Chryseobacterium flavum sp. nov., isolated from polluted soil

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A Gram-negative, non-motile, rod-shaped bacterial strain, designated CW-E 2^T, was isolated from a polluted soil sample collected from Jiangsu Province, China. A taxonomic study of the isolate, including phylogenetic analysis based on 16S rRNA gene sequences and phenotypic characteristics, was carried out. The predominant menaquinone was MK-6 and the major fatty acids were i-C_{15:0}, i-C_{17:0} 3-OH, i-C_{17:1} ω9c and summed feature 4. The G + C content of the DNA was 37.2 mol%. Based on phenotypic and genotypic characteristics, strain CW-E 2^T represents a novel species of the genus *Chryseobacterium* for which the name *Chryseobacterium flavum* sp. nov. is proposed. The type strain is CW-E 2^T (=KCTC 12877^T=CCTCC AB 206147^T).

The family *Flavobacteriaceae* represents the main bacterial lineage in the phylum *Bacteroidetes* (formerly the *Cytophaga–Flexibacter–Bacteroides* group) (Bernardet *et al.*, 1996, 2002). Among the genera currently included in the family, *Chryseobacterium*, *Bergeyella*, *Ornithobacterium*, *Empedobacter*, *Weeksella*, *Wautersiella*, *Elizabethkingia*, *Sejongia* and *Kaistella* form a separate branch in phylogenetic trees (Vandamme *et al.*, 1994; Kim *et al.*, 2004, 2005b; Bernardet *et al.*, 2002, 2006). At the time of writing, the genus *Chryseobacterium* contains 18 species with validly published names (Shimomura *et al.*, 2005; Kim *et al.*, 2005a; Young *et al.*, 2005; de Beer *et al.*, 2005, 2006; Bernardet *et al.*, 2006; Quan *et al.*, 2007). Members of the genus *Chryseobacterium* are strictly aerobic, non-motile, Gram-negative rods that produce flexirubin-type pigments and have a DNA G + C content of about 29–39 mol%. Members of the genus *Chryseobacterium* occur in a variety of environments, including soil, fresh- and seawater, raw milk and chicken, diseased fish, bioreactor sludge and

clinical samples (Yamaguchi & Yokoe, 2000; Hugo *et al.*, 2003; Kim *et al.*, 2005a; de Beer *et al.*, 2005, 2006; Bernardet *et al.*, 2006; Quan *et al.*, 2007). In the course of an investigation of the bacterial community in a soil sample contaminated by atrazine, ethametsulfuron, glyphosate and other herbicides from a chemical factory in the suburb of Nanjing City, Jiangsu Province, China, one bacterial strain, CW-E 2^T, which had the ability to decompose atrazine (data not shown), was isolated and subjected to a polyphasic taxonomy study. It is proposed that this strain represents a novel species in the genus *Chryseobacterium*, *Chryseobacterium flavum* sp. nov.

Strain CW-E 2^T was isolated by plating dilutions of a soil sample on LB agar (Sambrook & Russell, 2002) incubated at 30 °C for 3 days. The strain was preserved in a 20 % (v/v) glycerol solution in distilled water at –80 °C. Biomass for molecular systematic and chemotaxonomic studies was obtained from LB broth (pH adjusted to 7.0) cultures incubated at 30 °C for 2 days in shake flasks (about 180 r.p.m.).

Cellular morphology of the isolate was observed by transmission electron microscopy (TEM) (model H-7650; Hitachi) examination of bacterial cells grown for 18 h on LB agar and

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Abbreviation: TEM, transmission electron microscopy.

The GenBank/EMBL/DDBJ accession number for the 16S rRNA gene sequence of strain CW-E 2^T is EF154516.

