

Taxonomic Note

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Falsibacillus pallidus to replace the homonym *Bacillus pallidus* Zhou *et al.* 2008

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The prokaryotic species name *Bacillus pallidus* Zhou *et al.* 2008 is illegitimate because it is a later homonym of the name *Bacillus pallidus* Scholz *et al.* 1988 (*Bacteria*, *Firmicutes*, *Bacillales*, *Bacillaceae*, *Bacillus*). Although *Bacillus pallidus* Scholz *et al.* 1988 was transferred to the genus *Geobacillus* as *Geobacillus pallidus* comb. nov. in 2004, the species name *Bacillus pallidus* Scholz *et al.* 1988 remains validly published and has priority at that position [Principle 8 of the Bacteriological Code (1990 Revision)]. In order to solve this problem, a new name was sought for this taxon, represented by the type strain CW 7^T. In addition, the placement of this species in the genus *Bacillus* was reassessed. The data collected were re-examined as part of a taxonomic study using a polyphasic approach. It was concluded, primarily based on the chemotaxonomic data, that strain CW 7^T did not belong to the genus *Bacillus*, but should be placed in a new genus, for which the name *Falsibacillus* gen. nov. is proposed. The genus currently comprises a single species, *Falsibacillus pallidus* comb. nov. represented by the type strain CW 7^T (=KCTC 13200^T=CCTCC AB 207188^T=LMG 24451^T).

There are a number of important principles on which the Bacteriological Code (Lapage *et al.*, 1992) is based. One of them is the fact that the same strain may bear, depending on differing taxonomic opinions, different names. The name to be used according to a particular taxonomic opinion is determined by establishing whether the name is legitimate, is validly published and has priority at a given rank, circumscription and position. There are two important consequences, namely that an organism may have more than one validly published name and such names remain validly published, even if it is generally accepted that an earlier name has become redundant (see Tindall, 1999). Thus on the transfer of the type strain of *Bacillus pallidus* Scholz *et al.* 1988 (Scholz *et al.*, 1987, 1988) to the genus *Geobacillus* as *Geobacillus pallidus* (Scholz *et al.* 1988) Banat *et al.* 2004 (Banat *et al.*, 2004), the name *Bacillus pallidus* Scholz *et al.* 1988 remains validly published, although according to the taxonomic opinion published by Banat *et al.* (2004) it is currently not the correct name of this taxon. The proposed transfer of this species to the genus *Aeribacillus* Miñana-Galbis *et al.* 2010 as *Aeribacillus pallidus* (Scholz *et al.* 1988) Miñana-Galbis

et al. 2010 in a paper in press (Miñana-Galbis *et al.*, 2010) will simply create the situation whereby there are three synonyms, *Bacillus pallidus* Scholz *et al.* 1988, *Geobacillus pallidus* (Scholz *et al.* 1988) Banat *et al.* 2004 and *Aeribacillus pallidus* (Scholz *et al.* 1988) Miñana-Galbis *et al.* 2010, all of which are validly published. However, only one name may serve as the correct name according to the taxonomic opinion of whether this species belongs in the genus *Bacillus*, *Geobacillus* or *Aeribacillus*.

Zhou *et al.* (2008) published a description of a new taxon based on strain CW 7^T, for which they also proposed the name *Bacillus pallidus*. However, Rule 51b states that:

‘Among the reasons for which a name may be illegitimate are the following.

(4) If it is a later homonym of a name of a taxon of prokaryotes/bacteria, fungi, algae, protozoa, or viruses’.

Rule 51a states:

‘A name contrary to a Rule is illegitimate and may not be used’.

Thus the name *Bacillus pallidus* Zhou *et al.* 2008 is illegitimate because it is a homonym of the name *Bacillus pallidus* Scholz *et al.* 1988 and may not be used. This is also confirmed by Rule 23a, which states:

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'Each taxon above species, up to and including order; with a given circumscription, position, and rank can bear only one correct name, that is, the earliest that is in accordance with the Rules of this Code'.

Principle 8 states:

'Each order or taxon of a lower rank with a given circumscription, position, and rank can bear only one correct name, i.e. the earliest that is in accordance with the Rules of this Code'.

Rule 24a also states:

'Valid publication of names (or epithets) which are in accordance with the Rules of this Code dates from the date of publication of the Code'.

These Principles and Rules emphasize the importance of names that are in accordance with the Code, i.e. legitimate names (Rule 23a, note 5), calling into question whether illegitimate names may be validly published (see Tindall, 2008).

