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Bacillus songklensis sp. nov., isolated from soil

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A Gram-stain-positive, spore-forming, rod-shaped, motile, strictly aerobic bacterial strain, designated CAU 1033^T, was isolated from soil and its taxonomic position was investigated using a polyphasic approach. Phylogenetic analysis based on 16S rRNA gene sequences revealed that strain CAU 1033^T formed a distinct lineage within the genus *Bacillus* and was most closely related to *Bacillus drentensis* KCTC 13025^T (similarity 95.9 %). CAU 1033^T contained MK-7 as the only isoprenoid quinone and iso-$C_{15:0}$ and anteiso-$C_{15:0}$ as the major fatty acids. The cell wall peptidoglycan of strain CAU 1033^T contained *meso*-diaminopimelic acid and the major whole-cell sugars were arabinose, sucrose and ribose. The polar lipids were composed of diphosphatidylglycerol, phosphatidylglycerol, phosphatidylethanolamine, two unidentified phospholipids, four unidentified aminophospholipids, an unidentified aminolipid, two unidentified glycolipids and another unidentified polar lipid. The DNA G+C content was 41.4 mol%. On the basis of phenotypic data and phylogenetic inference, strain CAU 1033^T was classified as a representative of a novel species in the genus *Bacillus* for which the name *Bacillus songklensis* sp. nov. is proposed. The type strain is CAU 1033^T (=KCTC 13881^T=CCUG 61889^T).

The genus *Bacillus* of the family *Bacillaceae*, belonging to the phylum *Firmicutes*, contains aerobic or facultatively anaerobic bacteria that are able to produce bacterial endospores (Ash *et al.*, 1991). Members have physiologically diverse characteristics and are ubiquitous in nature because they have been isolated from a wide variety of natural environments (Priest *et al.*, 1981; Claus & Berkeley, 1986; Slepecky & Hemphill, 1992). The members of this genus have menaquinone 7 (MK-7) as the major respiratory quinone and iso-$C_{15:0}$ as the predominant cellular fatty acid (Ash *et al.*, 1991; Ahmed *et al.*, 2007a). In the course of the screening of bacteria with biotechnological potential from environmental samples, a bacterial strain, designated CAU 1033^T, was isolated from a non-saline soil sample at a depth of 20–25 cm collected in Wang Sai Thong Waterfall (7° 05′ 27.02″ N 99° 54′ 34.13″ S) in Thailand. The purpose of the present study was to establish the taxonomic position of this bacterial strain by using a polyphasic characterization that included the determination of phenotypic (including chemotaxonomic) properties, a detailed phylogenetic investigation based on 16S rRNA gene sequences and genetic analysis.

Abbreviation: TEM, transmission electron microscopy.

The GenBank/EMBL/DDBJ accession number for the 16S rRNA gene sequence of strain CAU 1033^T is JN036548.

Three supplementary figures are available with the online version of this paper.

Isolation was performed according to the methods of Gordon & Mihm (1962) using glucose-yeast extract agar (GYEA; 10 g yeast extract, 10 g glucose, 15 g agar, l^{-1}; Difco), supplemented with cycloheximide (50 mg l^{-1}) and nalidixic acid (20 mg l^{-1}). The sample was diluted with sterilized 0.9 % sodium chloride solution. Two dilution series were made of the sample; one series was plated directly on GYEA and the other was plated after heating for 15 min at 80 °C to select for endospore-forming bacteria. The agar plates were incubated under aerobic conditions at 30 °C for 14 days. Single colonies on the plates were purified by subculturing. Strain CAU 1033^T was one of the isolates that appeared after direct plating on GYEA. Pure cultures were preserved at −70 °C in glucose-yeast extract broth (GYEB) supplemented with 25 % (v/v) glycerol.

The type strains of eight closely related species were used as reference strains in most analyses. *Bacillus drentensis* KCTC 13025^T, *Bacillus novalis* KCTC 13026^T, *Bacillus soli* KCTC 13572^T, *Bacillus bataviensis* KCTC 13024^T and *Bacillus subtilis* KCTC 1022^T were obtained from the Korean Collection for type Cultures (KCTC; Taejon, Korea). *Bacillus herbersteinensis* DSM 16534^T, *Bacillus methanolicus* DSM 16454^T and *Bacillus marisflavi* KCCM 41588^T were obtained from the Deutsche Sammlung von Mikroorganismen und Zellkulturen (DSMZ; Braunschweig, Germany) and the culture centre of microorganisms (KCCM; Seoul, Korea).