Fig. 1. TEMs of cells of strain CW-E 2^T grown on LB agar for 18 h at 30 °C. Arrows indicate fimbriae. Bars, 200 nm.

prepared as described by Nedashkovskaya *et al.* (2005) (Fig. 1). Acid production from carbohydrates was investigated using the media and methods described by Yamaguchi & Yokoe (2000). Utilization of various carbon sources was investigated by using modified ammonia salt-sugar medium [0.2 % (NH₄)₂SO₄, 0.05 % NaH₂PO₄ · H₂O, 0.02 % MgSO₄ · 7H₂O, 0.01 % CaCl₂ · 2H₂O and 0.05 % K₂HPO₄] containing 1 % of each tested carbon source. Growth was tested at 0, 5, 10, 25, 30, 35, 37, 40, 42, 45 and 50 °C on LB agar. NaCl and pH tolerance were tested using LB broth adjusted to various pH values (3.0, 6.0, 7.0, 7.8, 8.0, 10.0, 11.0 and 12.0) and NaCl concentrations (1, 4, 7 and 10 %) (Zhou *et al.*, 2007). Growth was tested on cetrimide agar, β -hydroxybutyrate agar, MacConkey agar and Simmons' citrate agar. Susceptibility to antibiotics was examined as described by Nedashkovskaya *et al.* (2003, 2005). Flexirubin-type pigments were identified using the KOH test as described by Bernardet *et al.* (2002). Gram-staining and other phenotypic characteristics were determined

according to the methods of Gerhardt *et al.* (1994). Detailed morphological, physiological and biochemical characteristics of strain CW-E 2^T are given in Table 1 and the species description.

Menaquinones were extracted from lyophilized cells and determined according to Hu *et al.* (2001). The predominant menaquinone was MK-6. The fatty acids were extracted, methylated and analysed using the standard MIDI (Microbial Identification) system as described by Sasser (1990). The major fatty acids of strain CW-E 2^T were i-C_{15:0}, i-C_{17:0} 3-OH, i-C_{17:1} ω 9c and summed feature 4 (comprising i-C_{15:0} 2-OH and/or C_{16:1} ω 7c and/or C_{16:1} ω 7t). Detailed fatty acid profiles of strain CW-E 2^T and its closest relatives are given in Table 2.

Genomic DNA of strain CW-E 2^T was prepared following the method of Marmur (1961) and the DNA G + C content was determined using the thermal denaturation method (Mandel & Marmur, 1968). The DNA G + C content of strain CW-E 2^T was 37.2 mol%.

Table 1. Phenotypic characteristics that differentiate *Chryseobacterium flavum* CW-E 2^T from its closest relatives

Species: 1, *C. flavum* (n=1); 2, *C. joostei* (n=11); 3, *C. gleum* (n=5); 4, *C. indologenes* (n=45); 5, *C. shigense* (n=1); 6, *C. vrystaatense* (n=36). +, Positive; –, negative; v, variable; D, delayed; NA, no data available; PS, polluted soil; RM, raw milk; RC, raw chicken; CS, clinical samples; LAB, lactic acid beverage. For all species, indole production was positive. Data from Hugo *et al.* (2003), Shimomura *et al.* (2005) and de Beer *et al.* (2005).

Characteristic	1	2	3	4	5	6
Habitat	PS	RM	CS	CS	LAB	RC
Growth on/at:						
MacConkey agar	+	+	+	+	–	–
5 °C	+	+	–	–	+	+
37 °C	+	–	+	+	–	–
42 °C	+	–	–	–	–	–
Growth on nutrient agar supplemented with 5 % NaCl	+	NA	NA	NA	NA	–
Enzyme activities:						
DNase	–	+	+	+	NA	+
Urease	–	v	+	–	–	+
β -Galactosidase	+	–	–	–	NA	NA
Nitrate reduction	–	–	+	–	–	–
Nitrite reduction	–	–	+	–	NA	+
Hydrolysis of:						
Tyrosine	+	+	+	–	NA	–
Tween 80	+	+	+	+	NA	+
Starch	+	+	+	+	+	–
Acid production from:						
D-Fructose	+	+	+	D	+	NA
D-Mannitol	–	v	–	–	–	NA
D-Xylose	+	–	+	–	–	NA
Mean DNA G + C content (mol%)	37.2	36.8	38	38.5	36.6	37.1

Table 2. Cellular fatty acid content (%) of *Chryseobacterium flavum* CW-E 2^T and its closest relatives

Species: 1, *C. flavum* (n=1); 2, *C. joostei* (n=11); 3, *C. gleum* (n=5); 4, *C. indologenes* (n=45); 5, *C. shigense* (n=1); 6, *C. vrystaatense* (n=36). Some of the *Chryseobacterium* species included in this table were cultivated under different conditions. Means $\pm$ SD are given where more than one strain was tested. ND, Not detected; TR, trace (fatty acids amounting to less than 1 %). Data from Hugo *et al.* (2003), Shimomura *et al.* (2005) and de Beer *et al.* (2005).

