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DNA–DNA hybridization study of strains of *Chryseobacterium*, *Elizabethkingia* and *Empedobacter* and of other usually indole-producing non-fermenters of CDC groups IIc, IIe, IIh and IIi, mostly from human clinical sources, and proposals of *Chryseobacterium bernardetii* sp. nov., *Chryseobacterium carnis* sp. nov., *Chryseobacterium lactis* sp. nov., *Chryseobacterium nakagawai* sp. nov. and *Chryseobacterium taklimakanense* comb. nov

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Abstract

The taxonomic classification of 182 phenotypically similar isolates was evaluated using DNA–DNA hybridization and 16S rRNA gene sequence analysis. These bacterial isolates were mainly derived from clinical sources; all were Gram-negative non-fermenters and most were indoleproducing. Phenotypically, they resembled species from the genera *Chryseobacterium*, *Elizabethkingia* or *Empedobacter* or belonged to CDC groups IIc, IIe, IIh and IIi. Based on these analyses, four novel species are described: *Chryseobacterium bernardetii* sp. nov. (type strain NCTC 13530^T=CCUG 60564^T=CDC G229^T), *Chryseobacterium carnis* sp. nov. (type strain NCTC 13525^T=CCUG 60559^T=CDC G81^T), *Chryseobacterium lactis* sp. nov. (type strain NCTC 11390^T=CCUG 60566^T=CDC KC1864^T) and *Chryseobacterium nakagawai* sp. nov. (type strain NCTC 13529^T=CCUG 60563^T=CDC G41^T). The new combination *Chryseobacterium taklimakanense* comb. nov. (type strain NCTC 13490^T=X-65^T=CCTCC AB 208154^T=NRRL B-51322^T) is also proposed to accommodate the reclassified *Planobacterium taklimakanense*.

INTRODUCTION

Phenotypically similar bacteria belonging to the family *Flavobacteriaceae* have long been recognized as genetically diverse. Sottile *et al.* (1973) noted high levels of nucleotide sequence divergence within the species *Flavobacterium meningosepticum* (now *Elizabethkingia meningoseptica*) compared with the type strain of *F. meningosepticum*,

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NCTC 10016^T, and Owen & Snell (1976) showed that the type strain shared on average only 30% of its DNA sequence with most other strains of *F. meningosepticum*, including strains of the same serotype. Another reference strain proved to be highly related to all the other strains, confirming the atypical nature of the type strain. Subsequently, Ursing & Bruun (1987) studied DNA reassociation in 52 strains of *F. meningosepticum* and found two main hybridization groups that were about 40–55% interrelated and comprised four and 48 strains, respectively. The larger group could be divided further into four subgroups if differences in thermal stability of reassociated duplexes were taken into consideration.

Owen & Holmes (1980) studied ten strains that had been identified as *Flavobacterium breve* (now *Empedobacter brevis*) and found that six had high levels of intraspecific nucleotide sequence similarity, whilst the other four contained a high degree of nucleotide sequence divergence. Amongst clinically important members of the genus *Flavobacterium*, as the genus was defined at the time, four natural phenotypic groups were discerned (Holmes & Owen, 1981). Group A comprised saccharolytic, proteolytic, mostly indole-producing strains (now assigned to the genera *Chryseobacterium*, *Elizabethkingia* and *Empedobacter*; see Vandamme *et al.*, 1994). Group B comprised non-saccharolytic, proteolytic, non-indole-producing strains (now assigned to *Myroides*). Group C comprised saccharolytic, non-actively proteolytic, non-indole-producing strains (now assigned to *Sphingobacterium*). Group D was less like *Flavobacterium* and comprised non-saccharolytic, proteolytic, indole-producing strains, but which were not yellow-pigmented and were more susceptible to antimicrobial agents (now assigned to *Bergeyella* and *Weeksella*).

Owen & Holmes (1981) also described a relatively broad range of G+C contents in *Flavobacterium odoratum* (which was reflected in DNA–DNA hybridizations and which later led to a division into *Myroides odoratus* and *Myroides odoratimimus*) and CDC group IIb (which was also reflected in DNA–DNA hybridizations). Subsequently, Ursing & Bruun (1991) investigated DNA reassociation in 42 strains presumptively identified as belonging to *F. breve* or to CDC group IIb. *F. breve* was found to constitute two genomic groups (approx. 45% interrelated) comprising eight and three strains, respectively. Strains of CDC group IIb demonstrated great genomic diversity, with *Flavobacterium gleum* and *Flavobacterium indologenes* (now *Chryseobacterium gleum* and *Chryseobacterium indologenes*) constituting the largest groups, with nine and 11 strains, respectively. The presumably related CDC groups IIc, IIe, IIh and IIi (Weyant *et al.*, 1995) were also usually indole-producing, but remained unnamed. The present study was undertaken to resolve the taxonomic position of strains representing the four latter unnamed groups.

METHODS

Details of the strains examined in the present study are given in Table 1. An additional 15 strains were examined, but are not described in this paper as they could not be assigned to any group.

The methods used for the extraction and purification of DNA from the strains studied and the hydroxyapatite hybridization method for determining levels of DNA relatedness between them have been described previously (Brenner *et al.*, 1982). Since the optimal temperature

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for DNA reassociation is T_m0230 °C (T_m−215 °C for the stringent condition), and members of the family *Flavobacteriaceae* have a lower G+C content than members of the family *Enterobacteriaceae*, the temperatures used in this study were slightly lower than those used by Brenner *et al.* (1982). Reference DNAs were labelled enzymatically *in vitro* with [^{32}P]dCTP by using a nick translation reagent kit (Invitrogen), as directed by the manufacturer. DNA hybridization tests were performed in duplicate at 55 °C and in some cases also at 70 °C. Percentage divergence was calculated to the nearest 0.5%.

