Description and Genome Analysis of *Luteimonas Viscosa* Sp. Nov., A Novel Bacterium Isolated from Soil of a Sunflower Field

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Abstract

Strain XBU10^T was isolated from a soil sample of a sunflower plot in Inner Mongolia, China. The isolate was a Gram-stain-negative, aerobic, non-motile, rod-shaped bacterium, and its colonies were bright yellow in colour. Phylogenetic analysis based on 16S rRNA gene sequences indicated that strain XBU10^T belonged to the genus *Luteimonas* of the family *Lysobacteraceae* and was most closely related to *Luteimonas panaciterrae* Gsoil 068^T (97.8%), *Luteimonas marina* FR1330^T (97.6%), *Luteimonas aquatica* RIB1-20^T (97.4%) and *Luteimonas huabeiensis* HB2^T (97.2%). Growth occurred at 4–40 °C (optimum, 28–30 °C), with 0–5.0% (w/v) NaCl (optimum, 0.5%) and at pH 6.0–10.0 (optimum, pH 7.0–8.0). The chemotaxonomic characteristics of strain XBU10^T, which had Q-8 as its predominant quinone and iso-C_{17:1} ω9c, iso-C_{15:0}, iso-C_{17:0} and iso-C_{16:0} as its major fatty acids, were consistent with classification in the genus *Luteimonas*. The polar lipid profile of strain XBU10^T comprised phosphatidylglycerol, diphosphatidylglycerol, phosphatidylethanolamine, one unidentified phospholipid, two unidentified aminophospholipids and three unidentified polar lipids. The genome of strain XBU10^T was 4.17 Mbp with a G+C content of 69.9 mol%. Its genome sequence showed genes encoding alkaline phosphatase and catalase. Protein-coding genes related to carbohydrate-active enzymes were also observed. Average nucleotide identity (ANI) values between XBU10^T and other species of the genus *Luteimonas* were found to be low (ANIm < 88.0%, ANIb < 85.0% and OrthoANIu < 85.0%). Furthermore, digital DNA-DNA hybridization (dDDH) and average amino acid identity (AAI) values between strain XBU10^T and the closely related species ranged from 20.3 to 28.9% and from 64.2 to 82.3%, respectively. Based on the results of our phylogenetic, phenotypic, genotypic and chemotaxonomic analyses, it is concluded that strain XBU10^T represents a novel species within the genus *Luteimonas*, for which the name *Luteimonas viscosa* sp. nov. is proposed. The type strain is XBU10^T (=CGMCC 1.12158^T =KCTC 23878^T).

Introduction

The genus *Luteimonas* was initially proposed by Finkmann et al. (2000), with *Luteimonas mephitis* as the type species. The genus is a member of the family *Lysobacteraceae*, class *Gammaproteobacteria* and closely to the genera *Lysobacter*, *Xanthomonas*, *Pseudoxanthomonas* and *Stenotrophomonas*. According to LPSN (<https://lpsn.dsmz.de/genus/luteimonas>), the genus *Luteimonas* currently contains 24 species with validly published names. These members have been isolated from various environments such as sea water (Baik et al. 2008), soil (Wang et al. 2015; Lin et al. 2020; Xiong et al. 2020; Rani et al. 2016), rhizosphere (Zhao et al. 2018; Cheng et al. 2016), sediment (Fan et al. 2014; Roh et al. 2008), fresh water (Chou et al. 2008), food waste (Young et al. 2007), plant (Sun et al. 2012) and intertidal macroalga (Verma et al. 2016). The species within the genus *Luteimonas* have several common characteristics, including Gram-stain-negative, non-flagellated, non-spore-forming, aerobic, rod-shaped, yellow coloured, iso-C_{15:0} as the predominant fatty acid and the presence of ubiquinone-8 (Q-8) as the major ubiquinone. Some *Luteimonas* species not only play an important role in the ecosystem, but also have a considerable potential value in the field of basic research and biological engineering. Notably, *Luteimonas arsenica* is

tolerant to arsenite and arsenate (Mu et al. 2016), *Luteimonas terricola*, *L. mephitis*, "*Luteimonas dalianensis*" and *Luteimonas huabeiensis* are capable of reducing petroleum (Zhang et al. 2010; Finkmann et al. 2000; Xin et al. 2014; Wu et al. 2013). These species have potentials to be applied in bioremediation. Thus, isolation and characterization of a novel *Luteimonas* species from soil are crucial for our understanding of soil-related bacterial communities as well as for developing commercially useful metabolites. Accordingly, we here describe a novel bacterium belonging to the genus *Luteimonas*, designated strain XBU10^T, which was isolated from the soil of a sunflower field. It is interesting to note that genes related to degradation of aromatic compounds were found in strain XBU10^T, suggesting that the bacterium could be used in bioremediation. Apart from these, function genes related to alkaline phosphatase and catalase were also observed in strain XBU10^T.