Fatty acid	1	2	3	4	5	6
i-C _{15:0}	35.1	34.6 $\pm$ 2.0	35.4 $\pm$ 2.9	34.3 $\pm$ 4.9	39.7	41.8 $\pm$ 1.4
i-C _{15:0} 3-OH	3.7	2.9 $\pm$ 0.3	2.5 $\pm$ 0.1	2.6 $\pm$ 0.2	4.1	2.7 $\pm$ 0.3
C _{16:0}	1.1	TR	1.3 $\pm$ 0.4	TR	TR	1.1 $\pm$ 0.3
C _{16:0} 3-OH	2.0	1.2 $\pm$ 0.2	1.1 $\pm$ 0.1	1.0 $\pm$ 0.2	1.3	1.3 $\pm$ 0.3
i-C _{17:0}	TR	TR	1.6 $\pm$ 0.6	TR	TR	TR
i-C _{17:0} 3-OH	18.5	20.1 $\pm$ 1.2	21.8 $\pm$ 0.3	19.2 $\pm$ 1.8	19.6	15.4 $\pm$ 1.8
i-C _{17:1ω9c}	16.8	22.9 $\pm$ 1.9	20.2 $\pm$ 3.9	24.2 $\pm$ 3.1	11.9	19.7 $\pm$ 2.3
Unknown 13.566*	1.9	1.1 $\pm$ 0.2	1.2 $\pm$ 0.4	2.1 $\pm$ 0.7	2.4	1.4 $\pm$ 0.3
Unknown 16.580*	1.5	1.6 $\pm$ 0.1	1.7 $\pm$ 0.1	1.7 $\pm$ 0.2	1.0	1.2 $\pm$ 0.2
Summed feature 4†	13.3	12.1 $\pm$ 1.3	11.8 $\pm$ 0.8	11.1 $\pm$ 1.3	12.4	9.1 $\pm$ 2.3

*Numbers indicate the equivalent chain-length.

†Fatty acids that could not be separated by GC using the Microbial Identification System (Microbial ID) software were considered summed features. Summed feature 4 contains i-C_{15:0} 2-OH and/or C_{16:1 ω 7c} and/or C_{16:1 ω 7t}.

PCR amplification of the 16S rRNA gene was performed as described by Xu *et al.* (2003). Phylogenetic analysis was performed using the software packages PHYLIP (Felsenstein, 1993) and MEGA version 3.1 (Kumar *et al.*, 2001) after multiple alignment of data by CLUSTAL_X (Thompson *et al.*, 1997). Distances (distance options according to the Kimura two-parameter model; Kimura, 1980, 1983) and clustering were based on the neighbour-joining (Saitou & Nei, 1987) and maximum-likelihood (Felsenstein, 1981) methods. Bootstrap analysis (1000 resamplings) was used to evaluate the topology of the neighbour-joining tree (Felsenstein, 1985).

The almost complete 16S rRNA gene sequence (1440 bp in length) of strain CW-E 2^T and reference sequences selected from GenBank following BLAST searches were used to draw the phylogenetic tree. Phylogenetic analyses showed that strain CW-E 2^T belongs to the genus *Chryseobacterium* but shares only 94.9, 93.9 and 93.9 % 16S rRNA gene sequence similarity with its closest relatives *Chryseobacterium joostei* LMG 18212^T, *Chryseobacterium gleum* ATCC 35910^T and *Chryseobacterium indologenes* ATCC 29897^T, respectively (Fig. 2).

Differential phenotypic characteristics (Table 1) could be used to distinguish the isolate from other *Chryseobacterium* species. Hence, strain CW-E 2^T represents a novel species for which the name *Chryseobacterium flavum* sp. nov. is proposed.