For DNA extraction and 16S rRNA gene sequencing, colonies were removed from a culture plate using a 1 µl loop, suspended in 200 µl DMSO and incubated at room temperature for 30 min. The 16S rRNA gene was amplified from the DNA suspension by using the Expand High-Fidelity PCR system (Roche Diagnostics). An aliquot of 2.5 µl DNA suspension was used as template in a 50 µl PCR mix containing 2.5 U polymerase, 1.5 mM $MgCl_2$, 200 µM dNTPs and 400 nM primers fD1 and rP2 in order to amplify the gene from nucleotide positions 8 to 1492 of the *Escherichia coli* 16S rRNA gene (GenBank accession no. J01695). The PCR primer sequences are from Weisburg *et al.* (1991).

Amplification was performed on an ABI 9700 thermocycler (Applied Biosystems) using a program of 94 °C for 5 min followed by 35 cycles of 94 °C for 15 s, 50 °C for 15 s and 72 °C for 90 s, with a final single extension of 72 °C for 5 min, and then held at 4 °C. Amplified products were examined by electrophoresis of 5 µl of each reaction on a 1.2% agarose e-gel (Invitrogen) for 30 min at 85 V. Excess nucleotides and primers were removed using a QIAquick PCR Purification kit (Qiagen). The purified PCR product was used as a template in 20 µl cycle sequencing reactions with Big Dye version 3.1 (Applied Biosystems). Sixteen sequencing primers were used, as listed in Table S1 (available in IJSEM Online). Primer sequences beginning with BSF or BSR are from the European rRNA database (http://bioinformatics.psb.ugent.be/webtools/rRNA/), primer sequences named R357 and R519 are from Stackebrandt & Charfreitag (1990) and fD1 is from Weisburg *et al.* (1991), F357, F530, and R530 are from Sacchi *et al.* (2002), and fD1-5p, F785, R802, and rP2-5p are from Morey *et al.* (2006). Sequencing reaction products were purified with Centri-Sep plates (Princeton Separations). Reactions were electrophoresed on an ABI 3130 or 3730 system using POP-7 polymer (Applied Biosystems). Chromatograms were assembled and analysed in Seqmerge (Wisconsin package version 10.3; Accelrys).

Phylogenetic and molecular evolutionary analyses were conducted using BioEdit (Hall, 1999) and CLUSTAL X2 (Larkin *et al.*, 2007). Alignments were corrected manually where necessary, and end-trimming was performed so that percentage identities between 16S rRNA gene sequences were calculated over the range shared by both sequences. Trees were visualized using MEGA version 4 (Tamura *et al.*, 2007).

All strains were characterized biochemically in all or most of a range of 68 conventional biochemical tests by methods described previously (Holmes *et al.*, 1975).

RESULTS AND DISCUSSION

The DNA–DNA hybridization results are presented in Tables 2, 3, 4, 5 and 6, and published names of the various genomic groups are summarized in Table 7. Strains representing

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species of the genera *Empedobacter*, *Elizabethkingia* and *Sphingobacterium* will be discussed first, roughly in the order in which they appear in Tables 2, 3, 4, 5 and 6, followed by strains representing known species of the genera *Chryseobacterium* and *Flavobacterium*. Novel *Chryseobacterium* species will then be discussed in alphabetical order.

Fourteen strains could be ascribed to *Empedobacter brevis*, as they showed high levels of DNA–DNA relatedness (69–82%) to the type strain (Table 2). These included the six strains studied by Owen & Holmes (1980) found to have high levels of intraspecific nucleotide sequence similarity (>70%). Four other strains from that study, which were identified biochemically as *Empedobacter brevis*, proved only 35–42% related, and these were distributed over different groups in the present study; ATCC 14234 and CL 94/78 together formed the 92 group (Table 5), NCTC 11163 was a member of the large 58 group (Table 3), whilst CL93/78 (data not shown) remained one of the 15 lone strains.

The 16S rRNA gene sequence of ATCC 14234, representing the two strains of the 92 group (Table 5), was >99.9% similar to that of *Empedobacter brevis* LMG 4011^{T} (GenBank accession no. AM177497). Stackebrandt & Ebers (2006) proposed that a 16S rRNA gene sequence similarity above 98.7–99% should be mandatory for testing the genomic uniqueness of a novel isolate. Ursing & Bruun (1991) found that DNA–DNA hybridization of ATCC 14234 showed a reassociation of >70% with the type strain of *Empedobacter brevis* (NCTC 11099^{T}), but Owen & Holmes (1980) and the present study (Table 2) found only 35–48 and 34–45% relatedness, respectively, to the type strain of *Empedobacter brevis*. No direct comparison of the isolates used by each group has been made, so none of the possible reasons for this discrepancy (such as differences in technique or a mix-up of strains) have been ruled out. Although indistinguishable in biochemical tests and in 16S rRNA gene sequence similarity, strains in this group may represent separate genomospecies. Strain 92 (=ATCC 14234) has been preserved as NCTC 13469 to represent '*Empedobacter brevis*-like species 1'.

