Ornithinibacillus gen. nov., with the species *Ornithinibacillus bavariensis* sp. nov. and *Ornithinibacillus californiensis* sp. nov.

R. Mayr,¹ H.-J. Busse,² H. L. Worliczek,² M. Ehling-Schulz¹ and S. Scherer¹

Correspondence

S. Scherer

siegfried.scherer@wzw.tum.de

¹Lehrstuhl für Mikrobielle Ökologie, Department für Grundlagen der Biowissenschaften, WZW, Technische Universität München, D-85354 Freising, Germany

²Institut für Bakteriologie, Mykologie und Hygiene, Veterinärmedizinische Universität, A-1210 Wien, Austria

A Gram-positive, aerobic, rod-shaped, motile, endospore-forming bacterium was isolated from pasteurized milk from Bavaria, Germany. 16S rRNA gene sequence similarities indicated that strain WSBC 24001^T was most closely related to *Virgibacillus* species (95.3–96.1 %), *Oceanobacillus* species (95.6–95.7 %), *Bacillus firmus* IAM 12464^T (95.5 %) and *Bacillus niacini* IFO 15566^T (95.2 %). However, strain WSBC 24001^T showed the highest level of sequence similarity to an unnamed strain, MB-9^T (97.6 %), which was isolated from coastal surface sediments in California. Hence, this strain was included in our study. The genomic DNA G + C contents of strains WSBC 24001^T and MB-9^T were 36.4 mol and 40.8 mol%, respectively. The major respiratory quinone of both strains was menaquinone MK-7 and the peptidoglycan type was A4β (L-Orn←D-Asp). The polar lipid profiles of these strains contained a predominance of diphosphatidylglycerol and moderate to minor amounts of phosphatidylglycerol, an unknown phospholipid and an unknown aminophospholipid. However, strain WSBC 24001^T could be distinguished from strain MB-9^T by the presence of an unknown lipid. The fatty acid profiles of the two strains comprised mainly iso- and anteiso-branched acids, but showed some significant quantitative differences in the amounts of certain acids. The DNA–DNA relatedness value (15.5 %) clearly demonstrated that strains WSBC 24001^T and MB-9^T are representatives of two different species. On the basis of their phylogenetic position and morphological, physiological and chemotaxonomic properties, a novel genus is proposed, *Ornithinibacillus* gen. nov., with two novel species, the type species *Ornithinibacillus bavariensis* sp. nov. (type strain WSBC 24001^T = DSM 15681^T = CCM 7096^T) and *Ornithinibacillus californiensis* sp. nov. (type strain MB-9^T = DSM 16628^T = CCM 7237^T).

Aerobic endospore-forming bacteria represent an obligate and well-known part of the microbiota of pasteurized milk, which has been studied extensively over decades. Nevertheless, analysis of the microbiota of commercial high-temperature, short-time (HTST) pasteurized milk from a Bavarian dairy revealed the presence of an endospore-forming bacterium, designated as strain WSBC 24001^T, that could not be identified as a member of a species with a validly published name. 16S rRNA gene sequence analysis revealed a close relationship between strain WSBC 24001^T and strain MB-9^T, which was isolated from coastal surface sediments in California (Francis & Tebo, 2002). The two strains were studied in detail to find a taxonomic home for

these closely related organisms isolated from two such different environments.

A retail sample of Bavarian HTST pasteurized (72 °C, 30 s) milk was plated on plate-count agar containing (l⁻¹) 5.0 g casein peptone (Oxoid), 2.5 g yeast extract (bioMérieux), 1.0 g glucose and 15 g agar, pH 7.0. Strain WSBC 24001^T was isolated from plates incubated aerobically at 30 °C for 72 h. The isolate lost its viability during repeated subcultivation on plate-count agar, but grew well within 3 days at 30 °C on modified trypticase soy agar (TSA) containing (l⁻¹) 17.0 g casein peptone (Oxoid), 3.0 g phytone peptone (Becton Dickinson), 5 g NaCl, 2.5 g K₂HPO₄, 2.5 g glucose, 6 g yeast extract (bioMérieux) and 15 g agar, pH 7.3. Strain MB-9^T was isolated by Francis & Tebo (2002) from surface sediment samples collected at the waterline from the shore of Mission Bay (San Diego, CA, USA) during low tide; sediment samples were diluted in seawater,

Abbreviation: HTST, high-temperature, short-time.