Description of *Chryseobacterium flavum* sp. nov.

Chryseobacterium flavum (fla'vum. L. neut. adj. *flavum* yellow, the colour of colonies or pigment).

Cells are Gram-negative, non-motile, non-spore-forming, aerobic and chemo-organotrophic rods (0.5–0.7 $\times$ 1.1–

1.3 μ m) that are covered by fimbriae. Colonies on LB agar plates are circular, convex, shiny, bright-yellow-coloured and smooth, 1.0–1.5 mm in diameter, with a ropy consistency. Growth occurs on trypticase soy agar, cetrimide agar, MacConkey agar and β -hydroxybutyrate (β -hydroxybutyrate inclusions are not produced). No growth occurs on Simmons' citrate agar. Growth occurs at 5–42 °C, pH 5.0–9.0 and with 0–6 % NaCl (optimum growth at 30–35 °C, pH 6.0–7.0 and 1 % NaCl). Non-diffusible flexirubin-type yellow pigments are produced. Casein, gelatin, starch and Tween 20 are hydrolysed. DNA, cellulose and chitin are not hydrolysed. Acid is produced from D-fructose, maltose, trehalose and D-xylose; but not from adonitol, D-arabinose, cellobiose, dulcitol, L-fucose, D-galactose, D-glucose, inositol, D-lactose, D-mannitol, D-melezitose, D-melibiose, L-raffinose, L-rhamnose, D-sorbitol, L-sorbose or sucrose. Adonitol, aesculin, D-arabinose, cellobiose, dextrin, D-fructose, D-galactose, glucosamine, D-glucose, inositol, inulin, D-lactose, maltose, D-mannose, D-melezitose, D-melibiose, D-ribose, D-sorbitol, sucrose, trehalose, turanose, xylitol and D-xylose are utilized. Citrate, acetate, adonitol, dulcitol, D-erythrose, malonate, D-mannitol, melezitose, D-raffinose, tartrate, salicin and D-sorbitose are not utilized. Oxidase, catalase, β -galactosidase, phenylalanine decarboxylase, arginine dihydrolase and lipase activities are present. Methyl α -D-glucosidase, ornithine decarboxylase, lysine decarboxylase, arginine decarboxylase, urease and DNase activities are absent. Nitrate and nitrite are not reduced. H₂S (triple-sugar iron test) is not produced. Methyl red and Voges–Proskauer tests are negative. Indole is produced. KCN (0.0075 %) is not tolerated. Susceptible to carbenicillin, tetracycline, doxycycline, gentamicin, penicillin G, kanamycin, vancomycin, erythromycin, rifampicin, neomycin, streptomycin

Fig. 2. Neighbour-joining phylogenetic tree based on 16S rRNA gene sequences of strain CW-E 2^T, other members of the genus *Chryseobacterium*, and representatives of the family *Flavobacteriaceae*. Numbers at nodes indicate bootstrap values (%) greater than 50%. Bar, 0.02 substitutions per nucleotide position. The maximum-likelihood tree showed essentially the same topology (data not shown).

and chloramphenicol; resistant to ampicillin, lincomycin and polymyxin B. The predominant menaquinone is MK-6. The major fatty acids are i-C_{15:0} (35.1%), i-C_{17:0} 3-OH (18.5%), i-C_{17:1}ω9c (16.8%) and summed feature 4 (13.3%, comprising i-C_{15:0} 2-OH and/or C_{16:1}ω7c and/or C_{16:1}ω7t). The DNA G+C content is 37.2 mol%.

The type strain is CW-E 2^T (=KCTC 12877^T=CCTCC AB 206147^T), isolated from the surface layer of polluted soil from Nanjing, Jiangsu Province, China.

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References

Bernardet, J.-F., Segers, P., Vancanneyt, M., Berthe, F., Kersters, K. & Vandamme, P. (1996). Cutting a Gordian knot: emended classification and description of the genus *Flavobacterium*, emended description of the family *Flavobacteriaceae*, and proposal of

Flavobacterium hydatidis nom. nov. (basonym, *Cytophaga aquatilis* Strohl and Tait 1978). *Int J Syst Bacteriol* **46**, 128–148.