Elizabethkingia meningoseptica is a complex of several genomic groups (Sottile *et al.*, 1973; Owen & Snell, 1976; Ursing & Bruun, 1987). Many cultures from these previous studies were included in the present study, and the DNA–DNA hybridization results (Table 4) support these earlier findings. Only two strains in this study were found to show high levels of DNA–DNA relatedness (82–87%) to the type strain (Table 5), confirming the findings of Ursing & Bruun (1987). In view of the rarity of such strains, strain 256 has been preserved as NCTC 13393. In this study, the 245 group comprised five strains and corresponded to Ursing and Bruun group II: 1 (Table 4), the 251 group comprised three strains and corresponded to Ursing and Bruun group II: 2 (Table 4), the 255 group comprised six strains and corresponded to Ursing and Bruun group II: 3 (Table 4), whilst the 259 group comprised three strains and corresponded to Ursing and Bruun group II: 4 (Table 6). Many strains in the present study were included in that of Ursing & Bruun (1987), and all were assigned to the same groups they described. DNA–DNA hybridization levels above 70% between groups were seen (Table 4) between the 251 and 255 groups, 251 and 259 groups and 255 and 259 groups, but with a divergence in related sequences ≥4.0%. No characters to differentiate these genomic groups have yet been found, so they remain assigned for the time being to *Elizabethkingia meningoseptica*.

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The 16S rRNA gene sequence of CDC E8371, representing the two strains of the 231 group (Table 4), was 99.9% identical to that of *Sphingobacterium daejeonense* TR6-04^T (GenBank accession no. AB249372), suggesting that the 231 group can be assigned to this species, which was confirmed by a DNA–DNA relatedness value of 80% between the two (Table 8). The species was originally described to accommodate a single strain isolated from compost, but the members of this taxon identified in this study were both from human clinical sources. Both were also representatives of CDC group IIi, so the assignment of these two strains to this genus would be unexpected. Given the paucity of isolates of this species and the fact that the isolates identified in this study are the first to be recognized from clinical sources, CDC E8371 has been preserved as NCTC 13534 and CL714/92 as NCTC 13522.

The 16S rRNA gene sequence of CDC F859, representing the two strains of the 217 group (Table 4), was 99.8% identical to that of *Sphingobacterium lactis* DSM 22361^T (GenBank accession no. FN908501). These two strains were phenotypically similar to the strain description of *S. lactis* (Schmidt *et al.*, 2012), except that they grew on MacConkey agar and did not produce DNase. Strain CDC F859 had a DNA G + C content of 39.5 mol%, as opposed to the 44.2 mol% reported in the species description. Despite these phenotypic variations, CDC F859 is presumed to represent *S. lactis*, and has been preserved as NCTC 135265CCUG 60560.

Fifteen strains could be ascribed to *Chryseobacterium indologenes*, as they showed high levels of DNA–DNA relatedness (80–96%) to its type strain (Table 2). A single strain of *Chryseobacterium gleum* was identified (CL424/73, which showed 80% DNA–DNA relatedness to the type strain of *C. gleum*; Table 2) and can be added to the 12 identified in the original description of the species (Holmes *et al.*, 1984) and to the additional four found by Ursing & Bruun (1991). No additional strains of *Chryseobacterium balustinum* were found in this study to show high levels of DNA–DNA relatedness to the type strain, but two more strains of *Chryseobacterium indoltheticum*, from environmental sources, were identified (Table 2), as they showed 76–82% DNA–DNA relatedness to the type strain. Given the paucity of isolates of this species, CL743/78 has been preserved as NCTC 13532.

The 16S rRNA gene sequence of CL311/80, representing the 12 strains of the 93 group (Table 3), was 99.3% identical to that of *Chryseobacterium anthropi* NF 1366^T (GenBank accession no. AM982786), suggesting that the 93 group can be assigned to this species. This species assignment was confirmed by the fact that Kämpfer *et al.* (2009b), when describing the novel species, included in their study CDC F4391, which was also included in the present study. The species was originally described to accommodate eight strains of human clinical origin. Most of the strains identified in this study were similarly from human clinical specimens, but two were from a veterinary source and one from chiller water associated with poultry processing. CL311/80 has been preserved as NCTC 135285CCUG 60562.

The 16S rRNA gene sequence of CDC F5649, representing the 19 strains of the 224 group (Table 3), was 99.9% identical to that of *Chryseobacterium hominis* NF802^T (GenBank accession no. AM261868), suggesting that the 224 group can be assigned to this species. This species assignment was confirmed by a DNA–DNA relatedness value of 96% between the two (Table 9). The species was originally described to accommodate clinical isolates

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biochemically similar to CDC groups IIc and IIh. The members of this taxon identified in this study included several representatives of CDC groups IIc and IIh and were all from human clinical sources.

The 16S rRNA gene sequence of CL712/92, representing the four strains of the 212 group (Table 3), was 99.7% identical to that of *Planobacterium taklimakanense* X-65^T (GenBank accession no. EU718058) and 95.4% identical to that of *Chryseobacterium haifense* H38^T (EF204450), suggesting that the 212 group might have affinities to these species. *P. taklimakanense* was originally described to accommodate a single strain from desert soil (Peng *et al.*, 2009), and *C. haifense* was originally described as a psychrotolerant bacterium isolated from raw milk (Hantsis-Zacharov & Halpern, 2007), whereas the strains comprising the 212 group contained representatives of CDC groups IIc and IIe and all were from human clinical sources. However, any possible close relationship of the 212 group to *C. haifense* was not confirmed by DNA–DNA relatedness, with a value of only 18% between the two (Table 9). The four strains of the 212 group appear to correspond to *P. taklimakanense*, differing from the species description only in that they were non-motile in broth. CL712/92 has been preserved as NCTC 135275CCUG 60561. Fig. 1 includes species of the genus *Chryseobacterium* published after the initial description of *P. taklimakanense*, which clearly falls in the *Chryseobacterium* clade. There is no longer any justification for *Planobacterium* as a separate genus, and the new combination *Chryseobacterium taklimakanense* comb. nov. is formally proposed.