Materials And Methods

Isolation and culture conditions

Strain XBU10^T was isolated from a saline-alkaline soil sample collected from a sunflower field in Wuyuan county (41° 04' 39" N, 108° 00' 21" E) in Inner Mongolia, China. To isolate bacteria, 1 g soil sample was suspended in 100 ml sterile water, and then diluted and spread onto tryptic soy agar (TSA) plates, followed by incubation at 28°C for 5 days. Single colonies were picked and re-streaked repeatedly onto new TSA plates until the purity was confirmed. Purified colonies of strain XBU10^T were maintained on TSA medium slants at 4 °C and preserved in 20% (v/v) glycerol at -20 °C.

16S rRNA gene sequencing and phylogenetic analysis

For phylogenetic analysis, genomic DNA of strain XBU10^T was obtained by using a commercial genomic DNA extraction kit (TianGen, China). The 16S rRNA genes were amplified by using universal primers 27F (5'-AGAGTTTGATCCTGGCTCAG-3') and 1492R (5'-TACGGTTACCTTGTACGACTT-3') (Weisburg et al. 1991). The resultant 16S rRNA gene sequence of strain XBU10^T was acquired and compared with the recognized species in the EzTaxon database (www.ezbiocloud.net/identify) (Yoon et al. 2017) to determine the closest type strains and calculate the levels of sequence similarity. Multiple sequence alignments were performed using the Clustal_X program (Thompson et al. 1997). Evolutionary distances were calculated according to Kimura's two-parameter model (Kimura 1980). The neighbour-joining (NJ) (Saitou et al. 1987), maximum-parsimony (MP) (Kluge et al. 1969) and maximum-likelihood (ML) (Felsenstein 1981) algorithms were used to reconstruct phylogenetic trees by using MEGA X software (Kumar et al. 2018). The topologies of the resultant trees were evaluated by using the bootstrap resampling method of Felsenstein (1985) with 1000 replicates.

Genome sequencing, assembly and function analysis

The whole genome sequence of the reference strain *Luteimonas panaciterrae* Gsoil 068^T for genomic comparison with strain XBU10^T was not available. For the comparison of genome relatedness, genomic

DNA of strain XBU10^T together with reference strain *L. panaciterrae* Gsoil 068^T were extracted following the method of Marmur (1961). Genome sequencing was performed using the HiSeq X Ten sequencer system (Illumina) with paired-end reads of 150 bp. Reads of each data sets were filtered, and high quality paired-end reads were assembled using SOAPdenovo version 2.04 (<http://soap.genomics.org.cn/>) (Li et al. 2010). The draft genome sequence data has been deposited in the GenBank/DDBJ/ENA. The genomic DNA G+C content was calculated directly from the draft genome sequence. The function of coding genes in the assembled genome were annotated by Gene Ontology (GO) (Ashburner et al. 2000), Clusters of Orthologous Groups (COG) (Galperin et al. 2015) and Kyoto Encyclopedia of Genes and Genomes (KEGG) (Minoru et al. 2016). The carbohydrate-active enzyme coding genes were annotated by HMMER searching (Finn et al. 2011) against the Carbohydrate-Active enZymes database (Lombard et al. 2014).

DNA-DNA hybridization and genome-based phylogenetic analysis

The available whole genomes of *Luteimonas* species were collected from the National Center for Biotechnology Information (NCBI) database. Average nucleotide identity (ANI) values using BLAST (ANiB), MUMmer (ANIm) and the OrthoANiU algorithm between strain XBU10^T and the reference strains were calculated by the JSpecies Web Server (<http://jspecies.ribohost.com/jspeciesws/>) (Richter et al. 2009) and the EzGenome web service (<https://www.ezbiocloud.net/tools/ani>) (Yoon et al. 2017). Moreover, digital DNA-DNA hybridization (dDDH) was performed at the Genome-to-Genome Distance Calculator (GGDC) website (<http://ggdc.dsmz.de/distcalc2.php>) (Meier-Kolthoff et al. 2013). The average amino acid identity (AAI) was assessed using an online AAI calculator (<http://enve-omics.ce.gatech.edu/aai/>). Two-way AAI analysis was performed. Protein sequence data from NCBI database. Additionally, a phylogenetic tree based on the whole genome sequences of strain XBU10^T and related species was constructed using the Type (Strain) Genome Server (<https://tygs.dsmz.de/>) (Meier-Kolthoff et al. 2019).