Genomic DNA of strain CAU 1033^T was isolated by the method of Marmur (1961). The 16S rRNA gene was

amplified by PCR following established procedures (Nam *et al.*, 2004). The amplified 16S rRNA gene was sequenced directly using a BigDye Terminator Cycle Sequencing kit (Applied Biosystems) and an automatic DNA sequencer (model 3730; Applied Biosystems). Multiple alignments with sequences of a broad selection of strains of species of the genus *Bacillus* and calculation of sequence similarity levels were carried out by using the EzTaxon-e server (Kim *et al.*, 2012; http://eztaxon-e.ezbiocloud.net/) and CLUSTAL_X (Thompson *et al.*, 1997). Evolutionary distance matrices were generated by the neighbour-joining method described by Jukes & Cantor (1969). Phylogenetic trees were reconstructed using the neighbour-joining (Saitou & Nei, 1987), least-squares (Fitch & Margoliash, 1967) and maximum-likelihood (Felsenstein, 1981) algorithms in the PHYLIP package (Felsenstein, 1989), and tree topology was evaluated by the bootstrap resampling method (Felsenstein, 1985) with 1000 replicates for the neighbour-joining dataset with the SEQBOOT and CONSENSE programs from the PHYLIP package. The nearly complete 16S rRNA gene sequence of strain CAU 1033^T (1529 bp) was determined and compared with the corresponding sequences of other bacterial strains in the GenBank database. Phylogenetic analysis indicated that the strain represented a member of the genus *Bacillus*. The neighbour-joining tree is shown in Fig. 1. The trees obtained with the two other treeing methods used showed essentially the same topology (not shown). Pairwise analysis showed that the most closely related species were *B. drentensis* KCTC 13025^T (similarity 95.9 %), *B. novalis* KCTC 13026^T (similarity 95.8 %), *B. herbersteinensis* DSM 16534^T (similarity 95.8 %), *B. soli* KCTC 13572^T (similarity 95.8 %), *B. marisflavi* KCCM 41588^T (similarity 95.6 %), *B. bataviensis* KCTC 13024^T (similarity 95.6 %) and *B. methanolicus* DSM 16454^T (similarity 95.4 %).

CAU 1033^T was cultivated routinely on GYEA at 30 °C to investigate all morphological, physiological and biochemical characteristics, except for spore formation, which was assessed on nutrient sporulation medium (Nicholson & Setlow, 1990).

Cell morphology was examined by light microscopy (model DM 1000; Leica) and transmission electron microscopy (TEM; JEM 1010; JEOL) using cells from exponentially growing cultures. For TEM, the cells were negatively stained with 1 % (w/v) phosphotungstic acid and the grids were examined after being air-dried. Gram staining was carried out using the bioMérieux Gram staining kit according to the manufacturer's instructions. Motility was assessed using the hanging-drop method. After 7 days of growth, spore formation was determined by staining with malachite green as described previously by Conn *et al.* (1957).

Growth on GYEA at 4, 10, 30, 37, 45 and 55 °C in an aerobic incubator (model MIR-253; Sanyo) and in an anaerobic chamber (Bactron; Sheldon) was evaluated by measuring the turbidity of the broth by spectrophotometry after 7 days. Growth was tested at 30 °C in GYEB adjusted to pH 4.0–10.0 at increments of 0.5 pH units by using sodium acetate/acetic acid and Na_2CO_3 buffers. Growth in the absence of NaCl and in the presence of 1–15.0 % (w/v) NaCl at 0.5 % intervals was investigated at 30 °C in GYEB prepared according to the formula of the Difco medium except that NaCl was excluded and that 0.45 % (w/v) $MgCl_2 . 6H_2O$ and 0.06 % (w/v) KCl was added. Catalase, oxidase and urease activities, nitrate and nitrite reduction, hydrolysis of aesculin, production of indole, methyl red and Voges–Proskauer tests were evaluated as recommended by Smibert & Krieg (1994). Hydrolysis of casein, gelatin, starch, aesculin and citrate were examined as described by Lányi (1987) and Smibert & Krieg (1994). Acid production from carbohydrates, as well as utilization of carbon and energy sources, was analysed as recommended by Ventosa *et al.* (1982). Acid production from carbohydrates, as well as utilization of carbon and energy sources, was tested as recommended by Ventosa *et al.* (1982).