The 16S rRNA gene sequence of A86/68, representing the two strains of the 123 group (Table 6), was 99.3% identical to that of *Chryseobacterium ureilyticum* F-Fue-04IIIaaaa^T (GenBank accession no. AM232806) and 98.4% identical to that of *Chryseobacterium joostei* LMG 18212^T (AJ271010), suggesting that the 123 group can be assigned to the former species. Any possible close relationship to *C. joostei* was not confirmed by DNA–DNA relatedness, with a value of only 55% between the two (Table 9). *C. ureilyticum* was originally described to accommodate a single strain associated with a beer-bottling plant, whilst *C. joostei* was originally described to accommodate strains isolated from the dairy environment. The two strains identified in this study, however, were from human clinical specimens. A86/68 has been preserved as NCTC 135245CCUG 60558.

The 16S rRNA gene sequence of A104/68, representing the two strains of the 125 group (Table 6), was 98.9% identical to that of *Chryseobacterium shigense* GUM-Kaji^T (GenBank accession no. AB193101), suggesting that the 125 group can be assigned to this species, which was confirmed by a DNA–DNA relatedness value of 77% between the two (Table 9). The species was originally described to accommodate a single strain isolated from a lactic acid beverage, and the members of this taxon identified in this study were both from milk swabs. Given the paucity of isolates of this species, A104/68 has been preserved as NCTC 13533.

The 16S rRNA gene sequence of CL479/77, representing the three strains of the 71 group, was 98.6% identical to that of *Chryseobacterium gleum* CCUG 14555^T (GenBank accession no. AM232812). Although suggesting that the 71 group might have some affinity to this species, DNA–DNA hybridization results from the present study revealed only 43–65%

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relatedness (with a divergence in related sequences >9.0%; Table 2) to the type strain of *C. gleum*. Thus, despite showing high 16S rRNA gene sequence similarity, these three strains clearly form a distinct and homogeneous taxon.

The 16S rRNA gene sequence of CL88/78, representing the two strains of the 95 group, was 98.4% identical to that of *Chryseobacterium* (*Sejongia*) *jeonii* AT1047^T (GenBank accession no. AY553294), suggesting that the 95 group might have an affinity to this species. However, any possible close relationship was not confirmed by DNA–DNA relatedness, with a value of only 20% between the two (Table 8). The species was originally described to accommodate a single strain isolated from an Antarctic terrestrial sample; both members of the 95 group were from meat.

The 16S rRNA gene sequence of CL636/74, representing the two strains of the 137 group (Table 6), was 99.4% identical to that of *Flavobacterium lindanitolerans* IP-10^T (GenBank accession no. EF424395), suggesting that the 137 group can be assigned to this species. The species was originally described to accommodate a single strain from soil, whereas both strains identified in this study were from human clinical specimens. CL636/74 has been preserved as NCTC 13531=CCUG 60565.

The 16S rRNA gene sequence of strains representing the remaining four groups (58 group, 48 strains; 142 group, six strains; 63 group, two strains; and 78 group, two strains) did not show high levels of similarity to the type strains of any other named taxa in the GenBank database, suggesting they represent hitherto unnamed taxa. However, the 16S rRNA gene sequence of NCTC 11310, representing the 48 strains of the 58 group, was >99.9% similar to a strain of *Wautersiella falsenii* genomovar 2 (GenBank accession no. AM238678), suggesting that the 58 group might correspond to this hitherto-unnamed but phenotypically indistinguishable genomic species. Since *W. falsenii* was described to accommodate clinical isolates phenotypically resembling members of the genera *Chryseobacterium* and *Empedobacter* and CDC group IIh, and given that several of the strains of the 58 group were deposited as reference strains of *F. meningosepticum* serotypes (see, for example, Richard *et al.*, 1979) whilst one was of CDC group IIh, such a relationship was perfectly feasible. The synonymy of the 58 group with *W. falsenii* was confirmed by DNA–DNA relatedness values of ≥85% between the 58 group and the two biovars (Table 8).

Overall, five groups (71 group, three strains; 95 group, two strains; 142 group, six strains; 63 group, two strains; and 78 group, two strains) each constituted novel genomospecies of the genus *Chryseobacterium*. To determine their phylogenetic positions, a phylogenetic tree of all species of *Chryseobacterium* was reconstructed (Fig. S1). Although the 16S rRNA gene sequence of CL479/77, representing the three strains of the 71 group, fell in the same clade as that of the type strain of *C. gleum*, DNA–DNA hybridization results did not confirm these strains as members of *C. gleum*. This group is therefore clearly a candidate novel species, but no phenotypic differences from *C. gleum* could be found, so it is not appropriate to propose the 71 group as a novel species at this time. Strain 71 (CL479/77) has been preserved as NCTC 13470 to represent '*C. gleum*-like species 1'. The type strains of the nearest neighbours of each of the remaining four groups were then characterized to identify characteristics that might differentiate each from its nearest neighbours. These differential

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characteristics are displayed in Tables 10 and 11. On the basis of the 16S rRNA gene sequence of strains and the differential phenotypic characters, four novel species are proposed.

Although strains of CDC groups IIc, IIe, IIh and IIi were so assigned on the basis of phenotypic characters, there was little correlation with genomic data, as members of each group belonged to at least two different DNA–DNA hybridization groups as defined in this study (three in the case of CDC group IIi), or proved to be single isolates. The 93 group, however, was composed almost entirely of strains of or resembling CDC group IIe. The disposition of the various groups identified in this study and their correlation to the CDC phenotypic groups are summarized in Table 7. Fifteen strains (including CL93/78) were the sole representative of their groups, and it is likely that some of these represent novel taxa, but only those groups containing at least two representatives are described here.