Morphological, physiological and biochemical analysis

Growth on different bacteriological culture media was assessed using nutrient agar (NA), marine agar 2216 (MA), Reasoner's 2A (R2A), Luria–Bertani agar (LB), MacConkey agar and TSA medium, with sub culturing at 28 °C for 7 days. The Gram stain reaction of strain XBU10^T was determined according to the method of Smibert and Krieg (1994). Colony characteristics were observed after incubation of the bacterial cells at 28 °C for 3 days on TSA medium. Cell size and morphology of strain XBU10^T was observed by using light microscope (BH-2, Olympus) and transmission electron microscopy (JEM-1400, JEOL), with cells grown for 3 days at 28 °C on TSA medium. Cell motility was performed depending on turbidity development in a tube containing semisolid medium and was confirmed by the hanging-drop method (Leifson 1960; Skerman 1967). Poly- β -hydroxybutyrate granule accumulation was observed by using light microscopy after staining of the cells with Sudan black (Baomanbio). Growth at various temperatures (2, 4, 10, 15, 20, 25, 28, 30, 33, 35, 37, 40, 42 and 45 °C) was tested on TSA medium for 15 days. NaCl tolerance for salinity was assessed by inoculating the strain in tryptone soya broth (TSB) medium supplemented with different NaCl concentrations (0–10.0%, at intervals of 0.5%, w/v). To assess

the pH range and the optimal pH (4.0–12.0, at intervals of 1.0 pH unit) for growth of the strain, cultures were incubated in TSB medium. The pH of the basal medium was adjusted using the buffer system: pH 4.0–5.0: 0.1 M citric acid/0.1 M sodium citrate; pH 6.0–8.0: 0.1 M KH_2PO_4 /0.1 M NaOH; pH 9.0–10.0: 0.1 M NaHCO_3 /0.1 M Na_2CO_3 ; pH 11.0: 0.05 M Na_2HPO_4 /0.1 M NaOH; pH 12.0: 0.2 M KCl/0.2 M NaOH (Xu et al. 2005). Catalase activity was detected by the production of bubbles after the addition of 3% (v/v) H_2O_2 solution. Oxidase activity was determined by the oxidation of 1% (w/v) tetramethyl-*p*-phenylenediamine (Tarrand et al. 1982). The anaerobic growth test was performed on TSA medium with 1 g pyrogalllic acid and 2 ml 10% (w/v) NaOH in the plate, which was then sealed with vaseline and growth detected for 2 weeks (Ping et al. 2020). Endospore formation was examined according to the Schaeffer-Fulton method by staining bacterial cells with malachite green (Chaudhary et al. 2016). Nitrate reduction, hydrolysis of Tweens 20, 40, 60, 80, casein, xylan and starch were investigated according to the methods of Dong & Cai (2001). Cells for physiological and biochemical characterization of strain XBU10^T were prepared by routine cultivation on TSA medium at 28°C. Other physiological and biochemical tests were performed using API 20E, API 20NE and API 50CH kits (bioMérieux) and a GN2 MicroPlate (Biolog). Enzyme activities were tested using an API ZYM kit (bioMérieux) according to the manufacturer's instructions. Antibiotic resistance was tested by placing antibiotic-impregnated discs (TIAN TAN) on TSA medium inoculated with bacterial cultures. The strain was recorded as susceptible to the antibiotic if the clear zone of growth inhibition was > 10 mm in diameter (Goodfellow et al. 1974).

