose, D-trehalose, L-rhamnose, D-sorbitol, D-xylose, dulcitol, D-arabinose, adonitol, xylitol, *meso*-erythritol, inositol, D-cellobiose and D-mannitol but not D-melezitose or salicin as sole carbon and energy sources. Uses L-threonine, L-methionine, L-proline, L-serine, L-histidine, L-lysine, L-valine and L-tryptophan but not DL- α -alanine, glycine, L-isoleucine, L-glutamic acid, L-glutamine, L-aspartic acid or L-cysteine as sole nitrogen sources. Glycine and DL- α -alanine (10 mM)

inhibit growth. Growth is observed at 15–32 °C, but not at 5 or 37 °C. Grows at pH 4.0–9.0. Growth is seen in YMB medium containing up to 0.3 % NaCl. Does not utilize citrate, require vitamins, hydrolyse starch, liquefy gelatin or produce 3-ketolactose from lactose oxidation. Oxidase- and catalase-positive. Tolerates erythromycin (10 µg ml⁻¹), but is sensitive to ampicillin (50 µg ml⁻¹), penicillin G (100 µg ml⁻¹), kanamycin (10 µg ml⁻¹), chloramphenicol (50 µg ml⁻¹) and tetracycline (10 µg ml⁻¹). The major cellular fatty acids are C_{19:0} cyclo ω 8 ϵ , C_{16:0}, C_{18:1} ω 7 ϵ and C_{18:0}. The major quinone is ubiquinone Q-10.

The type strain is G24103^T (=NBRC 101365^T=NCIMB 14143^T), which was isolated from a grassland soil in Sendai in Miyagi, Japan. Strain G24116 (=NBRC 101366=NCIMB 14144) is a reference strain. The DNA G+C contents of strains G24103^T and G24116 are 61.4 and 61.0 mol%, respectively.

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