Acidicapsa dinghuensis sp. nov., a novel acidobacterium isolated from forest soil

Tian-na Ou-yang,† Fan Xia† and Li-hong Qiu*

Abstract

A bacterial strain, designated 4GSKX<sup>T</sup>, isolated from the forest soil of Dinghushan Biosphere Reserve, Guangdong Province, PR China (112° 31’ E 23° 10’ N), is proposed as a novel species of the genus Acidicapsa. Cells of strain 4GSKX<sup>T</sup> were aerobic, non-motile, Gram-stain-negative short rods that multiplied by binary division. The strain grew at 12–37 °C (optimum, 25–30 °C), pH 4.0–6.5 (optimum, pH 4.5–5.0) and NaCl concentrations of 0–1.0 % (w/v; optimum, 0 %). Strain 4GSKX<sup>T</sup> utilized various carbon sources as growth substrates, including both sugars and amino acids. The major fatty acids (>10 %) were iso-C<sub>15</sub>:0 (48.8 %) and iso-C<sub>17:0</sub>ω9c/C<sub>16</sub>:0 10-methyl (14.7 %). The major polar lipids were phosphatidylethanolamine, an unidentified glycolipid, three unidentified phospholipids and two unidentified aminophospholipids. The only quinone detected was MK-8 and the DNA G+C content was 52.8 mol%. Phylogenetic analysis based on 16S rRNA gene sequences showed that strain 4GSKX<sup>T</sup> belongs to the genus Acidicapsa in the family Acidobacteriaceae in subdivision 1 of the phylum Acidobacteria, with the highest similarity of 97.1 % to Acidicapsa ligni WH120<sup>T</sup>. Based on all phenotypic, chemotaxonomic and phylogenetic data obtained, it is proposed as a novel species of genus Acidicapsa, for which the name Acidicapsa dinghuensis sp. nov. is proposed, with 4GSKX<sup>T</sup> (=CGMCC 1.15449<sup>T</sup>=LMG 29213<sup>T</sup>) as the type strain.

The phylum Acidobacteria is one of the most dominant bacterial groups in many habitats, especially soil environments, based on 16S rRNA gene sequence analysis [1]. It currently contains 26 subdivisions but there are only 57 species with validly published names, with most of them belonging to subdivision 1 [2–4]. The genus Acidicapsa was established by Kulichevskaia et al. [5], which is one of the 12 genera in the family Acidobacteriaceae. Currently, there are five published Acidicapsa species: Acidicapsa borealis KA1<sup>T</sup> [5], Acidicapsa ligni WH120<sup>T</sup> [5], Acidicapsa acidisolii SK-11<sup>T</sup> [6], Acidicapsa ferriducens MCF9<sup>T</sup> [7] and Acidicapsa acidiphila MCF10<sup>T</sup> [7]. Members of the genus Acidicapsa are acidophilic and mesophilic, Gram-stain-negative, non-spore-forming rods that reproduce by binary fission, and occur singly, in pairs or in short chains. DNA G+C content ranges from 51.7 to 56.9 %, with MK-8 as the only quinone and iso-C<sub>15</sub>:0 as the dominant cellular fatty acid (>45 %).

Dinghushan Biosphere Reserve (DHSBR) lies in the middle part of Guangdong Province, China (112° 31’ E 23° 10’ N). A research based on the analysis of 16S rRNA gene clone library indicated that Acidobacteria was one of the dominant phyla in the soil bacterial communities of DHSBR [8]. Recently, an acidophilic and mesophilic strain, 4GSKX<sup>T</sup>, was isolated from the forest soils of DHSBR. The 16S rRNA gene analysis showed that 4GSKX<sup>T</sup> has the highest 16S rRNA gene sequence similarities to members of the genus Acidicapsa, ranging from 95.4–98.7 %. The purpose of this study is to elucidate the taxonomic position of strain 4GSKX<sup>T</sup> using a polyphasic approach.

To isolate strain 4GSKX<sup>T</sup>, the soil sample collected from the upper layer (0–25 cm) of forest soil from DHSBR was suspended in 100 mM phosphate buffered saline, 1/10 diluted three times, and then innoculated on MM1F agar medium (0.04 g MgSO<sub>4</sub>·7H<sub>2</sub>O, 0.02 g CaCl<sub>2</sub>·2H<sub>2</sub>O, 0.05 g yeast extract, 0.5 g fructose in 1 litre of distilled water, pH 5.0) which was equivalent to the medium MM1 [9] supplemented with fructose. A single colony, obtained after incubation at 28 °C for 2 weeks and purified by subculturing three times, was designated as 4GSKX<sup>T</sup> and then stored in 25 % (v/v) glycerol in a freezer at −80 °C. The reference strain, A. ligni WH120<sup>T</sup> (purchased from DSMZ) used in this study was cultured on the same medium under the same conditions.