The GenBank/EMBL/DDBJ accession number for the 16S rRNA gene sequence of strain WSBC 24001^T is Y13066.

incubated at 80 °C for 10 min and then plated on Mn(II)-containing K agar (Francis & Tebo, 2002).

16S rRNA gene sequence analysis was performed as described previously (Lechner *et al.*, 1998). Sequence comparisons were done using FASTA3 (Pearson & Lipman, 1988). Strain WSBC 24001^T showed highest levels of 16S rRNA gene sequence similarity (97.6 and 97.3 %, respectively) to strain MB-9^T (GenBank accession no. AF326365) and strain AS-39 (GenBank accession no. AJ391200), a marine bacterium isolated from the Adriatic Sea. For strains WSBC 24001^T and MB-9^T, sequence similarities indicated that the most closely related established taxa with validly published names were species of the genera *Virgibacillus* (94.6–96.1 %) and *Oceanobacillus* (94.7–95.7 %), *Bacillus firmus* IAM 12464^T (95.5 and 93.6 %, respectively) and *Bacillus niacini* IFO 15566^T (95.2 and 94.0 %, respectively). Sequences were aligned and manually edited using CLUSTAL X (Thompson *et al.*, 1997) and BioEdit (Hall, 1999). The phylogenetic position of strain WSBC 24001^T was calculated using software included in the PHYLIP package (Felsenstein, 1993). Sequence distances were calculated on the basis of Kimura's two-parameter model (Kimura, 1980). In all of the phylogenetic trees constructed using neighbour joining (Fig. 1), maximum likelihood and maximum parsimony (not shown), strain WSBC 24001^T was most closely related to the unnamed strain MB-9^T and more distantly to *Oceanobacillus* species.

Whilst strain AS-39 was not accessible, strain MB-9^T was kindly provided by Bradley M. Tebo (Scripps Institute of

Oceanography, San Diego, USA) and thus we could include it in our studies.

Cell morphology and spore formation were examined using a Laborlux S microscope (Leitz). On TSA, the frequency of endospores was low. However, spore formation could usually be observed after 10 days at 30 °C. Motility was tested by spot-inoculation of a plate with trypticase soy soft agar (0.4 % agar agar) supplemented with 3 % NaCl (w/v). After incubation for 3 days at 30 °C, motility was manifested macroscopically as a diffuse zone of growth spreading from the point of inoculation. Swimming motility was observed under the microscope. Morphological characteristics are given in the species description.

Under anaerobic conditions using Anaerocult A (Merck) in an anaerobic jar (Oxoid), no growth was observed for strain WSBC 24001^T or strain MB-9^T after 7 days on TSA at 30 °C. When trypticase soy soft agar (0.4 % agar agar) was prepared in a tube and inoculated by stabbing through the centre of the medium with an inoculation needle, growth was observed only in the upper 1 mm after 7 days at 30 °C. Growth tests were performed in trypticase soy broth (TSB) and TSB supplemented with 3 % NaCl on a rotary shaker at 170 r.p.m. Growth was determined by monitoring the optical density at 585 nm with a portable WinLab photometer LF2400 (Windaus Labortechnik) after 1, 2, 7, 14, 21 and 28 days incubation at various temperatures. Growth was defined as an increase of at least 0.1 optical density units. The growth profile of WSBC 24001^T differed significantly from that of MB-9^T. Generally, WSBC 24001^T

Fig. 1. Distance-matrix tree clustered by neighbour joining based on 16S rRNA gene sequences showing the position of strains WSBC 24001^T and MB-9^T among phylogenetic neighbours of the genera *Bacillus*, *Oceanobacillus*, *Virgibacillus*, *Halobacillus*, *Filobacillus*, *Tenuibacillus* and *Gracilibacillus*. *Paenibacillus polymyxa* DSM 36^T was used as an outgroup. Bootstrap percentages (from 200 replications) greater than 70 % are shown at branching points. Bar, 0.01 substitutions per nucleotide position.