Chemotaxonomic characterization

For analysis of fatty acid methyl esters, strain XBU10^T and its reference strains were cultured under the same conditions in TSB medium at 28 °C for 3–4 days. Fatty acid methyl esters were further prepared and analysed using the Sherlock Microbial Identification System (MIDI) (Athalye et al. 1985). The fatty acid methyl esters were obtained from the cells through saponification, methylation and extraction and were then separated on a gas chromatograph. The peaks were automatically integrated, the fatty acid names and percentages were determined using the MIDI Sherlock MIS system (Library: TSBA6; Version, 6.0B). Cells of strain XBU10^T for quinones and polar lipids were obtained by cultivating in TSB medium at 28 °C for 3–4 days. Cells were harvested by centrifugation, washed with distilled water and freeze-dried. The respiratory quinones were extracted and subsequently analysed by HPLC as previously described by Collins et al. (1977). Polar lipids were extracted from 100 mg freeze-dried cultures by the method described by Minnikin et al. (1984) and then they were separated by two-dimensional TLC on 10 × 10 cm silica-gel plates (Silica gel 60 F254, Merck), with chloroform/methanol/water (65:25:4, v/v) and chloroform/glacial acetic acid/methanol/water (80:15:12:4, v/v) used in the first and second dimensions, respectively. The lipid profile of strain XBU10^T was identified by spraying the following reagents: 10% ethanolic phosphomolybdic acid hydrate reagent (for total lipids), molybdenum blue reagent (for phospholipids) and 0.4% ninhydrin reagent (for aminolipids).

Results And Discussion

Phylogenetic characteristics

The almost complete 16S rRNA gene sequence for strain XBU10^T contained 1459 bp nucleotides. Comparative 16S rRNA gene sequence analyses showed that strain XBU10^T was phylogenetically affiliated with species of the genus *Luteimonas*, showing high similarity with strains *L. panaciterrae* Gsoil 068^T (97.8%), *Luteimonas marina* FR1330^T (97.6%), *Luteimonas aquatica* RIB1-20^T (97.4%) and *L. huabeiensis* HB2^T (97.2%). Moreover, 16S rRNA gene sequence similarities between strain XBU10^T and other type strains with validly published names of the genus *Luteimonas* were less than 96.9%. The NJ phylogenetic tree (Fig. 1) indicated that strain XBU10^T clustered with the members of the genus *Luteimonas* and formed a robust clade with the related species *L. marina* FR1330^T and *Luteimonas composti* CC-YY255^T. This phylogenetic relationship was also confirmed in the trees generated with the maximum-parsimony (Fig. S1, available in the online version of this article) and maximum-likelihood (Fig. S2) algorithms. According to phylogenetic analysis and sequence similarities of the 16S rRNA genes, strain XBU10^T represented a member of the genus *Luteimonas*.

Genome composition and DNA-DNA hybridisation

The assembled genome sequence of strain XBU10^T has been deposited in the GenBank database under the accession number VTFT000000000. The total length of the assembly contained 4 169 055 bp, comprising 9 contigs with an N50 value of 3 026 340 bp and 283× coverage. The draft genome contained 3579 predicted protein-coding sequences, 47 tRNAs, 3 rRNAs and 5 ncRNAs were predicted. In addition, the GenBank accession number for the whole genome sequence of the closest related type strain *L. panaciterrae* Gsoil 068^T is JAJAVH000000000. The genome of *L. panaciterrae* Gsoil 068^T was 4 093 429 bp with a 257× average genome coverage. A total of 3682 genes were predicted, including 53 RNA genes (46 tRNAs, 3 rRNAs and 4 other RNAs) and 3565 protein-coding genes. The DNA G+C content of strain XBU10^T was calculated directly from the genome sequence and determined to 69.9 mol%. This value falls within the range reported for the genus *Luteimonas* (64.3–73.4 mol%). General features of the draft genome of strain XBU10^T are given in Table S1. The ANI values between strain XBU10^T and several other *Luteimonas* species were found to be low (ANIm < 88.0%, ANIb < 85.0% and OrthoANIu < 85.0%), as given in Table S2. All the ANI values are below the 95–96% cut-off value previously proposed species delineation (Stackebrandt et al. 1994). dDDH values inferred by the GGDC were from 20.3 to 28.9%, which are lower than the accepted threshold value (70%) to separate two species in prokaryotic systematics (Meier-Kolthoff et al. 2013). Furthermore, the result of AAI analyses between strain XBU10^T and other reference strains were in the range 64.2–84.3%, and all values are far below the threshold of 95% for differentiating bacterial species (Konstantinidis et al. 2005; Luo et al. 2014). A genome-based phylogenetic tree was included in Fig. S3, which showed that strain XBU10^T was affiliated to the genus *Luteimonas*. These results strongly support that strain XBU10^T is different from all closely related species described previously and represents a novel species.