The morphological, cultural, physiological and biochemical characteristics of strain CAU 1033^T are given in Table 1 and in the species description. Overall, the results obtained in this study are in agreement with previously published data for the species of the genus *Bacillus* (Arfman *et al.*, 1992; Yoon *et al.*, 2003; Heyrman *et al.*, 2004, 2005; Wieser *et al.*, 2005). Strain CAU 1033^T was found to consist of Gram-stain-positive, strictly aerobic, motile, rod-shaped cells. Central ellipsoidal endospores were observed in slightly swollen sporangia (Fig. S1, available in IJSEM Online). Colonies grown on GYEA plates for 3 days were cream-coloured, smooth, circular and convex with entire margins. Oxidase and catalase reactions were positive. Despite the fact that CAU 1033^T was isolated from a non-saline environment, it was shown to tolerate NaCl concentrations of up to 4 % (w/v), though it did not require NaCl for growth. Cells are rods approximately 0.6–1.5 µm in diameter and 1.8–4.3 µm in length. The isolate was observed to be motile and TEM demonstrated the presence of peritrichous flagella (Fig. S2). CAU 1033^T did not utilize erythritol, L-arabinose, D-ribose, D-xylose, D-mannose, *N*-acetylglucosamine, amygdalin, arbutin, salicin, cellobiose, raffinose, glycogen, gentiobiose, D-tagatose or potassium 5-ketogluconate as sole carbon sources, did not hydrolyse casein, gelatin, starch and aesculin and did not produce acid from D-glucose, D-mannitol, inositol, D-sorbitol, L-rhamnose or sucrose. However, CAU 1033^T differed from its closest relatives by its inability to hydrolyse aesculin (Table 1).

For fatty acid analysis, the cell mass of strain CAU 1033^T and eight closely related strains were harvested from tryptic soy agar (TSA; Difco) after cultivation for 3 days at 30 °C. The physiological age of the biomasses was standardized by observing growth during incubation of the two cultures and choosing the moment of harvesting according to the standard MIDI protocol (Sherlock Microbial Identification System version 6.1). Cellular fatty acid methyl esters