showed better growth than MB-9^T at temperatures $\geq 37^\circ\text{C}$, whereas MB-9^T showed better growth than WSBC 24001^T at temperatures $\leq 30^\circ\text{C}$. The pH range for growth was determined on PYE agar adjusted to pH 5, 6, 7, 8, 9 and 10 by the addition of HCl or NaOH; growth was monitored for 10 days. Growth at various NaCl concentrations was tested on PYE agar (Hauser *et al.*, 2004) supplemented with 0, 0.5, 2, 4, 6, 8, 10, 12, 14 and 16 % NaCl (w/v).

Catalase was tested with 3 % H₂O₂ and oxidase was tested with ready-to-use test strips (Merck). Gram-behaviour was tested by staining cells grown on TSA for 2 and 18 days and by means of the KOH test and L-alanine aminopeptidase activity (Moaledj, 1986). Acid production from carbohydrates was examined over a period of 6 weeks using the API 50 CHB system according to the instructions of the manufacturer (bioMérieux), but covering of the tubes with paraffin oil was necessary to obtain reproducible results.

No clearly reproducible results could be obtained for the two strains with the API 50 CH system and AUX inoculation medium (bioMérieux), but aesculin hydrolysis tested as positive for WSBC 24001^T and MB-9^T. The API 20E system was applied as recommended by the manufacturer (bioMérieux). Biolog GP2 microplates were also used. For strain WSBC 24001^T, the microplates were used according to the manufacturer's instructions without modification; for strain MB-9^T, the medium was adjusted to 3 % (w/v) NaCl. Microplates were incubated for 48 h at 28 °C. Lecithinase activity was tested on TSA supplemented with 3 % NaCl and egg-yolk emulsion (Oxoid) at 50 ml l⁻¹. Formation of dihydroxyacetone was investigated on glycerol/J agar (Claus & Berkeley, 1986) supplemented with 12.0 g casein peptone l⁻¹, 3.0 g phytone peptone l⁻¹ and 30.0 g NaCl l⁻¹, and the ability to hydrolyse starch was tested as described by Claus & Berkeley (1986). Haemolysis was assessed by spot-inoculation on TSA supplemented with 5 % ovine blood (Oxoid) followed by incubation at 37 °C for 1–3 days.

The fatty acid profiles were analysed at the Identification Service of the Deutsche Sammlung von Mikroorganismen und Zellkulturen (DSMZ, Braunschweig, Germany) as described by Kämpfer & Kroppenstedt (1996) and are listed in the species descriptions below. The fatty acid profiles of WSBC 24001^T and MB-9^T contained a predominance of branched acids of the iso and anteiso types (> 80 %). The two strains were distinguishable by the significant differences in the content of C_{16:0}, iso-C_{16:0} and iso-C_{14:0}.

Quinones were extracted and analysed according to Tindall (1990) and Altenburger *et al.* (1996). Strain WSBC 24001^T contained a quinone system that consisted of the major compound MK-7 (98 %) and the minor compound MK-8 (2 %). In strain MB-9^T, the quinone system consisted exclusively of MK-7. These quinone systems are consistent with those found in numerous aerobic, endospore-forming bacteria, including members of the genera *Virgibacillus* (Heyrman *et al.*, 2003) and *Oceanobacillus* (Lu *et al.*, 2001; Yumoto *et al.*, 2005). Polar lipids of WSBC 24001^T, MB-9^T

and *Oceanobacillus iheyensis* DSM 14371^T were analysed according to Tindall (1990). Highly similar polar lipid profiles were detected in WSBC 24001^T (Fig. 2a) and MB-9^T (results not shown). Both strains contained the predominant compound diphosphatidylglycerol, minor amounts of phosphatidylglycerol and an unknown phosphate-containing lipid, PL10. Additionally, minor amounts of an unknown aminophospholipid, APL1, and an unknown