The novel species *Chryseobacterium bernardetii* and *Chryseobacterium nakagawai* were both derived from clinical samples and share a set of near-neighbours that includes *C. indologenes* (Fig. 2), which is a known human pathogen (Chen *et al.*, 2013). The possibility that this cluster of species of *Chryseobacterium* shares virulence characteristics remains to be investigated.

Description of *Chryseobacterium taklimakanense* comb. nov

Chryseobacterium taklimakanense (tak.li.ma.kan.en′se. N.L. neut. adj. *taklimakanense* pertaining to the desert of Taklimakan,Xinjiang,China,wherethetypestrainwasisolated).

Basonym: Planobacterium taklimakanense Peng et al. 2009.

The description is that of Peng *et al.* (2009). The type strain is X-65^{T}=CCTCC AB 208154^{T}=NRRL B-51322^{T}=NCTC 13490^{T}.

Description of *Chryseobacterium bernardetii* sp. nov

Chryseobacterium bernardetii (ber.nar.de′ti.i. N.L. masc. gen. n. *bernardetii* of Bernardet, named after Jean-François Bernardet, a French microbiologist long associated with this group of organisms).

Described in this study based on six strains comprising the 142 group. Cells are Gram-negative. Colonies are circular, convex, entire, opaque, shiny, smooth and yellow-pigmented. Positive for acid production (in ammonium salt medium) from glucose, arabinose, fructose, glycerol, maltose, sucrose and trehalose, aesculin hydrolysis, casein digestion, catalase production, cytochrome oxidase production, gelatinase production (stab method), growth at 37 °C, at room temperature (18–22 °C), on MacConkey agar and on β-hydroxybutyrate, hydrolysis of tyrosine, oxidative metabolism in Hugh and Leifson O-F test (one strain positive only after incubation for more than 5 days) and production of brown melanin-like pigment on tyrosine agar. Negative for acid production (in ammonium salt medium) from adonitol, cellobiose, dulcitol, ethanol, inositol, lactose, mannitol, raffinose, rhamnose, salicin and sorbitol, acid and gas production from glucose in peptone water medium, acid from 10% (w/v) lactose, arginine dihydrolase production, fluorescence on

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King's B medium, gluconate oxidation, growth at 5 °C and on cetrimide agar, H_2S production (by both lead acetate paper and triple-sugar iron agar methods), KCN tolerance, lipid inclusions after growth on β-hydroxybutyrate, lysine decarboxylase production, malonate utilization, motility (hanging drop preparation at both 37 °C and room temperature), nitrate reduction, ornithine decarboxylase production, phenylalanine deamination, reduction of 0.4% (w/v) selenite, utilization of citrate (Simmons' medium), β-galactosidase production (ONPG test) and 3-ketolactose production. The six strains studied differ in the following tests (result in parentheses for the type strain): acid production (in ammonium salt medium) from xylose (+), acid from 10% (w/v) glucose (+), gelatinase production (plate method; +), growth at 42 °C (+), hydrolysis of Tween 20 (+), Tween 80 (−) and starch (−), lecithinase production (−), nitrite reduction (+), production of extracellular DNase (+), urease production (−) and utilization of citrate (Christensen's medium; +). Phenotypic characteristics (between one and four in number) useful for the differentiation of *C. bernardetii* (142 group) from its nearest neighbours are shown in Table 10. Only the inability to grow at 5 °C distinguishes *C. bernardetii* from *C. joostei*.

The G+C content of the type strain is 37.0 mol%. The type strain is NCTC 13530^T=CCUG 60564^T=CL318/82^T=CDC G229^T; it was isolated from sputum in Doncaster, UK.

Description of *Chryseobacterium carnis* sp. nov

Chryseobacterium carnis (car′nis. L. gen. n. *carnis* of flesh).

Described in this study based on two strains comprising the 95 group. Cells are Gram-negative. Colonies are circular, convex, entire, opaque, shiny, smooth and yellow-pigmented. Positive for acid production (in ammonium salt medium) from glucose and maltose, casein digestion, catalase production, cytochrome oxidase production, gelatinase production (plate and stab methods), growth at 37 °C, at room temperature (18–22 °C), on β-hydroxybutyrate and on MacConkey agar, hydrolysis of starch and Tween 20 and production of extracellular DNase. Negative for acid production (in ammonium salt medium) from adonitol, arabinose, cellobiose, dulcitol, ethanol, fructose, glycerol, inositol, lactose, mannitol, raffinose, rhamnose, salicin, sorbitol, sucrose, trehalose and xylose, acid from 10% (w/v) glucose and from 10% (w/v) lactose, aesculin hydrolysis, arginine dihydrolase production, fluorescence on King's B medium, gas production from glucose in peptone water medium, gluconate oxidation, growth at 42 °C and on cetrimide agar, Hugh and Leifson O-F test, hydrolysis of Tween 80 and tyrosine, H_2S production (by both lead acetate paper and triple-sugar iron agar methods), lecithinase production, lipid inclusions after growth on β-hydroxybutyrate, lysine decarboxylase production, malonate utilization, motility (hanging drop preparation at both 37 °C and room temperature), nitrate reduction, nitrite reduction, ornithine decarboxylase production, phenylalanine deamination, production of brown melanin-like pigment on tyrosine agar, reduction of 0.4% (w/v) selenite, urease production, utilization of citrate (Christensen's and Simmons' media), β-galactosidase production (ONPG test) and 3-ketolactose production. The two strains studied differ in the following tests (type strain positive): acid production from glucose in peptone water medium, growth at 5 °C and KCN tolerance. Phenotypic characteristics (between three and 10 in number) useful for the differentiation of *C. carnis* (95 group) from its nearest

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neighbours, several of which were originally described as belonging to the genus *Sejongia*, are shown in Table 11. The assertion that species of the genus *Sejongia* be transferred to the genus *Chryseobacterium* (Kämpfer *et al.*, 2009a) was supported (Fig. 3), so this novel species is assigned to the genus *Chryseobacterium*.