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Keywords: Acidicapsa; acidophilic; phylogeny.
Abbreviations: CGMCC, China General Microbial Culture Collection; DHSBR, Dinghushan Biosphere Reserve; DSMZ, Deutsche Sammlung von Mikroorganismen und Zellkulturen; LMG, Laboratorium voor Microbiologie, Universiteit Gent (UGent); MP, maximum-parsimony; ML, maximum-likelihood; PDE, phosphatidylethanolamine; PE, phosphatidylethanolamine; NJ, neighbour-joining.
†These authors contributed equally to this work.
The 16S rRNA gene sequence of strain 4GSKX<sup>T</sup> has been deposited in DDBJ/EMBL/GenBank under accession number MF150300.

Two supplementary figures are available with the online version of this article.
Cell morphology of strain 4GSKX was observed under a Nikon light microscope at ×1000 magnification and a transmission electron microscope (JEM-1400) after cultivation on MM1F agar medium at 28°C for 2 weeks. An ultrathin section was operated as described by Pankratov et al. Growth of strain 4GSKX under various growth conditions, including pH ranging between 3.0 and 8.0 at 0.5 interval (buffered with 0.2 M Na₂HPO₄+0.1 M citric acid for pH ≤5.5 and 0.2 M KH₂PO₄+0.2 M NaOH for pH ≥6.0), temperatures at 4, 10, 20, 25, 28, 33, 37 and 42°C, and NaCl concentrations of 0–3.0% (w/v, at 0.5% intervals) [10], was monitored by measuring OD₆₀₀ of the cultures in MM1F liquid medium at 28°C for 2 weeks (4 and 6 weeks for temperatures of 10 and 4°C, respectively). Gram staining was determined as proposed by Doetsch [11]. Motility and anaerobic growth were tested by observing the growth of the bacteria inoculated by piercing into MM1F and hydrogen sulfide indole motility media (20.0 g tryptone, 6.0 g polyvalent peptone, 0.2 g NH₄Fe(SO₄)₂, 0.2 g Na₂S₂O₃, 3.5 g agar in 11 of distilled water) in test tubes. The flagellum staining was performed using both ammonium oxalate crystal violet and black ink. The catalase and oxidase activities were determined using the corresponding test with 3% (v/v)

### Table 1. Differential phenotypic characteristics of strain 4GSKX and its closely related neighbours

All data were obtained from this study except the DNA G+C content, motility, pH optimum and NaCl tolerance which were from the original studies. +, Positive; w, weakly positive; −, negative; n.d, not determined. Strain 4GSKX, A. ligni WH120, A. borealis KA1 and A. acidisoli SK11 test positive for following enzymic activities: alkaline phosphatase, α-fucosidase, β-galactosidase, β-glucosidase, β-glucuronidase, cystine arylamidase, esterase (C4), leucine arylamidase, naphthol-AS-BI-phosphohydrolase and valine arylamidase, while none of them appears positive for lipase (C14).