Genome features and function prediction

During the comparison from different databases, the GO annotation indicated that gene functions of strain XBU10^T covered all aspects of cellular metabolism, with the most diverse gene types and number associated with biological process (6164 genes), followed by the genes associated with cellular component (4824 genes) and the genes associated with molecular function (3675 genes). Notably, among these genes related to molecular function, the number of the genes associated with catalytic activity was high and up to 1593 (Fig. S4). The COG classification annotation indicated that 74.7% protein-coding genes could be annotated to COG. Among the 21 general COG functional categories, the detailed distribution of genes was as follows: transcription, 186 genes; cell wall/membrane/envelope biogenesis, 177 genes; translation, ribosomal structure and biogenesis, 172 genes; amino acid transport and metabolism, 170 genes; signal transduction mechanisms, 164 genes. Other information of the COG functional categories was presented in Fig. S5. KEGG metabolic pathways were classified according to the relationship between KO (KEGG ORTHOLOGY) and Pathway. Functional annotation of genes by comparisons against the manually curated KEGG GENES database revealed that there were 296 genes related to the amino acid metabolism, 105 genes related to the xenobiotics biodegradation and metabolism, 142 genes related to the lipid metabolism, 364 genes related to the carbohydrate metabolism, 162 genes related to the energy metabolism and 197 genes related to the signal transduction (Fig. S6). Regarding the carbohydrate related genes, including cellobiose phosphorylase (EC. 2.4.1.20), *N*-acetylglucosamine-6-phosphate deacetylase (EC. 3.5.1.25), lactase (EC 3.2.1.108) and trehalose 6-phosphate phosphatase (EC. 3.1.3.12). The presence of genes associated with D-cellobiose, *N*-acetyl-D-glucosamine, α -D-lactose and D-trehalose utilization supported the results obtained for the physiological tests. Apart from these, the genes for alkaline phosphatase (EC. 3.1.3.1) and catalase (EC. 1.11.1.6) were found in genome of strain XBU10^T, and these enzyme activities were positive in the enzyme tests. Analysis of the genome sequence of strain XBU10^T showed 253 genes encoding different CAZymes in six different classes: glycoside hydrolases (GHs), enzymes that catalyze the hydrolysis of glycosidic linkage of glucoside—71 gene counts; glycosyltransferases (GTs), involved in the formation of glycosidic bonds—70 gene counts; carbohydrate esterases (CEs), which hydrolyze carbohydrate esters—65 gene counts; auxiliary activities (AAs), redox enzymes that act in conjunction with CAZymes—19 gene counts; carbohydrate-binding modules (CBMs)—19 gene count and polysaccharide lyases (PLs), which perform non-hydrolytic cleavage of glycosidic bonds—9 gene counts (Fig. S7).

Morphological, physiological, and biochemical characteristics

Strain XBU10^T was a Gram-stain-negative, aerobic, non-motile and non-spore-forming bacterium. Colonies grown on TSA medium plates for 3 days at 28°C were circular, convex, smooth, bright yellow in colour, with entire margins and approximately 1.0–2.0 mm in diameter. Cells were rod-shaped and were 0.4–0.6 μ m in width and 1.3–2.4 μ m in length (Fig. 2). These phenotypic characteristics of strain XBU10^T were in line with the genus *Luteimonas*. Growth occurred at temperatures ranging from 4°C to 40°C (optimum, 28–30°C), at pH 6.0–10.0 (optimum, pH 7.0–8.0) and with 0–5.0% (optimum, 0.5%, w/v) NaCl concentration. Strain XBU10^T was positive for hydrolysis of aesculin and gelatin, assimilation of D-glucose and maltose, consistent with data for other reference strains. Detailed cultural, biochemical and

physiological properties of strain XBU10^T are given in the species description and the features that differentiate strain XBU10^T from closely related species of the genus *Luteimonas* are shown in Table 1. Several phenotypic and biochemical characteristics of strain XBU10^T clearly distinguished it from its closest related phylogenetic neighbours in the genus *Luteimonas* such as nitrate reduction, temperature range, NaCl tolerance, hydrolysis of casein, assimilation of malate, and utilization of glycyl L-aspartic acid, lactulose, L-proline, L-serine, L-threonine and Tween 40. In addition, enzyme activity of valine arylamidase was negative for strain XBU10^T while positive for the reference strains. These phenotypic characteristics clearly distinguished strain XBU10^T from other related species of the genus *Luteimonas*.