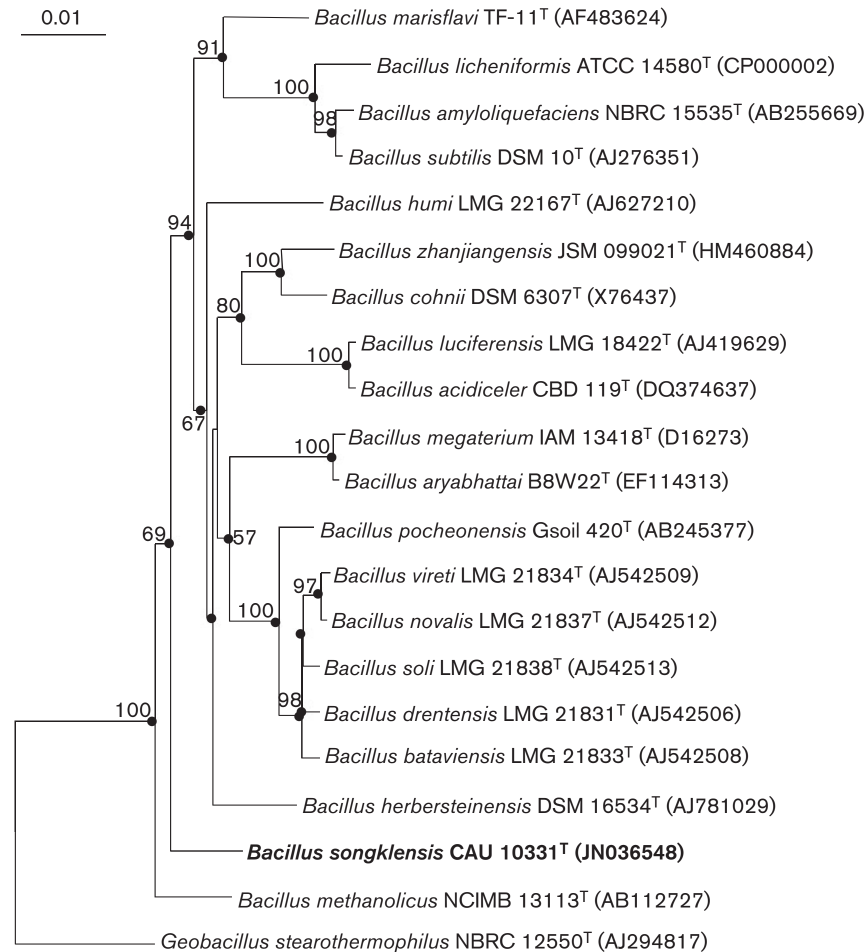

Fig. 1. Neighbour-joining phylogenetic tree based on nearly complete 16S rRNA gene sequences showing the relationships between strain CAU 1033^{T} and the type strains of closely related species of the genus *Bacillus*. Filled circles indicate that the corresponding nodes were also recovered in the trees generated with the maximum-likelihood and least-squares algorithms. Numbers at nodes indicate levels of bootstrap support based on a neighbour-joining analysis of 1000 resampled datasets; only values >70 % are given. Bar, 0.01 substitutions per nucleotide position. *Geobacillus stearothermophilus* NBRC 12550^{T} (AJ294817) is used as an outgroup organism.

(FAMEs) were obtained according to the method of Minnikin *et al.* (1980) and separated by an automated gas chromatography system (model 6890N and 7683 autosampler; Agilent). Peaks were identified by using the Microbial Identification software package (MOORE library v. 5.0; MIDI database TSBA6). The polar lipids of strain CAU 1033^{T} was identified using two-dimensional TLC by the method of Minnikin *et al.* (1984). The plate were sprayed with 10 % ethanolic molybdatophosphoric acid (for the total lipids), molybdenum blue (for phospholipids), ninhydrin (for aminolipids) and α-naphthol/sulphuric acid reagent (for glycolipids) (Sigma-Aldrich). Isoprenoid quinones were separated by HPLC using an isocratic solvent system [methanol/isopropyl ether (3 : 1, v/v)] and a flow rate of 1 ml min^{-1} (Komagata & Suzuki, 1987). Whole-cell sugars were analysed by TLC according to the method of Komagata & Suzuki (1987). Peptidoglycan was analysed as described by Schleifer (1985), with the modification that the cellulose sheet was a substitute for chromatography paper. The G+C content of the genomic DNA was determined using HPLC by the method of Tamaoka & Komagata (1984) with the modification that DNA was hydrolysed and the resultant nucleotides were analysed by reversed-phase HPLC.

The peptidoglycan of strain CAU 1033^{T} contained *meso*-diaminopimelic acid and menaquinone 7 (MK-7) was the only quinone present. These characteristics are in agreement with those of numerous species of the genus *Bacillus*, including the type species, *B. subtilis* (Claus & Berkeley, 1986). However, these characteristics were significantly different from those of any other closely related genus, *Viridibacillus*, (Albert *et al.*, 2007), *Kurthia* (Shaw & Keddie, 1983), *Lysinibacillus* (Ahmed *et al.*, 2007b) or

Table 1. Differential properties of strain CAU 1033^T and the type strains of the most closely related species of the genus *Bacillus*

Strain: 1, CAU 1033^T; 2, *B. methanolicus* DSM 16454^T; 3, *B. drentensis* KCTC 13025^T; 4, *B. herbersteinensis* DSM 16534^T; 5, *B. marisflavi* KCCM 41588^T; 6, *B. novalis* KCTC 13026^T; 7, *B. soli* KCTC 13572^T; 8, *B. bataviensis* KCTC 13024^T; 9, *B. subtilis* KCTC 1022^T. Data were obtained in this study unless indicated. +, Positive; −, negative; w, weakly positive.