Bernardet, J.-F., Nakagawa, Y. & Holmes, B. (2002). Proposed minimal standards for describing new taxa of the family *Flavobacteriaceae* and emended description of the family. *Int J Syst Evol Microbiol* **52**, 1049–1070.

Bernardet, J.-F., Hugo, C. & Bruun, B. (2006). The genera *Chryseobacterium* and *Elizabethkingia*. In *The Prokaryotes: Handbook on the Biology of Bacteria*, 3rd edn, vol. 7, pp. 638–676. Edited by M. Dworkin, S. Falkow, E. Rosenberg, K.-H. Schleifer & E. Stackebrandt. New York: Springer-Verlag.

de Beer, H., Hugo, C.-J., Jooste, P.-J., Willems, A., Vancanneyt, M., Coenye, T. & Vandamme, P. (2005). *Chryseobacterium vrystaatense* sp. nov., isolated from raw chicken in a chicken-processing plant. *Int J Syst Evol Microbiol* **55**, 2149–2153.

de Beer, H., Hugo, C.-J., Jooste, P.-J., Vancanneyt, M., Coenye, T. & Vandamme, P. (2006). *Chryseobacterium piscium* sp. nov., isolated from fish of the South Atlantic Ocean off South Africa. *Int J Syst Evol Microbiol* **56**, 1317–1322.

Felsenstein, J. (1981). Evolutionary trees from DNA sequences: a maximum likelihood approach. *J Mol Evol* **17**, 368–376.

Felsenstein, J. (1985). Confidence limits on phylogenies: an approach using the bootstrap. *Evolution* **39**, 783–791.

Felsenstein, J. (1993). PHYLIP (phylogeny inference package), version 3.5c. Distributed by the author. Department of Genome Sciences, University of Washington, Seattle, USA.

Gerhardt, P., Murray, R. G. E., Wood, W. A. & Krieg, N. R. (editors) (1994). *Methods for General and Molecular Bacteriology*. Washington, DC: American Society for Microbiology.