Chemotaxonomic characteristics

A comparison of the fatty acid compositions of strain XBU10^T and reference strains is presented in Table S3. The major fatty acids were iso-C_{17:1} ω 9c (34.7%), iso-C_{15:0} (21.0%), iso-C_{17:0} (11.9%) and iso-C_{16:0} (10.3%), similar to other members of the genus *Luteimonas*. Although the fatty acid composition of strain XBU10^T was consistent with that of the genus *Luteimonas*, there were marked between-strain differences in the proportions of these fatty acids. In particular, strain XBU10^T differed from recognized *Luteimonas* species based on its higher content of iso-C_{17:1} ω 9c and lower content of iso-C_{15:0} and C_{16:0}. The predominant respiratory quinone of strain XBU10^T was ubiquinone Q-8, which is in consistent with the quinone system of the genus *Luteimonas*. The main polar lipids detected in strain XBU10^T, as in many members of the genus *Luteimonas*, contained phosphatidylglycerol (PG), diphosphatidylglycerol (DPG) and phosphatidylethanolamine (PE). In addition, the polar lipids of strain XBU10^T included one unidentified phospholipid (PL), two unidentified aminophospholipids (APL1, APL2) and three unidentified polar lipids (L1–L3) (Fig. S8). All in all, the chemotaxonomic characteristics confirmed the phylogenetic affiliation of strain XBU10^T to the genus *Luteimonas*.

On the basis of phylogenetic analysis of its 16S rRNA gene sequence, together with phenotypic and chemotaxonomic analyses, and genome comparisons presented in this study, strain XBU10^T represents a novel species within the genus *Luteimonas*, for which the name *Luteimonas viscosa* sp. nov. is proposed.

Description of *Luteimonas viscosa* sp. nov.

Luteimonas viscosa (vis.co'sa. L. fem. adj. *viscosa*, sticky, a property of the colonies).

Cells are Gram-stain-negative, aerobic, non-motile, rod-shaped, 0.4–0.6 μ m in width and 1.3–2.4 μ m in length. Colonies on TSA medium plates are circular, convex, smooth, bright yellow in colour, with entire margins and approximately 1.0–2.0 mm in diameter after incubation for 3 days at 28°C. Cells grow well on TSA, NA, MA, LB agar and MacConkey agar medium, but poorly on R2A agar medium. Growth occurs at 4–40°C (optimum, 28–30°C), with 0–5.0% (w/v) NaCl (optimum, 0.5%) and at pH 6.0–10.0 (optimum, 7.0–8.0). Poly- β -hydroxybutyrate granule is not accumulated. Oxidase and catalase activities are positive. Nitrate could be reduced. Tweens 20, 40, 60 and 80 are hydrolysed, but casein, starch or xylan are not hydrolysed. According the result of antibiotic sensitivity tests, resistant to gentamicin (10 μ g),

lincomycin (10 µg), novobiocin (30 µg) and streptomycin (10 µg), but sensitive to ampicillin (10 µg), chloramphenicol (30 µg), erythromycin (15 µg), kanamycin (30 µg), nalidixic acid (30 µg), neomycin (30 µg), penicillin G (10 U), rifampicin (5 µg) and tetracycline (30 µg). In API ZYM strips, positive for *N*-acetyl- β -glucosaminidase, acid phosphatase, alkaline phosphatase, α -chymotrypsin, esterase (C4), esterase lipase (C8), lipase (C14) (weakly) and naphthol-AS-BI-phosphohydrolase, but negative for cystine arylamidase, α -fucosidase, α -galactosidase, β -galactosidase, α -glucosidase, β -glucosidase, β -glucuronidase, leucine arylamidase, α -mannosidase, trypsin and valine arylamidase. In the API 20NE and API 20E systems, there are positive results for hydrolysis of aesculin, gelatin (as slow reaction in 2–3 days) and 4-nitrophenyl- β -D-galactopyranoside (PNPG), assimilation of *N*-acetyl-D-glucosamine (weakly), L-arabinose, D-glucose and maltose, but negative results for arginine dihydrolase, lysine decarboxylase, ornithine decarboxylase, tryptophan deaminase, urease, citrate utilization, H₂S and indole production, as well as assimilation of adipate, caprate, gluconate, malate, mannitol, mannose and phenylacetate. In the API 50CH tests, positive for acid production from aesculin ferric citrate and D-turanose (weakly), but negative for acid production from other substrates of the system. The following carbon sources are utilized (positive in the Biolog GN2 system): L-alaninamide, L-alanyl glycine, L-arabinose, D-cellobiose, dextrin, D-fructose, gentiobiose, α -D-glucose, glycyl L-glutamic acid, β -hydroxybutyric acid, α -D-lactose, lactulose, maltose, D-melibiose, pyruvic acid methyl ester, sucrose (weakly), D-trehalose, turanose and Tween 80. The remaining substrates of the Biolog GN2 system are not utilized. The major isoprenoid quinone is ubiquinone Q-8. Cellular fatty acids mainly consist of iso-C_{17:1} ω 9c, iso-C_{15:0}, iso-C_{17:0} and iso-C_{16:0}. The polar lipid profile consists of phosphatidylglycerol, diphosphatidylglycerol, phosphatidylethanolamine, one unidentified phospholipid, two unidentified aminophospholipids and three unidentified polar lipids.