<table>
<thead>
<tr>
<th>Characteristic</th>
<th>4GSKX</th>
<th>A. ligni WH120</th>
<th>A. borealis KA1</th>
<th>A. acidisoli SK11</th>
</tr>
</thead>
<tbody>
<tr>
<td>Origin</td>
<td>Forest soil</td>
<td>Decaying wood</td>
<td>Sphagnum peat</td>
<td>Forest soil</td>
</tr>
<tr>
<td>Cell size (µm)</td>
<td>0.4–0.8×0.5–1.5</td>
<td>0.5–0.8×1.0–2.0</td>
<td>0.6–0.9×1.0–3.0</td>
<td>0.7–1.0×1.0–1.4</td>
</tr>
<tr>
<td>Colour of colony</td>
<td>Beige</td>
<td>Colourless</td>
<td>Pale pink</td>
<td>White</td>
</tr>
<tr>
<td>Motility</td>
<td>−</td>
<td>−</td>
<td>−</td>
<td>−</td>
</tr>
<tr>
<td>pH for growth:</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Range</td>
<td>4.0–6.5</td>
<td>3.5–6.4</td>
<td>3.5–7.3</td>
<td>4.0–5.5</td>
</tr>
<tr>
<td>Optimum</td>
<td>4.5–5.0</td>
<td>4.0–4.5</td>
<td>5.0–5.5</td>
<td>5.0</td>
</tr>
<tr>
<td>Temperature for growth (°C):</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Range</td>
<td>12–37</td>
<td>10–33</td>
<td>10–33</td>
<td>10–35</td>
</tr>
<tr>
<td>NaCl tolerance (w/v, %):</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Range</td>
<td>0–1.0</td>
<td>0–2.0</td>
<td>0–2.0</td>
<td>0–0.4</td>
</tr>
<tr>
<td>Optimum</td>
<td>0</td>
<td>0–1.0</td>
<td>0–1.0</td>
<td>0–0.1</td>
</tr>
<tr>
<td>DNA G+C content (mol%)</td>
<td>52.8</td>
<td>51.7</td>
<td>54.1</td>
<td>56.9</td>
</tr>
<tr>
<td>Menaquinone</td>
<td>MK-8</td>
<td>MK-8</td>
<td>MK-8</td>
<td>MK-8</td>
</tr>
<tr>
<td>Catalase/oxidase activity</td>
<td>−/−</td>
<td>+/−</td>
<td>+/−</td>
<td>+/+</td>
</tr>
<tr>
<td>Carbon source utilization:</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Arbutin</td>
<td>−</td>
<td>−</td>
<td>+</td>
<td>+</td>
</tr>
<tr>
<td>D- Arabinose</td>
<td>−</td>
<td>−</td>
<td>+</td>
<td>w</td>
</tr>
<tr>
<td>D- Fructose</td>
<td>+</td>
<td>−</td>
<td>+</td>
<td>+</td>
</tr>
<tr>
<td>D- Fucose</td>
<td>−</td>
<td>−</td>
<td>−</td>
<td>w</td>
</tr>
<tr>
<td>D- Galactose</td>
<td>+</td>
<td>−</td>
<td>+</td>
<td>−</td>
</tr>
<tr>
<td>D- Glucose</td>
<td>+</td>
<td>−</td>
<td>+</td>
<td>+</td>
</tr>
<tr>
<td>D- Mannose</td>
<td>+</td>
<td>−</td>
<td>+</td>
<td>+</td>
</tr>
<tr>
<td>D- Ribose</td>
<td>−</td>
<td>−</td>
<td>+</td>
<td>−</td>
</tr>
<tr>
<td>D- Xylose</td>
<td>+</td>
<td>−</td>
<td>+</td>
<td>+</td>
</tr>
<tr>
<td>Starch</td>
<td>−</td>
<td>−</td>
<td>+</td>
<td>+</td>
</tr>
<tr>
<td>Enzymic activities:</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Acid phosphatase</td>
<td>+</td>
<td>w</td>
<td>+</td>
<td>+</td>
</tr>
<tr>
<td>α-Chymotrypsin</td>
<td>−</td>
<td>−</td>
<td>+</td>
<td>−</td>
</tr>
<tr>
<td>α-Galactosidase</td>
<td>+</td>
<td>+</td>
<td>+</td>
<td>w</td>
</tr>
<tr>
<td>α-Glucosidase</td>
<td>+</td>
<td>+</td>
<td>+</td>
<td>−</td>
</tr>
<tr>
<td>α-Mannosidase</td>
<td>+</td>
<td>+</td>
<td>+</td>
<td>−</td>
</tr>
<tr>
<td>C8 Esterase lipase</td>
<td>−</td>
<td>w</td>
<td>+</td>
<td>+</td>
</tr>
<tr>
<td>N- Acetyl-β-glucosaminidase</td>
<td>−</td>
<td>−</td>
<td>+</td>
<td>+</td>
</tr>
</tbody>
</table>
H₂O₂ and reaction with 1 % (w/v) tetramethyl-p-phenylene-
mediamine, respectively.

Enzymic activity profiles, including urease hydrolysis, the
indole production test, the Hugh–Leifson test, etc, and the
carbon source utilizations of strain 4GSKXᵀ in the refer-
ence strain were examined using API ZYM, API 20NE and
API 50 CHB/E galleries (bioMérieux) according to the man-
ufacturer’s instructions. Susceptibility to antibiotics was
determined on MM1F agar medium inoculated with tablets
(Hangzhou Microbial Reagent Co.) containing following
antibiotics: tobramycin (10 µg), penicillin (10 IU), tetracy-
cline (30 µg), neomycin (30 µg), vancomycin (30 µg), genta-
micin (10 µg), chloramphenicol (30 µg), erythromycin
(15 µg), ciprofloxacin (5 µg), kanamycin (30 µg), streptomycin
(10 µg), amikacin (30 µg), nalidixic acid (30 µg) and poly-
myxin (300 U).