Fig. 2. Two-dimensional thin-layer chromatograms of polar lipids of *Ornithinibacillus bavariensis* WSBC 24001^T (a) and *Oceanobacillus iheyensis* DSM 14371^T (b). Abbreviations: DPG, diphosphatidylglycerol; PG, phosphatidylglycerol; PL1–6 and PL10, unknown phospholipids; APL1, APL2 and APL4, unknown aminophospholipids; GL1, unknown glycolipid; L1–5 and L7, unknown lipids.

lipid, L7, were detected in WSBC 24001^T and trace amounts of an aminolipid were found in MB-9^T. Although *Oceanobacillus iheyensis* DSM 14371^T contained diphosphatidylglycerol and phosphatidylglycerol like the other two strains, it could be readily distinguished by the more complex profile, additionally consisting of six unknown phospholipids, two unknown aminophospholipids and an unknown glycolipid (Fig. 2b). The polar lipid profile also distinguished WSBC 24001^T and MB-9^T from *Virgibacillus* species (Heyrman *et al.*, 2003). Their profiles were less complex and phosphatidylglycerol was detected only in minor amounts, whereas it was reported to be present in moderate to major amounts in *Virgibacillus* species.

Analysis of the cell-wall composition of WSBC 24001^T and MB-9^T was carried out by the DSMZ as described by Schleifer & Kandler (1972), Schleifer (1985), MacKenzie (1987) and Groth *et al.* (1996). Gas chromatographic analysis of derivatized amino acids indicated Ala/Asp/Orn/Glu molar ratios of 2.1:1.2:1.0:1.6 for WSBC 24001^T and 2.7:0.9:1.0:1.9 for MB-9^T (additionally, traces of lysine were detected). On the basis of the presence of Asp→Orn and L-Ala→D-Glu, it was concluded that peptidoglycan type A4β (L-Orn←D-Asp) was present. In contrast, *Oceanobacillus iheyensis* DSM 14371^T and *Virgibacillus picturae* DSM 14867^T, which were also analysed, had the same peptidoglycan type, A1γ (*meso*-diaminopimelic acid direct) (Table 1). The latter type is found in the majority of endospore-forming rod-shaped bacteria; a murein type based on L-ornithine is somewhat unusual for this group of bacteria, though it has been described for members of the genera *Halobacillus* (Amoozegar *et al.*, 2003) and *Filobacillus* (Schlesner *et al.*, 2001), genera that are distantly related to MB-9^T and WSBC 24001^T. Other characteristics that differentiate WSBC 24001^T and MB-9^T from related species are given in Table 1.

Extraction of DNA (Cashion *et al.*, 1977), determination of the G + C content by HPLC (Tamaoka & Komagata, 1984; Mesbah *et al.*, 1989) and spectrophotometric DNA–DNA hybridization (De Ley *et al.*, 1970; Huß *et al.*, 1983; Escara & Hutton, 1980) were carried out by the Identification Service of the DSMZ. Renaturation rates were computed with the TRANSFER.BAS program (Jahnke, 1992). The G + C content of the genomic DNA of WSBC 24001^T and MB-9^T was determined to be 36.4 and 40.8 mol%, respectively. Genomic DNA of strains WSBC 24001^T and MB-9^T revealed DNA–DNA relatedness of 15.5%. This result clearly demonstrates that strains WSBC 24001^T and MB-9^T are representatives of two distinct species.

The phylogenetic analyses of WSBC 24001^T and MB-9^T and 16S rRNA gene sequence similarities indicate that these two strains may be neighbours of the genera *Virgibacillus* and *Oceanobacillus* (Fig. 1). However, there is no statistical support for the branching (bootstrap values of 45 and 20%, respectively). The two strains have the same peptidoglycan type and they have very similar polar lipid profiles, both features that distinguish them from related taxa (Table 1). Their degree of DNA–DNA relatedness demonstrates unambiguously that strains WSBC 24001^T and MB-9^T are members of separate species. These data suggest that they should be classified as separate species in a novel genus, for which we propose the name *Ornithinibacillus*, with *Ornithinibacillus bavariensis* and *Ornithinibacillus californiensis* as the two species.

Description of *Ornithinibacillus* gen. nov.