The type strain, XBU10^T (=CGMCC 1.12158^T =KCTC 23878^T) was isolated from soil of a sunflower plot in Inner Mongolia, China. The DNA G+C content of the type strain is 69.9 mol%. Accession numbers are MN850872 (16S rRNA gene) and VTFT00000000 (whole genome).

Declarations

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Author's contributions

Jianli Zhang, Huancheng Pang, Yuyi Li and Ya Chen designed research and project outline. Ya Chen, Ying Zhang, Xiaonan Luo and Di Xin performed isolation, deposition and polyphasic taxonomy. Ya Chen and Ying Zhang performed genome analysis. Ya Chen, Ying Zhang and Di Xin drafted the manuscript. Jianli Zhang revised the manuscript. All authors read and approved the final manuscript.

Compliance with ethical standards

Conflict of interest

The authors declare that they have no conflict of interest.

Ethical approval

This article does not contain any studies with human participants or animals performed by any of the authors.

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Tables

Table 1. Differential characteristics between strain XBU10^T and the closely related species of *Luteimonas*

Strains: 1, Strain XBU10^T; 2, *Luteimonas panaciterrae* Gsoil 068^T; 3, *Luteimonas marina* FR1330^T; 4, *Luteimonas aquatica* RIB1-20^T; 5, *Luteimonas huabeiensis* HB2^T; 6, *Luteimonas composti* CC-YY255^T. All were positive for the following: hydrolysis of aesculin and gelatin; assimilation of D-glucose and maltose; activity of *N*-acetyl- β -glucosaminidase. All were negative for the following: assimilation of phenylacetate; activity of α -fucosidase, α -galactosidase and β -glucuronidase. All data are from this study except where otherwise indicated. +, Positive; –, negative; w, weakly positive.

Characteristic	1	2	3	4	5	6
Source of isolation	Soil	Soil	Seawater	Fresh water	Stratum water	Food waste
Colony colour	Yellow	Cream-light yellow	Light yellow	Yellow	Yellow	Yellow
Temperature range (°C)	4-40	15-45	20-42	15-37	20-45	20-45
Temperature optimum (°C)	28-30	30	30	25	30	30
pH range	6.0-10.0	5.0-8.0	6.0-10.0	7.0-8.0	6.0-11.0	7.0-10.0
pH optimum	7.0-8.0	6.0-7.0	8.0	7.0	7.0	7.0
NaCl range (% w/v)	0-5.0	0-3.0	0-1.0	0-3.0	0-5.0	0-6.0
NaCl optimum (% w/v)	0.5	0	0	0	0-1.0	0-0.5
Nitrate reduction	+	-	-	-	-	+
Catalase	+	-	+	w	+	+
Hydrolysis of:						
Urea	-	-	-	-	+	-
Starch	-	-	-	w	-	-
Casein	-	+	+	+	+	+
Assimilation of (API 20NE):						
L-Arabinose	+	-	+	-	-	+
N-Acetyl-D-glucosamine	w	+	+	+	+	+
Caprate	-	+	-	-	-	-
Adipate	-	-	-	-	+	-
Malate	-	+	+	+	+	-
Citrate	-	w	+	-	-	-
Activity of (API ZYM):						
Alkaline phosphatase	+	-	+	+	+	+
Esterase (C4)	+	-	+	+	+	+
Esterase lipase (C8)	+	-	+	+	+	+
Lipase (C14)	w	-	-	-	+	-
Leucine arylamidase	-	-	+	+	+	+
Valine arylamidase	-	+	+	+	+	+
Cystine arylamidase	-	-	-	+	+	-
Trypsin	-	+	+	-	+	-
α -Chymotrypsin	+	+	+	-	+	+
Acid phosphatase	+	-	+	+	+	+
Naphthol-AS-BI-phosphohydrolase	+	-	+	+	+	+
β -Galactosidase	-	-	-	+	-	+
α -Glucosidase	-	+	+	-	-	+
β -Glucosidase	-	+	+	-	-	+
α -Mannosidase	-	+	+	-	-	-
Utilization of (Biolog GN2):						
α -Cyclodextrin	-	-	+	-	-	-
Glycogen	-	+	-	+	-	-