The situation is further complicated by the fact that Rule 12b states:

'No specific or subspecific epithets within the same genus may be the same if based on different types'.

Rule 53 states:

'An epithet is illegitimate if it duplicates a specific or subspecific epithet previously validly published for a species or subspecies of the same genus but which is a different bacterium whose name is based upon another type'.

Rule 51a also states:

'However; a name of a taxon which is illegitimate when the taxon is in one taxonomic position is not necessarily illegitimate when the taxon is in another taxonomic position'.

Rule 41a states:

'When a species is transferred to another genus without any change of rank, the specific epithet must be retained, or if it has not been retained (in a previous publication), it must be re-established...'

In the present case, it is clear that the Code appears to treat both the combination *Bacillus pallidus* as a homonym (a result of which is that it is illegitimate) and the epithet *pallidus* as being illegitimate. It should be noted that the Code is not exact in that it does not require that the name or epithets be specifically linked to the appropriate authority (authors and date), e.g. *Bacillus pallidus* Zhou *et al.* 2008 is illegitimate or the epithet *pallidus* in *Bacillus pallidus* Zhou *et al.* 2008 is illegitimate. The problem of illegitimate names has been summarized by Tindall (2008), with counter arguments published by Young (2009). In the current case, it is clear that the name *Bacillus pallidus* Zhou

et al. 2008 is illegitimate because it is a homonym of *Bacillus pallidus* Scholz *et al.* 1988 and that a new name must be proposed for strain CW 7^T. The Code currently also appears to rule that the epithet *pallidus* in *Bacillus pallidus* Zhou *et al.* 2008 is illegitimate and that it may not be used (at that position), but Rule 51a would also determine that the epithet *pallidus* is legitimate if placed in another genus where there are no such duplications (and attributed to a different authority, i.e. date and authors). However, it would appear that the purpose of Rules 12b and 53 is to avoid homonyms and also the duplication of epithets across the ranks of species and subspecies within the same genus, while homonyms are dealt with by Rule 51a (4) and cross rank duplication by Rule 53. There would appear to be a problem with the wording overlapping and causing unnecessary confusion and over regulation. It is surprising that the name *Bacillus pallidus* Zhou *et al.* 2008 could pass the review and editorial process without the fact that it is clearly a homonym being questioned. There is a clear need to refer back to the Code (Lapage *et al.*, 1992) and the interpretation published previously (Tindall, 1999).

In the original article, Zhou *et al.* (2008) published the description of the new taxon based on the type strain CW 7^T, the major fatty acids of the bacterium were ai-C_{15:0}, i-C_{15:0} and ai-C_{17:0}, the predominant menaquinone was MK-7, the cell-wall peptidoglycan contained meso-diaminopimelic acid and the G + C content of the genomic DNA was 42.3 mol% (Zhou *et al.*, 2008). All of the chemotaxonomic characteristics listed above are fairly common properties of members of the genus *Bacillus*. Analysis of the 16S rRNA gene sequence also showed that the strain CW 7^T was close to the cluster of the genus *Bacillus* and showed gene sequence similarity values of about 96 % with its closest neighbours (96.1 % to *Bacillus flexus* DSM 1320^T, 96.1 % to *Bacillus simplex* DSM 1321^T, 95.5 % to *Bacillus cohnii* DSM 6307^T, but 93.9 % to the type species *Bacillus subtilis* DSM 10^T).

Furthermore, polar lipid profiles have proved to be a most useful tool for the classification and determination of members of the family *Bacillaceae* in recent years. The polar lipid profile of *Bacillus subtilis* (the type species of the genus *Bacillus*) has been shown to be characteristic and should form the basis of part of the description of the genus *Bacillus* (Kämpfer *et al.*, 2006). Based on the importance of polar lipids in members of the family *Bacillaceae*, the phospholipids and glycolipid for strain CW 7^T were reanalysed in this study and stained with different reagents (Minnikin *et al.*, 1984). The polar lipid profile of the type species *Bacillus subtilis* comprises diphosphatidylglycerol, phosphatidylglycerol, phosphatidylethanolamine, β -gentiobiosyldiacylglycerol, an unknown aminophospholipid and an unknown glycolipid (Kämpfer *et al.*, 2006). However, diphosphatidylglycerol, phosphatidylglycerol, phosphatidylethanolamine, two aminophosphoglycolipids and an unknown lipid were identified in the strain CW 7^T by Zhou *et al.*, (2008). In this study, the polar lipids of