Ornithinibacillus (Or'ni.thi.ni.ba.cil'lus. N.L. n. *ornithina* ornithine; L. masc. n. *bacillus* a small staff, a wand; N.L. masc. n. *Ornithinibacillus* a rod with ornithine).

Table 1. Characteristics useful for differentiation of strains WSBC 24001^T and MB-9^T (*Ornithinibacillus* gen. nov.) from the genera *Oceanobacillus*, *Virgibacillus* and *Halobacillus*

Symbols: +, present; –, absent; v, variable; ND, not determined; DAP, diaminopimelic acid. Unless indicated otherwise, data for *Virgibacillus* were taken from Heyrman *et al.* (2003) and Yoon *et al.* (2004) and data for *Halobacillus* were taken from Amoozegar *et al.* (2003).

Characteristic	<i>Ornithinibacillus</i>	<i>Oceanobacillus</i>	<i>Virgibacillus</i>	<i>Halobacillus</i>
iso-C _{15:0} /anteiso-C _{15:0} ratio	>1	<1	0.05–1.7	0.2–0.4
Peptidoglycan type	A4β (L-Orn←D-Asp)	A1γ (<i>meso</i> -DAP direct)*	A1γ (<i>meso</i> -DAP direct)*	A4β (L-Orn←D-Asp)
Phosphatidylglycerol content	Minor	Moderate	Moderate to major	Present ^{a†}
Upper limit of NaCl tolerance (% w/v)	10–12	>20	>10–25	>20–30
Growth temperature range (°C)	10–45	15–42 ^b	5–50	10–49
Acid production from:				
D-Mannose	–	+ ^b	v	+
D-Trehalose	+	– ^b	v	ND
DNA G + C content (mol%)	36–41	36 ^b	36–42	40–43

*Determined in this study for *Oceanobacillus iheyensis* and *V. picturae*. *Virgibacillus pantothenticus* contains *meso*-diaminopimelic acid direct (Claus & Berkeley, 1986) and *Virgibacillus halodenitrificans* and *Virgibacillus salexigens* contain *meso*-diaminopimelic acid (Yoon *et al.*, 2004).

†Data taken from: a, Wainø *et al.* (1999); b, Lu *et al.* (2001).

Halotolerant to moderately halophilic. The characteristic diamino acid in the cell wall is ornithine and the peptidoglycan type is A4 β (L-Orn $\leftarrow$ D-Asp). Cellular fatty acids consist mainly of iso- and anteiso-branched acids, with iso-C_{15:0} and anteiso-C_{15:0} predominating and moderate amounts of iso-C_{16:0} and anteiso-C_{17:0}. The predominant compound in the polar lipid profile is diphosphatidylglycerol; moderate to minor amounts of an unknown phospholipid, an unknown aminophospholipid and phosphatidylglycerol are present. Menaquinone MK-7 is the predominant respiratory quinone. The DNA G+C content is 36–41 mol%. The type species is *Ornithinibacillus bavariensis*.

Description of *Ornithinibacillus bavariensis* sp. nov.

Ornithinibacillus bavariensis (ba.va.ri.en'sis. N.L. masc. adj. *bavariensis* of Bavaria, indicating the source of the type strain).