Tween 40	-	+	+	+	+	-
N-Acetyl-D-galactosamine	-	w	+	+	-	-
N-Acetyl-D-glucosamine	-	+	w	+	+	-
L-Arabinose	+	-	+	-	-	-
D-Cellobiose	+	+	+	-	+	-
D-Fructose	+	+	-	+	+	-
L-Fucose	-	-	-	+	-	-
Gentiobiose	+	-	+	+	+	+
α -D-Lactose	+	-	-	+	-	-
Lactulose	+	-	-	-	-	-
Maltose	+	+	+	+	-	+
D-Mannose	-	+	-	+	-	-
D-Melibiose	+	-	+	-	-	+
Methyl β -D-glucoside	-	-	-	+	-	-
D-Psicose	-	-	-	+	-	-
D-Sorbitol	-	-	-	+	-	-
Sucrose	w	-	-	+	-	-
D-Trehalose	+	-	-	+	-	-
Turanose	+	w	-	+	-	+
Succinic acid monomethyl eater	-	+	-	-	+	-
Acetic acid	-	+	w	+	+	-
Formic acid	-	-	-	+	+	-
D-Galactonic acid lactone	-	-	-	+	-	-
D-Galacturonic acid	-	-	-	+	-	-
D-Glucosaminic acid	-	-	-	+	-	-
α -Hydroxybutyric acid	-	-	+	-	+	-
β -Hydroxybutyric acid	+	+	-	+	+	-
<i>p</i> -Hydroxyphenylacetic acid	-	-	-	+	-	-
α -Ketobutyric acid	-	+	w	+	+	-
α -Ketoglutaric acid	-	w	-	+	+	-
α -Ketovaleric acid	-	-	-	+	-	-
DL-Lactic acid	-	-	-	+	+	-
Malonic acid	-	-	-	+	-	-
Propionic acid	-	+	-	+	+	-
Sebacic acid	-	-	-	+	-	-
Succinic acid	-	+	+	-	+	-
Bromosuccinic acid	-	+	w	+	-	-
Succinamic acid	-	w	-	+	-	-
L-Alaninamide	+	w	+	-	-	+
L-Alanine	-	-	-	+	-	-
L-Alanyl glycine	+	w	+	+	-	+
L-Aspartic acid	-	+	-	+	-	-

L-Glutamic acid	-	+	+	+	-	-
Glycyl L-aspartic acid	-	+	+	+	+	+
Hydroxy-L-proline	-	-	-	+	-	-
L-Leucine	-	-	-	+	-	-
L-Ornithine	-	-	w	+	-	-
L-Proline	-	+	+	+	+	-
D-Serine	-	w	-	-	-	-
L-Serine	-	+	+	+	+	-
L-Threonine	-	w	w	+	+	-
Urocanic acid	-	+	-	+	-	-
Inosine	-	-	-	+	-	+
Uridine	-	-	-	+	-	-
Glycerol	-	+	-	-	-	-
α -D-Glucose 1-phosphate	-	+	-	+	+	-
D-Glucose 6-phosphate	-	-	w	-	+	-
DNA G+C content (mol%)	69.9	67.0*	67.6 [†]	70.3 [‡]	67.0 [§]	68.1 [¶]

* Data from Ten et al. (2009)

† Data from Baik et al. (2008)

‡ Data from Chou et al. (2008)

§ Data from Wu et al. (2013)

¶ Data from Young et al. (2007)

Figures

Figure 1

The neighbour-joining tree based on 16S rRNA gene sequences of strain XBU10^T and phylogenetically related type strains in the family *Lysobacteraceae*. Bootstrap values of above 50% based on 1000 replications are shown at branch nodes. GenBank accession numbers are given in parentheses. The sequence of *Escherichia coli* ATCC 11775^T is included as an outgroup. Bar represents 0.01 substitutions per nucleotide position.

Figure 2

Transmission electron micrograph of a cell of strain XBU10^T, grown on TSA medium for 3 days at 28 °C. Bar, 0.2 μm.

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