The G + C content of the type strain is 34.0 mol%. The type strain is NCTC 13525^T=CCUG 60559^T=CL88/78^T=Hayes B19/1^T=CDC G81^T; it was isolated from beef.

Description of *Chryseobacterium lactis* sp. nov

Chryseobacterium lactis (lac′tis. L. gen. n. *lactis* of/from milk).

Described in this study based on two strains comprising the 63 group. Cells are Gram-negative. Colonies are circular, convex, entire, opaque, shiny, smooth and yellow-pigmented. Positive for acid production (in ammonium salt medium) from glucose, fructose, glycerol, maltose and trehalose, aesculin hydrolysis, casein digestion, catalase production, cytochrome oxidase production, gelatinase production (plate and stab methods), growth at 37 °C, at room temperature (18–22 °C), on *β*-hydroxybutyrate and on MacConkey agar, hydrolysis of Tweens 20 and 80, oxidative metabolism in the Hugh and Leifson O-F test, production of brown melanin-like pigment on tyrosine agar and production of extracellular DNase. Negative for acid production (in ammonium salt medium) from adonitol, arabinose, cellobiose, dulcitol, ethanol, inositol, lactose, mannitol, raffinose, rhamnose, salicin, sorbitol, sucrose and xylose, acid and gas production from glucose in peptone water medium, acid from 10% (w/v) glucose and from 10% (w/v) lactose, arginine dihydrolase production, fluorescence on King's B medium, gluconate oxidation, growth at 42 °C and on cetrimide agar, hydrolysis of starch, H_2S production (by both lead acetate paper and triple-sugar iron agar methods), KCN tolerance, lipid inclusions after growth on *β*-hydroxybutyrate, lysine decarboxylase production, malonate utilization, motility (hanging drop preparation at both 37 °C and room temperature), nitrate reduction, nitrite reduction, ornithine decarboxylase production, phenylalanine deamination, reduction of 0.4% (w/v) selenite, urease production, utilization of citrate (Christensen's and Simmons' media), *β*-galactosidase production (ONPG test) and 3-ketolactose production. The two strains studied differ in the following tests (type strain positive): growth at 5 °C, hydrolysis of tyrosine and lecithinase production. Phenotypic characteristics (between two and five in number) useful for the differentiation of *C. lactis* (63 group) from its nearest neighbours are shown in Table 10.

The G + C content of the type strain is 34.5 mol%. The type strain is NCTC 11390^T=CCUG 60566^T=A140/68^T=F68^T=CDC KC1864^T; it was isolated from a milk bottle rinse on a farm in Paisley, Scotland, UK.

Description of *Chryseobacterium nakagawai* sp. nov

Chryseobacterium nakagawai (na.ka.ga′wa.i. N.L. masc. gen. n. *nakagawai* of Nakagawa, named after Yasuyoshi Nakagawa, a Japanese microbiologist long associated with this group of organisms).

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Described in this study based on two strains comprising the 78 group. Cells are Gram-negative. Colonies are circular, convex, entire, opaque, shiny, smooth and yellow-pigmented. Positive for acid production (in ammonium salt medium) from glucose, maltose and trehalose, casein digestion, catalase production, cytochrome oxidase production, gelatinase production (plate and stab methods), growth at 37 °C, at room temperature (18–22 °C) and on β-hydroxybutyrate, hydrolysis of starch, Tween 20 and tyrosine, lecithinase production, oxidative metabolism in Hugh and Leifson O-F test, production of brown melanin-like pigment on tyrosine agar and production of extracellular DNase. Negative for acid production (in ammonium salt medium) from adonitol, arabinose, cellobiose, dulcitol, ethanol, fructose, inositol, lactose, mannitol, raffinose, rhamnose, salicin, sorbitol, sucrose and xylose, acid and gas production from glucose in peptone water medium, acid from 10% (w/v) glucose and 10% (w/v) lactose, arginine dihydrolase production, fluorescence on King's B medium, gluconate oxidation, growth at 5 and 42 °C and on MacConkey agar, H_2S production (by both lead acetate paper and triple-sugar iron agar methods), KCN tolerance, lipid inclusions after growth on β-hydroxybutyrate, lysine decarboxylase production, malonate utilization, motility (hanging drop preparation at both 37 °C and room temperature), nitrate reduction, nitrite reduction, ornithine decarboxylase production, phenylalanine deamination, reduction of 0.4% (w/v) selenite, urease production, utilization of citrate (Simmons' medium), β-galactosidase production (ONPG test) and 3-ketolactose production. The two strains studied differ in the following tests (type strain positive): acid production (in ammonium salt medium) from glycerol, aesculin hydrolysis, growth on cetrimide agar, hydrolysis of Tween 80 and utilization of citrate (Christensen's medium). Phenotypic characteristics (between three and five in number) useful for the differentiation of *C. nakagawai* (78 group) from its nearest neighbours are shown in Table 10.

The G + C content of the type strain is 35.0 mol%. The type strain is NCTC 13529^T=CCUG 60563^T=F91^T=CDC G41^T; it was isolated from a kidney abscess in Gloucester, UK.

Supplementary Material

Refer to Web version on PubMed Central for supplementary material.