Cells are regular, motile rods, 0.4 μ m wide and 2–6 μ m long, which occur singly or sometimes in short chains. Gram-behaviour of vegetative cells is positive with staining and in the KOH and aminopeptidase tests. Because of the presence of an oval spore in a terminal position, the swollen sporangium (width approx. 0.8 μ m) appears racket-shaped. On TSA, the type strain grows strictly aerobically, forming round, raised, slightly brownish/orange colonies with small regular margins. Within 10 days at 30 °C, the diameter of a single colony reaches about 10 mm. Halotolerant on PYE agar. Growth occurs with NaCl at 0–10% (w/v), with a growth optimum at 0.5–4%. Growth occurs at 15 and 45 °C, but not at 7 °C. Optimal growth is observed at 42 °C. Grows occurs at pH 7–10 on PYE agar but not at pH 6. Catalase-, oxidase- and gelatinase-positive. Results for acid production from carbohydrates (API 50 CHB) within 6 weeks and from Biolog GP2 testing are summarized in Table 2. Tests for β -galactosidase, arginine dihydrolase, lysine decarboxylase, ornithine decarboxylase, L-alanine aminopeptidase, H₂S production, urease, tryptophan deaminase, production of indole, production of acetoin, citrate utilization, nitrate reduction (API 20E), egg-yolk lecithinase, starch hydrolysis and formation of dihydroxyacetone are negative. Haemolysis is observed after incubation for 2 days at 37 °C on blood agar. The peptidoglycan type, quinone system and polar lipid profile are as given in the genus description, but, in addition, an unknown polar lipid is present that cannot be stained with any of the specific spray reagents. The cellular fatty acid profile consists of iso-C_{15:0} (39.0%), anteiso-C_{15:0} (19.9%), C_{16:0} (8.0%), anteiso-C_{17:0} (7.7%), iso-C_{16:0} (6.7%), iso-C_{17:0} (5.7%), iso-C_{14:0} (3.8%), C_{16:1} ω 11c (2.8%), C_{14:0} (1.4%), C_{16:1} ω 7c alcohol (1.3%), C_{15:0} (1.1%), iso-C_{17:1} ω 10c (1.1%), iso-C_{13:0} (1.0%) and summed feature 4 (iso-C_{17:1} and/or anteiso-C_{17:1}) (0.7%). The G+C content of the genomic DNA is 36.4 mol%, as determined by HPLC.

The type strain, WSBC 24001^T (= DSM 15681^T = CCM 7096^T), was isolated from pasteurized milk from Bavaria, Germany.

Table 2. Physiological characteristics of strains WSBC 24001^T and MB-9^T

API 50 CHB results were followed for up to 6 weeks at 30 °C. Biolog GP2 microplates were incubated for 48 h at 28 °C as described in the text. Characteristics are scored as follows: +, positive within 1–2 weeks; W, positive within 4–6 weeks (API 50 CHB) or weak colour development after 72 h (Biolog GP2); ?, plain-coloured to light-orange-coloured test volumes representing unclear results (API 50 CHB); V, not reproducible (Biolog GP2); –, negative. The two strains tested positive for acid production (API 50 CHB) from glycerol, D-glucose, trehalose and 5-ketoglucuronate within 1–2 weeks and tested positive for salicin within 4–6 weeks; both showed positive reactions in the Biolog tests for acetic acid, α -ketovaleric acid and pyruvic acid. The two strains tested negative for acid production (API 50 CHB) from erythritol, D- and L-arabinose, D- and L-xylose, adonitol, methyl β -D-xyloside, galactose, D-mannose, L-sorbose, rhamnose, dulcitol, inositol, sorbitol, methyl α -D-mannoside, methyl α -D-glucoside, amygdalin, arbutin, lactose, melibiose, melezitose, D-raffinose, glycogen, xylitol, β -gentiobiose, D-turanose, D-lyxose, D- and L-fucose, D- and L-arabitol, gluconate and 2-ketogluconate. Both strains were negative in the Biolog reactions for α -cyclodextrin, mannan, N-acetyl-D-glucosamine, N-acetyl- β -D-mannosamine, L-arabinose, D-arabitol, D-cellobiose, L-fucose, D-galactose, D-galacturonic acid, gentiobiose, D-gluconic acid, α -D-glucose, *myo*-inositol, α -D-lactose, lactulose, maltose, maltotriose, D-mannitol, D-mannose, D-melezitose, D-melibiose, methyl α -D-galactoside, methyl β -D-galactoside, 3-methyl D-glucose, methyl α -D-glucoside, methyl β -D-glucoside, methyl α -D-mannoside, palatinose, D-psicose, D-raffinose, L-rhamnose, D-ribose, sedoheptulosan, stachyose, sucrose, D-tagatose, turanose, xylitol, D-xylose, α -hydroxybutyric acid, γ -hydroxybutyric acid, *p*-hydroxyphenylacetic acid, lactamide, D-lactic acid methyl ester, L-lactic acid, D-malic acid, succinamic acid, succinic acid, N-acetyl-L-glutamic acid, L-alaninamide, D-alanine, L-asparagine, putrescine, 2,3-butanediol, adenosine, 2'-deoxyadenosine, inosine, thymidine, adenosine 5'-monophosphate, thymidine 5'-monophosphate, uridine 5'-monophosphate, D-fructose 6-phosphate, α -D-glucose 1-phosphate and D-glucose 6-phosphate. Strain WSBC 24001^T tested negative in the following Biolog reactions: β -cyclodextrin, dextrin, glycogen, inulin, Tween 80, arbutin, salicin, D-trehalose, β -hydroxybutyric acid, α -ketoglutaric acid, propionic acid, L-alanine, L-pyrogutamic acid, L-serine, uridine and DL- α -glycerol phosphate. Strain MB-9^T tested negative for the following Biolog reactions: amygdalin, D-fructose, D-sorbitol, succinic acid monomethyl ester, L-alanyl glycine, L-glutamic acid and glycyl L-glutamic acid.