strain CW 7^T comprised diphosphatidylglycerol, phosphatidylglycerol, phosphatidylethanolamine, two aminophosphoglycolipids, five unknown phospholipids, two phosphoglycolipids and one glycolipid (Fig. 1). The differences in the polar lipid profiles found in this study and by Zhou *et al.*, (2008) are attributed to the amounts of lipid material applied to the thin layer plates and the spray reagents used. Based on the significant polar lipid differences found in this study between strain CW 7^T and the type species of the genus *Bacillus*, *Bacillus subtilis*, the new isolate described by Zhou *et al.* (2008) should be reassigned to a new genus as a novel species. Some important phenotypic features of strain CW 7^T compared with the most closely related species currently placed in the genus *Bacillus* and the type

species *Bacillus subtilis*, based on 16S rRNA gene sequence similarity, are listed in Table 1 and Table 2.

According to Principle 6 of the Bacteriological Code (1990 Revision) (Lapage *et al.*, 1992), the correct name of a taxon is based upon valid publication, legitimacy and priority of publication. An illegitimate name may not be used, is not taken into consideration with regards determining priority and cannot be a correct name, and must be replaced. Considering the results discussed above, a new genus name is proposed for the taxon previously published as *Bacillus pallidus* Zhou *et al.* 2008, based on the type strain CW 7^T. Based on a consideration of the current Rules of the Code, we consider that only a new genus name is required to avoid homonymy. We therefore propose the new genus name *Falsibacillus* gen. nov.

There is no indication that the taxon names *Falsibacillus* and *Falsibacillus pallidus* have been used previously to designate taxa of prokaryotes, protists, algae, fungi, plants or viruses. The primary resources for searching botanical and zoological names were: Index Nominum Genericorum (ING) Plantarum (<http://botany.si.edu/ing/>), Names in

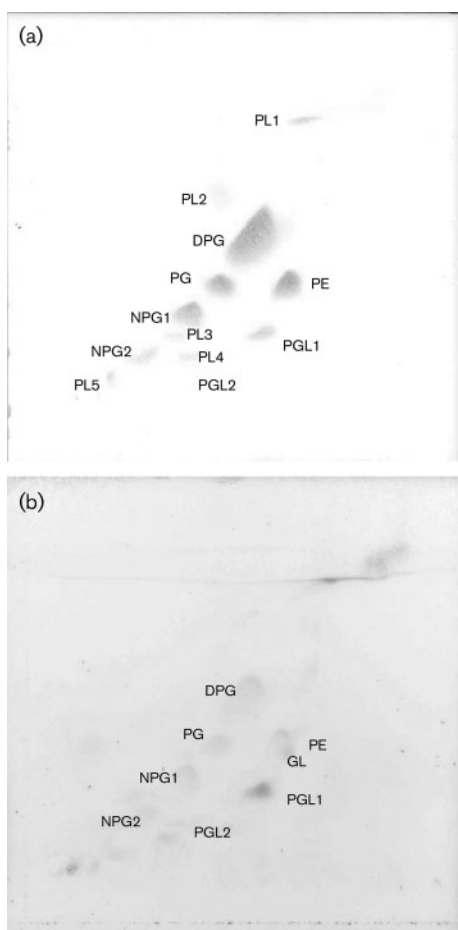


Fig. 1. Two-dimensional TLC of a polar lipid extract from strain CW 7^T. (a) first stained with ninhydrin and restained using molybdenum blue; (b) stained with α -naphthol. The staining of DPG, PG and PE with α -naphthol is a side reaction caused by overheating of the plates, which causes the fatty acids in these spots to char. DPG, diphosphatidylglycerol; PG, phosphatidylglycerol; PE, phosphatidylethanolamine; NPG1 and NPG2, ninhydrin-positive phosphoglycolipids; PGL1 and PGL2, phosphoglycolipid; PL1–PL5, unknown phospholipids, GL, glycolipid.

Table 1. Phenotypic characteristics that differentiate strain CW 7^T from its closest phylogenetic neighbours and the type species of the genus *Bacillus*, *Bacillus subtilis*

Taxa: 1, strain CW 7^T; 2, *B. flexus* DSM 1320^T; 3, *B. simplex* DSM 1321^T; 4, *B. cohnii* DSM 6307^T; 5, *B. subtilis* DSM 10^T. +, Positive; (+), weakly positive; –, negative; v, variable; NA, not available. The colony colour of *B. subtilis* was determined in the present study; all other data were obtained from Zhou *et al.* (2008).