Acknowledgments

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The GenBank/EMBL/DDBJ accession numbers for the 16S rRNA gene sequences of *C. anthropi* NCTC 13528, *C. bernardetii* sp. nov. NCTC 13530^T, *C. carnis* sp. nov. NCTC 13525^T, *C. gleum*-like species 1 strain NCTC 13470, *C. haifense* NCTC 13466^T, *C. hominis* CDC F5649, *C. lactis* sp. nov. NCTC 11390^T, *C. nakagawai* sp. nov. NCTC 13529^T, *C. shigense* strains NCTC 13533 and NCTC 13458^T, *C. taklimakanense* comb. nov. NCTC 13527, *C. ureilyticum* NCTC 13524, *F. granuli* NCTC 13460^T, *F. lindanitolerans* NCTC 13531, *S. daejeonense* strains NCTC 13534 and NCTC 13455^T, *S. lactis* NCTC 13526^T and *W. falsenii* NCTC 11310 are respectively JX100815–JX100832.

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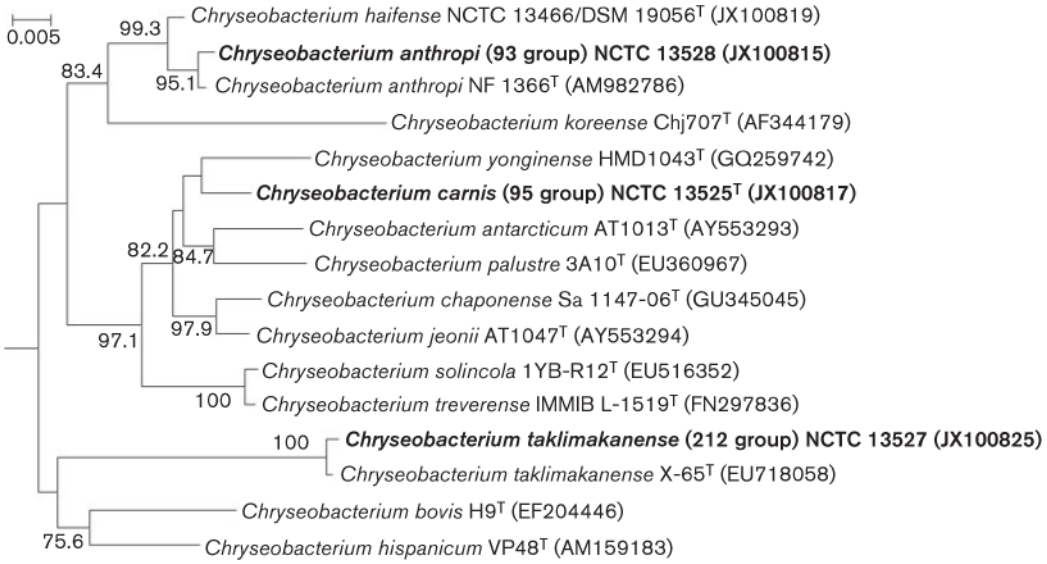

Fig. 1.

Neighbour-joining phylogenetic tree showing the relationships of 16S rRNA gene sequences from the type strains of *Chryseobacterium bernardetii* sp. nov., *Chryseobacterium carnis* sp. nov., *Chryseobacterium lactis* sp. nov. and *Chryseobacterium nakagawai* sp. nov. with sequences of the type strains of all other species of the genus *Chryseobacterium*. The tree is rooted, with *Weeksella virosa* ATCC 43766^T as the outgroup (not shown; GenBank accession no. M93152). GenBank accession numbers are given in parentheses. Bootstrap support from 1000 resamplings at nodes is displayed as percentages. Bar, 0.005 substitutions per nucleotide position. Strains described in this study are highlighted in bold. Full-length sequences were not available for all strains, so the alignment was trimmed to the 1336 bp for which data were available for all strains. The full tree is available as Fig. S1. Shown here is the node of the tree that supports the assertion that the species named *Planobacterium taklimakanense* is actually a species of *Chryseobacterium*.

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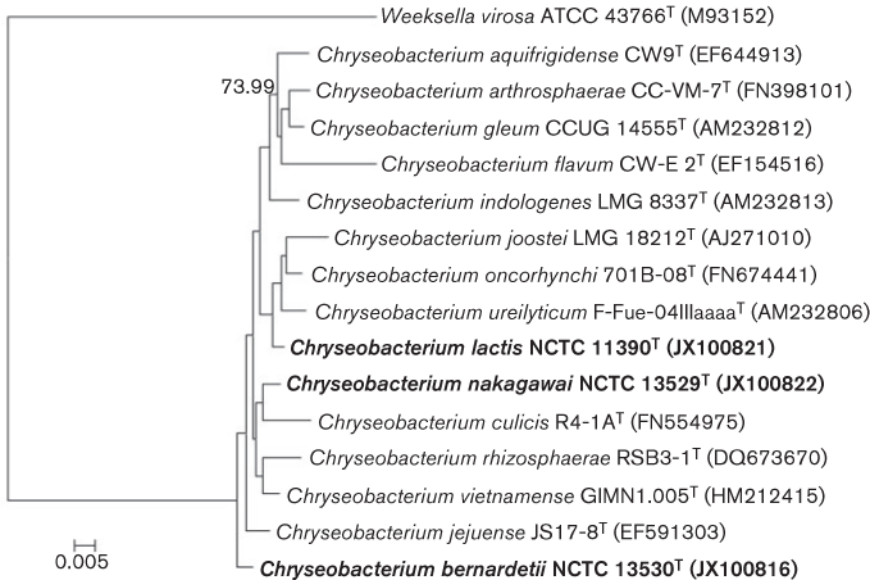

Fig. 2.