Characteristic	1	2	3	4	5	6	7	8	9
Spore morphology	Ellipsoidal/ cylindrical	Ellipsoidal[a]*	Ellipsoidal/ cylindrical[b]	Ellipsoidal[c]	Ellipsoidal[d]	Ellipsoidal[b]	Ellipsoidal[b]	Ellipsoidal[b]	Ellipsoidal[i]
Sporangium shape	Slightly swollen	Slightly swollen	Swollen	Unswollen	Swollen	Slightly swollen	Slightly swollen	Slightly swollen	Unswollen
Spore position	Paracentral/ terminal	Subterminal/ central	Paracentral/ terminal	Terminal	Subterminal/ central	Subterminal/ terminal	Paracentral	Central/ paracentral	Central
Temperature range (°C)	10–45	35–60	30–55	4–28	15–55	30–55	30–45	30–50	10–50
pH range	4.5–10.0	5.0–10.0	5.5–10.0†	7.0–12.0‡	4.5–9.0	4.0–10.0	5.0–9.0	4.0–6.0	4.5–10.0
Catalase	+	+	−	+	+	+	+	−	+
Oxidase	+	+	+	+	−	+	+	+	+
Nitrate reduction	−	+	−	+	−	+	−	+	+
Hydrolysis of:									
Casein	−	−	−	−	+	+	+	−	+
Gelatin	−	+	−	−	+	−	+	−	+
Starch	−	−	−	−	w	+	+	−	+
Citrate	+	−	−	−	+	+	+	+	+
Aesculin	−	+	+	+	+	+	+	+	+
Acid production from:									
D-Glucose	−	−	−	−	−	+	−	+	+
D-Mannitol	−	−	−	+	+	+	+	+	+
Inositol	−	−	−	−	−	+	−	+	+
D-Sorbitol	−	−	−	−	−	+	−	+	+
L-Rhamnose	−	−	−	−	−	−	−	+	+
Sucrose	−	−	−	−	−	−	−	−	+
Utilization of:									
Glycerol	+	−	−	+	+	−	+	+	−
Erythritol	−	−	−	−	−	+	−	−	−
L-Arabinose	−	−	−	−	−	−	+	−	+
D-Ribose	−	+	+	+	−	−	+	+	+
D-Xylose	−	−	−	−	+	+	+	−	+
D-Adonitol	+	−	+	−	+	−	−	−	−
D-Galactose	+	−	+	−	+	−	+	+	−
D-Glucose	+	−	+	+	+	+	+	+	−
D-Fructose	+	+	+	+	+	+	−	+	−
D-Mannose	−	−	−	+	+	+	+	+	−
D-Mannitol	+	+	+	+	+	+	+	+	−
D-Sorbitol	+	−	−	−	−	+	−	−	+
N-Acetylglucosamine	−	−	−	−	−	+	−	+	−
Amygdalin	−	−	−	−	−	+	−	−	+
Arbutin	−	−	+	+	+	+	−	−	+

Table 1. cont.

Characteristic	1	2	3	4	5	6	7	8	9
Salicin	−	−	+	+	+	−	−	+	+
Cellobiose	−	−	+	+	+	−	+	+	+
Maltose	+	+	+	+	+	+	−	+	+
Melibiose	+	−	+	+	+	−	+	+	+
Sucrose	+	−	+	+	+	−	+	+	−
Trehalose	+	−	+	+	+	+	+	+	−
Raffinose	−	−	+	+	+	−	−	−	+
Glycogen	−	−	−	−	−	−	−	−	+
Gentiobiose	−	−	−	−	−	+	−	+	+
D-Tagatose	−	+	−	−	−	−	−	−	−
Potassium 5-ketogluconate	−	+	−	−	−	−	−	−	−
DNA G+C content (mol%)	41.4	48–50[a]	39.4[b]	36.2–36.9[c]	49[d]	40.5[b]	40.1[b]	39.6–40.1[b]	43[e]

*Data from: *a*, Arfman *et al.* (1992); *b*, Heyrman *et al.* (2004); *c*, Wieser *et al.* (2005); *d*, Yoon *et al.* (2003); *e*, Lim *et al.* (2006).