Cell biomass for chemotaxonomic analysis was obtained
from batch cultures grown on MM1F medium at 28 °C for
2 weeks. For fatty acid analysis, 40 mg bacterial cells were
harvested and the fatty acid methyl esters were determined
using the method proposed by Kuykendall et al. [12]. Fatty
acid methyl esters were quantitated using the Sherlock
Microbial Identification System (MIDI) according to the man-
ufacturer’s instructions [13]. Respiratory quinones were
extracted according to Collins [14] and separated by high-
performance liquid chromatography using the procedure
described by Tamaoka et al. [15]. Polar lipids were extracted
and analysed by two dimensional thin-layer chromatogra-
phy according to Minnikin et al. [16].

For the 16S rRNA gene sequence analysis, genomic DNA
of strain 4GSKXᵀ was extracted using a commercial bac-
teria genomic DNA-extraction kit (Omega Bio-Tek) and
the 16S rRNA gene was amplified by PCR using the uni-
versal primers 27F and 1492R [17]. The amplified gene
fragment was cloned into pMD18-T vector (TaKaRa) using
the TA cloning method. 16S rRNA gene sequencing was
performed by Sanger sequencing using the 3730XL DNA
analysers (Applied Biosystems). Applying the EZBioCloud
server, the closest phylogenetic neighbours were identified
[18]. The nearly complete sequence of the 16S rRNA gene
was compiled using MEGA5 program [19] and then sub-
mitted to GenBank. Multiple alignment of sequence data was
achieved using CLUSTAL_X 2.1 [20] and the gaps at the 5’
and 3’ ends were deleted using the software package BioE-
dit [21]. Neighbour-joining (NJ) [22], maximum-parsi-
moniy (MP) [23] and maximum-likelihood (ML) [24]
phylogenetic trees with 1000 bootstrap replications [25]
were reconstructed and evolutionary distances were deter-
mined using Kimura’s two-parameter model [26], which
was selected using the MEGA5 program. The similarity
and pairwise distance values were calculated using MegAlign
software (DNASTAR). The genomic G+C-content was deter-
mined by using reverse-phase high-performance liquid
chromatography using a protocol described by Mesbah
et al. [27].

The colonies of 4GSKXᵀ appeared semi-transparent, beige,
mucous, raised and edge-smooth on an MM1F agar plate after
2 weeks of cultivation. Cells of 4GSKXᵀ were Gram-stain-
negative, non-spore-forming, non-capsule-forming and non-
motile short rods (0.4–0.8×0.5–1.5 µm) that reproduced by
binary fission (Fig. S1). The pH range for growth of strain
4GSKXᵀ on MM1F agar medium was 4.0–6.5, with optimum
at 4.5–5.0. It grew at 12–37 °C with optimum at
25–30 °C and NaCl concentrations no higher than 1.0 % (w/
v). Capsules were not observed for any of them staining with
either safranine or black ink. The enzymic activities, as well as
the abilities to utilize various substrates as sole carbon source
for growth, are provided in detail in species descriptions. For
the 14 antibiotics tested, strain 4GSKXᵀ was highly sensitive
to gentamicin, kanamycin, amikacin and netilimicin, but resis-
tant to all other 10 antibiotics tested. Detailed differential
phenotypic characteristics of strain 4GSKXᵀ and its closely related
described species are given in Table 1.

The fatty acid composition of the strain 4GSKXᵀ, compared
with closely related species of the genus Acidicapsa, is pre-
sented in Table 2. The major fatty acids (>10 %) of the cells
of strain 4GSKXᵀ were iso-C₁₅:₀ (48.8 %) and iso-C₁₇:₁ω₉c/C₁₆:₁ ω₁₀-methyl (14.6 %). The high percentage of iso-C₁₅:₀
is a typical characteristic of the genus Acidicapsa [5–7]. The presence of C₁₉:₀ω₇c cyclo and C₁₈:₁ω₇c/C₁₈:₁ω₆c in strain
4GSKXᵀ could be used to separate it from all members of the
genus Acidicapsa (all absence of these two fatty acids).

Position of the double bond from the methyl end; c, cis-isomer.