- Hu, H.-Y., Lim, B.-R., Naohiro, G. & Koich, F.-J. (2001). Analytical precision and repeatability of respiratory quinones for quantitative study of microbial community structure in environmental samples. *J Microbiol Methods* **47**, 17–24.
- Hugo, C.-J., Segers, P., Hoste, B., Vancanneyt, M. & Kersters, K. (2003). *Chryseobacterium joostei* sp. nov., isolated from the dairy environment. *Int J Syst Evol Microbiol* **53**, 771–777.
- Kim, M. K., Im, W. T., Shin, Y. K., Lim, J. H., Kim, S. H., Lee, B. C., Park, M. Y., Lee, K. Y. & Lee, S. T. (2004). *Kaistella koreensis* gen. nov., sp. nov., a novel member of the *Chryseobacterium*–*Bergeyella*–*Riemerella* branch. *Int J Syst Evol Microbiol* **54**, 2319–2324.
- Kim, K. K., Bae, H.-S., Schumann, P. & Lee, S. T. (2005a). *Chryseobacterium daecheongense* sp. nov., isolated from freshwater lake sediment. *Int J Syst Evol Microbiol* **55**, 133–138.
- Kim, K. K., Kim, M. K., Lim, J. H., Park, H. Y. & Lee, S. T. (2005b). Transfer of *Chryseobacterium meningosepticum* and *Chryseobacterium miricola* to *Elizabethkingia* gen. nov. as *Elizabethkingia meningoseptica* comb. nov. and *Elizabethkingia miricola* comb. nov. *Int J Syst Evol Microbiol* **55**, 1287–1293.
- Kimura, M. (1980). A simple method for estimating evolutionary rates of base substitutions through comparative studies of nucleotide sequence. *J Mol Evol* **16**, 111–120.
- Kimura, M. (1983). *The Neutral Theory of Molecular Evolution*. Cambridge: Cambridge University Press.
- Kumar, S., Tamura, K., Jakobsen, I.-B. & Nei, M. (2001). MEGA2: molecular evolutionary genetics analysis software. *Bioinformatics* **17**, 1244–1245.
- Mandel, M. & Marmur, J. (1968). Use of ultraviolet absorbance-temperature profile for determining the guanine plus cytosine content of DNA. *Methods Enzymol* **12B**, 195–206.
- Marmur, J. (1961). A procedure for the isolation of deoxyribonucleic acid from microorganisms. *J Mol Biol* **3**, 208–218.
- Nedashkovskaya, O. I., Suzuki, M., Vysotskii, M. V. & Mikhailov, V. V. (2003). *Reichenbachia agariperforans* gen. nov., sp. nov., a novel marine bacterium in the phylum *Cytophaga*–*Flavobacterium*–*Bacteroides*. *Int J Syst Evol Microbiol* **53**, 81–85.
- Nedashkovskaya, O. I., Kim, S. B., Suzuki, M., Shevchenko, L. S., Lee, M. S., Lee, K. H., Park, M. S., Frolova, G. M., Oh, H. W. & other authors (2005). *Pontibacter actiniarum* gen. nov., sp. nov., a novel member of the phylum ‘*Bacteroidetes*’, and proposal of *Reichenbachia* gen. nov. as a replacement for the illegitimate prokaryotic generic name *Reichenbachia* Nedashkovskaya et al. 2003. *Int J Syst Evol Microbiol* **55**, 2583–2588.
- Quan, Z.-X., Kim, K. K., Kim, M. K., Jin, L. & Lee, S. T. (2007). *Chryseobacterium caeni* sp. nov., isolated from bioreactor sludge. *Int J Syst Evol Microbiol* **57**, 141–145.
- Saitou, N. & Nei, M. (1987). The neighbor-joining method: a new method for reconstructing phylogenetic trees. *Mol Biol Evol* **4**, 406–425.
- Sambrook, J. & Russell, D. W. (2002). *Molecular Cloning: a Laboratory Manual*, pp. 1595–1596, 3rd edn. Cold Spring Harbor, NY: Cold Spring Harbor Laboratory.
- Sasser, M. (1990). Identification of bacteria by gas chromatography of cellular fatty acids. *USFCC Newsletter* **20**, 1–6.
- Shimomura, K., Kaji, S. & Hiraishi, A. (2005). *Chryseobacterium shigense* sp. nov., a yellow-pigmented, aerobic bacterium isolated from a lactic acid beverage. *Int J Syst Evol Microbiol* **55**, 1903–1906.
- Thompson, J. D., Gibson, T. J., Plewniak, F., Jeanmougin, F. & Higgins, D. G. (1997). The CLUSTAL_X windows interface: flexible strategies for multiple sequence alignment aided by quality analysis tools. *Nucleic Acids Res* **25**, 4876–4888.
- Vandamme, P., Bernardet, J.-F., Segers, P., Kersters, K. & Holmes, B. (1994). New perspectives in the classification of the flavobacteria: description of *Chryseobacterium* gen. nov., *Bergeyella* gen. nov., and *Empedobacter* nom. rev. *Int J Syst Bacteriol* **44**, 827–831.
- Xu, P., Li, W.-J., Xu, L.-H. & Jiang, C.-L. (2003). A microwave-based method for genomic DNA extraction from Actinomycetes. *Microbiology* **30**, 82–84 (in Chinese).
- Yamaguchi, S. & Yokoe, M. (2000). A novel protein-deamidating enzyme from *Chryseobacterium proteolyticum* sp. nov., a newly isolated bacterium from soil. *Appl Environ Microbiol* **66**, 3337–3343.
- Young, C.-C., Kämpfer, P., Shen, F.-T., Lai, W.-A. & Arun, A. B. (2005). *Chryseobacterium formosense* sp. nov., isolated from the rhizosphere of *Lactuca sativa* L. (garden lettuce). *Int J Syst Evol Microbiol* **55**, 423–426.
- Zhou, Y., Wang, X., Liu, H., Zhang, K.-Y., Zhang, Y.-Q., Lai, R. & Li, W.-J. (2007). *Pontibacter akesuensis* sp. nov., isolated from a desert soil in China. *Int J Syst Evol Microbiol* **57**, 321–325.