Rummeliibacillus (Vaishampayan *et al.*, 2009). CAU 1033[T] contained branched-chain, saturated and unsaturated fatty acids (Table 2). The major compounds were iso-$C_{15:0}$ (50.1 %) and anteiso-$C_{15:0}$ (25.6 %), which are characteristic of numerous taxa within the bacilli (Kämpfer, 1994). The following fatty acids were present at a level of at least 1 %: summed feature 3 ($C1_{6:1}\omega7c$ and/or iso-$C_{15:0}$ 2-OH), $C_{14:0}$, $C_{16:0}$, iso-$C_{17:0}$, anteiso-$C_{17:0}$, iso-$C_{13:0}$ and iso-$C_{14:0}$. However, some qualitative differences in fatty acid content could be observed between strain CAU 1033[T] and its phylogenetically closest relatives. In particular, strain CAU 1033[T] could be differentiated from closely related species by the presence of iso-$C_{13:0}$ and summed feature 3 and by the absence of iso-$C_{16:0}$. Diphosphatidylglycerol, phosphatidylglycerol and phosphatidylethanolamine were the polar lipids identified in strain CAU 1033[T]. The other, unidentified polar lipids were two phospholipids, four aminophospholipids, an aminolipid, two glycolipids and another unidentified polar lipid (Fig. S3). Strain CAU 1033[T] differs from the type strain of the type species, *B. subtilis* DSM 10[T], by the absence of aminoacylphosphatidylglycerol and the additional occurrence of two unidentified phospholipids, four unidentified aminophospholipids, an unidentified aminolipid and another unidentified polar lipid. On the other hand, the presence of diphosphatidylglycerol, phosphatidylglycerol and diphosphatidylethanolamine in strain CAU 1033[T] has also been reported for *B. subtilis* (Minnikin & Goodfellow, 1981). The polar lipid pattern of strain CAU 1033[T] is very similar to that of the reference strain *B. herbersteinensis* DSM 16534[T] (Wieser *et al.*, 2005). However, phosphatidylethanolamine, four unidentified aminophospholipids and an unidentified aminolipid were absent in *B. herbersteinensis* DSM 16534[T]. TLC analysis of the cell wall sugars of strain CAU 1033[T] revealed the presence of arabinose, sucrose and ribose. The genomic DNA of strain CAU 1033[T] had a G+C content of 41.1 mol%. These data provide sufficient evidence to recognize strain CAU 1033[T] as a novel species of the genus *Bacillus*, for which the name *Bacillus songklensis* sp. nov. is proposed.

Description of *Bacillus songklensis* sp. nov.

Bacillus songklensis (song.klen′sis. N.L. masc. adj. *songklensis* belonging to Songkla, named after Prince of Songkla University, Thailand, where the first sample was collected).

Cells are Gram-stain-positive, motile, strictly aerobic rods approximately 0.6–1.5 µm in diameter and 1.8–4.3 µm in length. Endospores are mainly ellipsoidal and lie in paracentral or terminal positions in slightly swollen sporangia. Colonies on GYEA are cream coloured, circular and convex with entire margins after 3 days of incubation at 30 °C. Growth occurs at 10–45 °C (optimum, 30 °C) and at pH 4.5–10.0 (optimum, 8.0). NaCl is not required for growth but up to 4.0 % (w/v) NaCl is tolerated. Catalase and oxidase are positive. Casein, gelatin, starch, aesculin and urea are not hydrolysed. Nitrate is not reduced. Citrate is hydrolysed. Indole and H_2S are not

Table 2. Cellular fatty acid compositions (percentages) of strain CAU 1033^T and the type strains of the most closely related species of the genus *Bacillus*

Strains: 1, CAU 1033^T; 2, *B. methanolicus* DSM 16454^T; 3, *B. drentensis* KCTC 13025^T; 4, *B. herbersteinensis* DSM 16534^T; 5, *B. marisflavi* KCCM 41588^T; 6, *B. novalis* KCTC 13026^T; 7, *B. soli* KCTC 13572^T; 8, *B. bataviensis* KCTC 13024^T; 9, *B. subtilis* KCTC 1022^T. Data were obtained in this study unless indicated. Only those fatty acids amounting to >1.0 % in all strains are shown, −, Not detected.