Table 2. Cellular fatty acid compositions (%) of strains 4GSKXᵀ and A. ligni WH120ᵀ

<table>
<thead>
<tr>
<th>Fatty acid (%)</th>
<th>4GSKXᵀ</th>
<th>A. ligni WH120ᵀ</th>
</tr>
</thead>
<tbody>
<tr>
<td>Saturated:</td>
<td></td>
<td></td>
</tr>
<tr>
<td>C₁₄:₀</td>
<td></td>
<td>2.3</td>
</tr>
<tr>
<td>C₁₅:₀ iso</td>
<td>48.8</td>
<td>55.4</td>
</tr>
<tr>
<td>C₁₆:₀</td>
<td>7.3</td>
<td>8.3</td>
</tr>
<tr>
<td>C₁₆:₀ 3OH</td>
<td>2.2</td>
<td></td>
</tr>
<tr>
<td>C₁₇:₀ iso</td>
<td>5.2</td>
<td>2.8</td>
</tr>
<tr>
<td>C₁₇:₀ anteiso</td>
<td></td>
<td></td>
</tr>
<tr>
<td>C₁₇:₀ cyclo</td>
<td>8.1</td>
<td>1.4</td>
</tr>
<tr>
<td>C₁₉:₀ω₈c cyclo</td>
<td>4.5</td>
<td></td>
</tr>
<tr>
<td>Unsaturated:</td>
<td></td>
<td></td>
</tr>
<tr>
<td>C₁₇:₁ω₈c</td>
<td></td>
<td>1.9</td>
</tr>
<tr>
<td>C₁₉:₁ω₉c</td>
<td></td>
<td>2.9</td>
</tr>
<tr>
<td>Summed features:</td>
<td></td>
<td></td>
</tr>
<tr>
<td>C₁₅:₁ iso H/C₁₃:₀ 3OH</td>
<td>1.3</td>
<td></td>
</tr>
<tr>
<td>C₁₆:₁ω₇c/C₁₈:₁ω₆c</td>
<td>2.1</td>
<td>17.7</td>
</tr>
<tr>
<td>C₁₈:₁ω₇c/C₁₈:₁ω₆c</td>
<td>3.5</td>
<td></td>
</tr>
<tr>
<td>iso-C₁₇:₁ω₆c/C₁₆:₀ 10-methyl</td>
<td>14.7</td>
<td>3.3</td>
</tr>
</tbody>
</table>
A. ligni WH120\textsuperscript{T} (3.3 \%). The major polar lipids of strain 4GSKX\textsuperscript{T} were phosphatidylethanolamine, an unidentified glycolipid, three unidentified phospholipids and two unidentified aminophospholipids (Fig. S2, available in the online version of this article). The predominant isoprenoid quinone was menaquinone-8 (MK-8) which is the common characteristics of Acidicapsa species. The genomic G+C content of strain 4GSKX\textsuperscript{T} was 52.8 mol\%, which is within the ranges of Acidicapsa.

The topology of the phylogenetic tree based on 16S rRNA gene sequences generated by ML (Fig. 1) showed that strain 4GSKX\textsuperscript{T} lay within the radiation of the genus Acidicapsa in the family Acidobacteriaceae, which was very similar to those generated by NJ and MP algorithms. 16S rRNA gene sequence similarity between 4GSKX\textsuperscript{T} and the type strains of closely related described Acidicapsa species ranged from 95.4 to 98.7 \%, with the lowest similarity between Acidicapsa acidisoli SK11\textsuperscript{T} and Acidicapsa ferrireducens MCF9\textsuperscript{T} and the highest between Acidicapsa acidiphila MCF10\textsuperscript{T} and Acidicapsa ligni WH120\textsuperscript{T}.

![Fig. 1. Maximum-likelihood tree based on 16S rRNA gene sequence showing the phylogenetic relationship between Acidicapsa dinghuensis sp. nov. 4GSKX\textsuperscript{T} and other representatives of the subdivision 1 of phylum Acidobacteria. Bootstrap values (1000 data resamplings) of >70 \% are shown. Filled circles at nodes indicate generic branches that were also recovered using neighbour-joining and maximum-parsimony algorithms. Six members of the phylum ‘Planctomycetes’, including Isosphaera pallida, Gemmata obscuriglobus, Planctomyces maris, Schlesneria paludicola and Singulisphaera acidiphila, were used as outgroups. Bar, 0.01 substitutions per nucleotide position.](image-url)
A. ferrireducens MCF9ᵀ and A. acidiphila MCF10ᵀ. The branches that were recovered congruently using ML, NJ and MP algorithms are marked with filled circles at the nodes in Fig. 1. The analysis of the 16S rRNA gene sequence showed that 4GSKXᵀ had the highest 16S rRNA gene sequence similarity of 97.1 % to A. ligni WH120ᵀ.