Characteristic	1	2	3	4	5
Gram staining	+	NA	v	NA	+
Catalase	(+)	NA	+	NA	+
Oxidase	–	NA	–	NA	+
Colony colour	Light pink	Cream	Cream	White	Pale yellow
Optimal pH for growth	7.0–8.0	7.0	NA	9.0	NA
Growth in NaCl (w/v):					
5 %	–	+	v	+	+
8 %	–	+	–	–	v
Hydrolysis of:					
Aesculin	+	–	v	–	+
Starch	–	+	+		+
Acid production from:					
D-Fructose	–	+	+	NA	+
D-Glucose	–	+	+	–	+
Glycerol	–	+	+	NA	+
Maltose	–	+	+	–	+
D-Mannitol	–	+	+	–	+
Raffinose	–	+	–	–	+
Sucrose	–	+	+	–	+
Trehalose	–	+	+	NA	+
D-Xylose	+	–	–	–	v

Table 2. Cellular fatty acid composition of strain CW 7^T, its closest phylogenetic neighbours and the type species of the genus *Bacillus*, *Bacillus subtilis*

Taxa: 1, strain CW 7^T; 2, *B. flexus* DSM 1320^T; 3, *B. simplex* DSM 1321^T; 4, *B. cohnii* DSM 6307^T; 5, *B. subtilis* DSM 10^T. Values are percentages of total fatty acids. Fatty acids representing <1.0% in all species are omitted. Means ± SD are given where more than one test was performed. ND, Not detected; i, iso; ai, anteiso. Data for cellular fatty acid contents are from Zhou et al. (2008).

Fatty acid	1	2	3	4	5
i-C _{14:0}	1.2 ± 0.3	1.6	10.8 ± 4.3	0.8	2.3
C _{14:0}	3.4 ± 0.8	ND	2.8 ± 0.2	ND	1.1
i-C _{15:0}	11.5 ± 1.3	32.2	13.5 ± 1.2	22.9	13.2
ai-C _{15:0}	36.6 ± 2.8	35.4	48.5 ± 4.1	20.3	40.0
i-C _{16:0}	5.6 ± 1.1	8.6	4.9 ± 1.4	7.0	4.6
C _{16:0}	7.7 ± 0.9	1.2	5.4 ± 0.6	1.0	10.2
C _{16:1} ω7c alcohol	ND	ND	3.3 ± 0.6	ND	ND
C _{16:1} ω11c	ND	ND	6.6 ± 0.7	ND	4.8
i-C _{17:0}	1.5 ± 0.1	2.1	1.0 ± 0.2	35.3	7.8
i-C _{17:1} ω10c	ND	ND	ND	ND	1.6
ai-C _{17:0}	19.0 ± 3.1	3.4	1.9 ± 0.5	2.2	12.2
C _{18:0}	4.1 ± 1.0	ND	ND	ND	1.0
C _{18:1} ω9c	3.4 ± 0.6	ND	ND	ND	ND
Summed features*					
4	ND	ND	ND	ND	1.2
8	1.8 ± 0.2	ND	ND	ND	ND

*Summed features represent groups of two or three fatty acids that cannot be separated by GC with the MIDI system. Summed feature 4 contains i-C_{17:1} I and/or ai-C_{17:1} B; summed feature 8 contains C_{18:1}ω7c and/or C_{18:1}ω6c.

Current Use for Extant Plant Genera (NCU) (<http://www.bgbm.fu-berlin.de/iapt/ncu/genera/Default.htm>), algae BASE (<http://www.algaebase.org/>), Index of Organism Names (Thomson) (<http://www.organismnames.com>), Index Fungorum (<http://www.indexfungorum.org/Names/Names.asp>) and uBio (www.ubio.org). The date of the last search was 9 September 2009.

In proposing the name *Falsibacillus pallidus*, we have taken the following into consideration: While the name *Bacillus pallidus* Zhou et al. 2008 is a later homonym of *Bacillus pallidus* Scholz et al. 1988 and the epithet *pallidus* in *Bacillus pallidus* Zhou et al. 2008 is also illegitimate, using the epithet *pallidus* in another combination, *Falsibacillus pallidus* does not contravene the Code. In fact, Rule 41a states that one must use the earliest validly published and legitimate epithet. It is unclear, however, how this is applied to an epithet that is illegitimate in the position in which it was originally published, but legitimate in another. The question of whether the combination *Bacillus pallidus* Zhou et al. 2008 is validly published is also another issue and whether the name *Falsibacillus pallidus* should make reference to that name as its basonym. While it is acceptable to cite the paper by Zhou et al. (2008) as the