Fatty acid	1	2	3	4	5	6	7	8	9
Saturated									
$C_{14:0}$	2.8	−	−	−	−	−	−	−	−
$C_{16:0}$	3.4	1.5	2.1	1.5	2	−	2	1.8	1.8
Unsaturated									
$C_{16:1}\omega11c$	−	1.8	−	1.8	−	−	−	−	−
$C_{16:1}\omega7c$ alcohol	−	5.8	−	5.3	2.1	−	−	2.9	−
Branched-chain									
anteiso-$C_{15:0}$	25.6	15.1	24.3	15.3	28.5	36.5	24.7	23.5	19.2
anteiso-$C_{17:0}$	1.6	6.1	4.9	6.2	9.7	8.9	4.8	5.4	13.5
iso-$C_{13:0}$	1.5	−	−	−	−	−	−	−	−
iso-$C_{14:0}$	1.5	2.9	1.3	2.3	5.8	−	1.3	7.6	1.1
iso-$C_{15:0}$	50.1	44.4	52.5	45.5	38.5	48.6	52.6	46.9	52.1
iso-$C_{16:0}$	−	6.5	3.7	5.8	7.4	3.1	3.8	2.5	2.4
iso-$C_{17:0}$	2.3	3.9	7.7	4	2.4	2.1	7.4	5.6	5.3
iso-$C_{15:1}$ F	−	1.6	−	1.5	−	−	−	−	−
iso-$C_{17:1}\omega10c$	−	4	1.2	4.3	−	−	1.9	2.3	1.5
Summed feature 3*	8.3	−	−	−	−	−	−	−	−
Summed feature 4*	−	4	−	4.5	1.3	−	−	1.4	1.5

*Summed features represent groups of two or three fatty acids that cannot be separated by GLC with the MIDI system. Summed feature 3 contains $C_{16:1}\omega7c$ and/or $C_{15:0}$ iso 2-OH. Summed feature 4 contains $C_{15:1}\omega8c$ and/or $C_{15:2}$.

produced. β-Galactosidase, methyl red, Voges–Proskauer test and urease are negative. Arginine dihydrolase and lysine and ornithine decarboxylase test are negative. Acid production from D-glucose, D-mannitol, inositol, D-sorbitol, L-rhamnose and sucrose are negative. The following carbohydrates were utilized as sole carbon sources: glycerol, D-adonitol, D-galactose, D-glucose, D-fructose, D-mannitol, D-sorbitol, maltose, melibiose, sucrose and trehalose. The following carbohydrates were not utilized as sole carbon sources: erythritol, L-arabinose, D-ribose, D-xylose, D-mannose, *N*-acetylglucosamine, amygdalin, arbutin, salicin, cellobiose, raffinose, glycogen, gentiobiose, D-tagatose and potassium 5-ketogluconate. The cell wall peptidoglycan contains *meso*-diaminopimelic acid. The major isoprenoid quinone is MK-7. The cell wall sugars contain arabinose, sucrose and ribose. The polar lipid pattern consists of diphosphatidylglycerol, phosphatidylglycerol, phosphatidylethanolamine, two unidentified phospholipids, four unidentified aminophospholipids, an unidentified aminolipid, two unidentified glycolipids and an unidentified polar lipid. The major fatty acids were iso-$C_{15:0}$ and anteiso-$C_{15:0}$; summed feature 3, $C_{14:0}$, $C_{16:0}$, iso-$C_{17:0}$, anteiso-$C_{17:0}$, iso-$C_{13:0}$ and iso-$C_{14:0}$ were also present.

The type strain is CAU 1033^T (KCTC 13881^T=CCUG 61889^T), isolated from soil collected from Wang Sai Thong Waterfall in Thailand. The DNA G+C content of the type strain is 41.1 mol%.

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