The characteristics that can be used to distinguish strain 4GSKXᵀ from its closely related Acidicapsa species are listed in detail in Table 1. As can be seen, except for the 16S rRNA gene sequence, which strongly supported the novel species status of 4GSKXᵀ phylogenetically, many phenotypic characters, such as enzymic activities, ability to utilize various carbon sources etc, can be used to distinguish strain 4GSKXᵀ from the closely related members of the genus Acidicapsa. Based on the phenotypic, chemotaxonomic and phylogenetic data shown above, we propose that strain 4GSKXᵀ represents a novel species of genus Acidicapsa, for which the name Acidicapsa dinghuensis sp. nov. is proposed, with 4GSKXᵀ as the type strain.

**DESCRIPTION OF ACIDICAPSA DINGHUENSIS SP. NOV.**

*Acidicapsa dinghuensis* (ding.hu.en’sis. N.L. fem. adj. ding-huensis pertaining to Mount Dinghu, PR China, the source of the soil from which the type strain was isolated).

Cells are Gram-stain-negative, aerobic, non-motile, non-spore-forming, non-capsule-forming short rods (0.4–0.8×0.5–1.5 μm) that reproduce by binary fission. Colonies formed on MM1F agar medium are circular, semitransparent beige, convex and mucous. Adaptable conditions for the growth include pH 4.0–6.5 (optimum, pH 4.5–5.0), temperature 12–37 °C (optimum, 25–30 °C) and NaCl concentration no higher that 1 % (w/v). The major polar lipids consist of three unidentified phospholipids, two unidentified amphotil phospholipids, and an unidentified glycolipid and phosphatidylethanolamine. The major cellular fatty acids are iso-C₁₅:₀ and iso-C₁₇:₁ω9c/C₁₆:₀ 10-methyl. Menaquinone-8 (MK-8) is the predominant respiratory quionone. Oxidase- and catalase-negative. Nitrate is not reduced to nitrite. Indole production reaction appears negative, as well as glucose fermentation reaction. Aesculin hydrolysis and gelatin hydrolysis reaction presents positive. Cells appear positive for the following enzymic activities: β-galactosidase, alkaline phosphatase, esterase (C4), leucine arylamidase, valine arylamidase, cystine arylamidase, acid phosphatase, naphthal-AS-BI-phosphohydrolase, α-galactosidase, β-galactosidase, β-glucuronidase, α-glucosidase, β-glucosidase, α-mannosidase and α-fucosidase. Enzymic activities are negative for urease, esterase (C8), lipase (C14), trypsin, α-chymotrypsin and N-acetyl-β-glucosaminidase. The following substrates can be utilized as sole carbon sources for growth: D-xylene, D-galactose, D-glucose, D-fructose, D-mannose, melibiose, sucrose, melezitose, D-gentiobiose, turanose, D-rhamnose, aesculin ferric citrate, inulin, glycerol, potassium 2-ketogluconate, methyl-α-D-mannopyranoside, and methyl β-D-xylpyranoside. The following substrates cannot be used as sole carbon sources: adipate, amygdalin, arbutin, caprate, D-adonitol, D-arabinose, D-arabitol, cellobiose, D-fucose, lactose, D-lxysose, maltose, D-mannitol, raffinose, D-ribose, D-sorbitol, D-tagatose, trehalose, dulcitol, erythritol, glycerogen, inositol, L-arabinose, L-arabitol, L-fucose, L-sorbose, L-xyllose, malate, methyl α-D-glucopyranoside, N-acetyl-D-glucosamine, phenylacetic acid, potassium 5-ketogluconate, potassium gluconate, salicin, starch, xyitol and trisodium citrate. Cells show susceptibility towards gentamicin, kanamycin, amikacin and netilmicin, but high resistance to tobramycin, penicillin, tetracycline, neomycin, vancomycin, chloramphenicol, erythromycin, ciprofloxacin, streptomycin and polymyxin B.

The type strain is 4GSKXᵀ (=CGMCC 1.15449ᵀ=LMG 29213ᵀ), which was isolated from upper layer soil collected from Dinghushan Biosphere Reserve, located at Guangdong Province, PR China.

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**Conflicts of interest**

The authors declare that there are no conflicts of interest.

**References**


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