Characteristic	WSBC 24001 ^T	MB-9 ^T
Growth in 12% NaCl	–	W
Growth at pH 5–6	–	+
Growth on TSA	W	+
Acid formation (API 50 CHB) from:		
Ribose	+	W
D-Fructose	+	?
Mannitol	+	–
Cellobiose	+	–
Maltose	–	W
Sucrose	–	+

Table 2. cont.

Characteristic	WSBC 24001 ^T	MB-9 ^T
Inulin	—	?
Starch	—	W
D-Tagatose	?	W
Biolog reactions		
β-Hydroxybutyric acid	—	+
L-Malic acid	W	V
Pyruvic acid methyl ester	+	V

Description of *Ornithinibacillus californiensis* sp. nov.

Ornithinibacillus californiensis (ca.li.for.ni.en' sis. N.L. masc. adj. *californiensis* of California, indicating the source of the type strain).

Cells are regular, motile rods, 0.4 µm wide and 2–6 µm long. Gram-behaviour of vegetative cells is positive with staining and in the KOH and aminopeptidase tests. Because of the presence of an oval spore in a terminal position, the swollen sporangium (width approx. 0.8 µm) appears racket-shaped. On TSA, the type strain grows strictly aerobically, forming round, raised, slightly brownish/orange colonies with small regular margins. Within 10 days at 30 °C, the diameter of single colonies reaches about 5 mm. Grows at 10 and 37 °C but not at 7 or 42 °C. At 10 °C, growth is visible after an incubation period of 2 weeks. Optimal growth temperature is 30 °C. On PYE agar, growth occurs with NaCl at 0.5–12 % (w/v), with best growth between 0.5 and 8 %. No growth occurs in the absence of NaCl. Good growth is observed at pH 5–9 and weak growth at pH 10. Catalase-, oxidase- and gelatinase-positive. Results for acid production from carbohydrates (API 50 CHB) within 6 weeks and from Biolog testing are summarized in Table 2. Tests for β-galactosidase, arginine dihydrolase, lysine decarboxylase, ornithine decarboxylase, H₂S production, urease, tryptophan deaminase, indole production and acetoin production, citrate utilization, nitrate reduction, starch degradation, egg-yolk lecithinase and formation of dihydroxyacetone are negative. Haemolysis is observed after 3 days incubation on blood agar. The peptidoglycan type, quinone system and polar lipid profile are as given in the genus description. The cellular fatty acid profile consists of iso-C_{15:0} (32.8 %), anteiso-C_{15:0} (23.9 %), iso-C_{16:0} (14.7 %), iso-C_{14:0} (10.2 %), anteiso-C_{17:0} (5.4 %), C_{16:1ω7c} alcohol (4.8 %), C_{16:0} (2.8 %), iso-C_{17:0} (1.9 %), C_{16:1ω11c} (0.9 %), C_{14:0} (0.7 %), summed feature 4 (iso-C_{17:1} and/or anteiso-C_{17:1}) (0.7 %), iso-C_{13:0} (0.6 %) and C_{15:0} (0.6 %). The G+C content of the genomic DNA is 40.8 mol%.

The type strain, MB-9^T (= DSM 16628^T = CCM 7237^T), was isolated by Francis & Tebo (2002) from sediments of Mission Bay, San Diego, CA, USA.

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