source of some of the data, the citation of the name *Falsibacillus pallidus* may either be as a new species name (sp. nov.), a new combination (comb. nov.) or as a new name (nom. nov.), all of which would affect the way authorship is cited; see also Rule 54. In fact the whole issue of the way illegitimate names are handled needs to be examined carefully in order to avoid potential clashes in the interpretation of the Code (Tindall, 2008; Young, 2009). We have further noted that there are differences in the way the data are presented for the species description of *Bacillus pallidus* Zhou et al. 2008 (Zhou et al., 2008) and the way the data is presented here for the description of the genus *Falsibacillus* and the species *Falsibacillus pallidus*. We do not consider it appropriate to refer to the description of Zhou et al. (2008) in the species description of *Falsibacillus pallidus*, although the paper is relevant as the original source of the data and methods. It is also unclear whether the name *Falsibacillus pallidus* should be treated as a new species name (sp. nov.) and new name (nom. nov.) or a new combination (comb. nov.). In this paper, it is temporarily described as a new combination.

Description of *Falsibacillus* gen. nov.

Falsibacillus (Fal.si.ba.cil'lus. L. adj. *falsus* false; N.L. masc. n. *Bacillus* a bacterial generic name; N.L. masc. n. *Falsibacillus* false *Bacillus*).

Gram-positive, spore-forming, aerobic and rod-shaped cells (0.7–1.0 × 1.8–3.5 μm), motile by means of peritrichous flagella. Spores are ellipsoidal and subterminal. Mesophilic and neutrophilic growth requirements. Colonies are circular, flat and light pink after 2 days cultivation at 37 °C on TYB medium. The fatty acids comprise i-C_{14:0}, C_{14:0}, i-C_{15:0}, ai-C_{15:0}, i-C_{16:0}, C_{16:0}, i-C_{17:0}, ai-C_{17:0}, C_{18:0}, C_{18:1}ω9c, summed feature 8 (C_{18:1}ω7c and/or C_{18:1}ω6c) (Table 2). The predominant menaquinone is MK-7 and the cell-wall peptidoglycan contains meso-diaminopimelic acid. The polar lipid profile consists of diphosphatidylglycerol, phosphatidylglycerol, phosphatidylethanolamine, two aminophosphoglycolipids, five unknown phospholipids, two phosphoglycolipids and one glycolipid (Fig. 1). The G + C content of the genomic DNA is 42.3 mol% (type strain CW 7^T, determined by HPLC). The type and currently only species is *Falsibacillus pallidus*.

Description of *Falsibacillus pallidus* comb. nov.

Falsibacillus pallidus (pal.li'dus. L. masc. adj. *pallidus* pale or paleish, the light pink colour of colonies).

This name may be linked to the illegitimate name: *Bacillus pallidus* Zhou et al. 2008.

Cellular morphology, oxygen relationships, colony shape and pigmentation are as given in the genus description above. Growth occurs at 15–42 °C, with an optimum at 30–37 °C. The pH range for growth is pH 6.0–8.5, with an

optimum at pH 7.0–8.0. Growth in the absence of NaCl, but no growth in the presence of 3% (w/v) NaCl. Casein and gelatin are hydrolysed, but Tween 80, chitin, starch, DNA, pectin and tyrosine are not hydrolysed. Weak catalase activity is present. Oxidase, β -galactosidase, arginine dihydrolase, lipase, methyl α -D-glucosidase, ornithine decarboxylase, lysine decarboxylase and urease activities are absent. No reduction of nitrate and nitrite. H₂S is not produced in the triple-sugar iron test. Voges–Proskauer test is positive, indole is not produced. Utilizes aesculin, gluconate, 2-ketogluconate and D-xylose, but not N-acetylglucosamine, D-adonitol, D-arabinose, L-arabinose, D-arabitol, L-arabitol, arbutin, cellobiose, dulcitol, erythritol, D-fructose, D-fucose, L-fucose, D-galactose, D-gentiobiose, D-glucose, glycerol, glycogen, inositol, inulin, 5-ketogluconate, D-lactose, amygdalin, D-lyxose, maltose, mannitol, D-mannose, melezitose, melibiose, methyl α -D-glucoside, methyl α -D-mannoside, methyl β -D-xyloside, raffinose, L-rhamnose, D-ribose, D-salicin, sorbitol, L-sorbitose, starch, sucrose, D-tagatose, trehalose, turanose, xylitol or L-xylose (API CHB tests). The polar lipid, fatty acid, respiratory lipoquinone and G+C content of the DNA are as given in the genus description.

The type strain, CW 7^T (=KCTC 13200^T=CCTCC AB 207188^T=LMG 24451^T), was isolated from a forest soil in Anhui Province, China.

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