Three of the newly identified *Chryseobacterium* species, which were derived from clinical specimens, belong to a cluster of species that includes the known human pathogen *C. indologenes*. The rooted tree was generated using the same parameters as described for Fig. 1, but an additional 111 bp was available for all of the selected strains, and was included in the alignment. The tree included all 16S rRNA gene sequence data that were available for all of the selected strains.

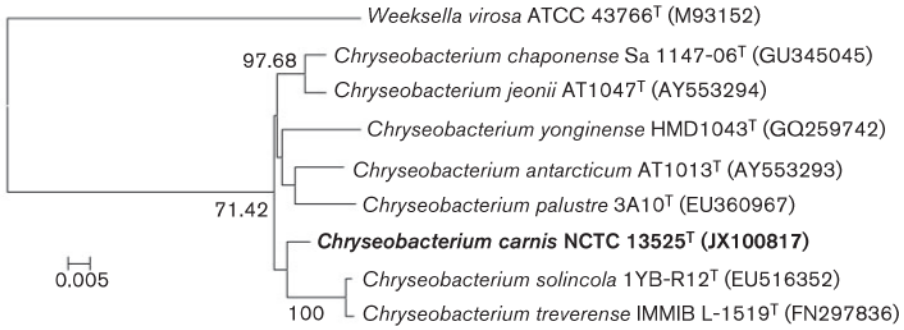

Fig. 3.

This analysis supports the suggestion that species of the genus *Sejongia* be transferred to the genus *Chryseobacterium*. As with Fig. 2, this analysis was done using the parameters described in Fig. 1.

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Table 1
Designation and source of strains studied

Culture numbers prefixed by the letters A, F or CL are strains received for identification at the NCTC. NCTC, National Collection of Type Cultures, Health Protection Agency, Colindale, London, UK;

Strain	Other strain designation(s)	Source
Chryseobacterium indologenes (16 strains)		
50^T	NCTC 10796^T=CDC KC1854^T=CDC 3716^T	Trachea at autopsy; unknown
65	CL361/70=F142=CDC F9967	Respirator; London, UK
67	CL471/75=CDC F9969	Nasal swab; Dublin, Ireland
68	CL45/78=CDC F9970	Urine; London, UK
70	CL44/78=CDC F9972	Catheter urine; London, UK
74	CL42/78=NCTC 11409=CDC F9976	Water; London, UK
75	CL514/78=CDC G38	Urine; Swansea, Wales, UK
76	CL145/70=F139=CDC G39	Blood culture; London, UK
77	CL46/78=CDC G40	Peritoneal dialysis fluid; London, UK
80	CL43/78=CDC G43	Catheter urine; London, UK
116	CL9/77=CDC G161	Urine; Liverpool, UK
124	F132=A57/70=CDC G187	Hospital kitchen surface; Northampton, UK
141	CL223/85=CDC G228	Oral ulcer; Coventry, UK
144	CL20/81=CDC G231	Blood; London, UK
145	CL187/82=CDC G232	Human clinical; Birmingham, UK
157	CL19/81=CDC G253	Blood; London, UK
Chryseobacterium gleum (2)		
51^T	NCTC 11432^T=F93^T=CDC KC1855^T	High vaginal swab; London, UK
59	CL424/73=CDC F9935	Blood; Dar es Salaam, Tanzania
Chryseobacterium balustinum (1)		
62^T	NCTC 11212^T=ATCC 33487^T=CDC KC1863^T	Heart blood of fish; River Dordogne, France
Chryseobacterium indoltheticum (3)		
106^T	CL252/80^T=ATCC 27950^T=CDC G141^T	Marine mud; UK
110	CL743/78=F32=A78/68=CDC G145	Milk sample; Paisley, Scotland, UK
140	CL97/78=Hayes S10/1=CDC G211	Soil; unknown
Empedobacter brevis (15)		
55^T	NCTC 11099^T=CL88/76^T=CDC KC1859^T	Human bronchial secretion; Zurich, Switzerland
54	NCTC 11162=CL626/75=CDC KC1858	Eye swab, human; Dublin, Ireland
81	CL666/76=CDC G59	Urine, human; Bratislava, Slovak Republic
83	CL42/79=CDC G61	Snake; London, UK
87	CL478/77=CDC G65	Post-mortem lung, human; London, UK
91	CL200/75=CDC G69	Human, unknown; London, UK
129	F149=A93/72=CL14/79=CDC G198	Presacral abscess; London, UK
159	CL309/80=CDC G348	Urine; Paris, France
160	CL277/81=CDC G349	Vagina; Strasbourg, France
161	CL476/81=Richard 6.81=CDC G350	Vagina; Strasbourg, France

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Strain	Other strain designation(s)	Source
168	CL40/85=CDC G361	Umbilical artery catheter, neonate; London, UK
183	CL167/82=Richard 19.81=CDC G376	CSF; Strasbourg, France
190	CL624/80=CDC G383	Nasal swab, tortoise; London, UK
191	CL297/80=CDC G384	Prairie marmot; London, UK
211	CDC F9036	CSF; USA
58 group (48)		
58	NCTC 11310=CIP 79.30=CDC KC1862	Urine; Strasbourg, France; *F. meningosepticum* serotype L
53	NCTC 11307=CIP 79.25=CDC KC1857	Blood culture; Strasbourg, France; *F. meningosepticum* serotype I
56	NCTC 11163=CL669/76=CDC KC1860	Urine, human; Bratislava, Slovak Republic
57	NCTC 11308=CIP 78.68=CDC KC1861	Skin swab; Strasbourg, France; *F. meningosepticum* serotype J
82	CL8/74=CDC G60	Wound; Wrexham, Wales, UK
84	CL444/73=CDC G62	Eye swab; Birmingham, UK
85	CL452/80=CDC G63	Swan faeces; London, UK
88	CL7/74=CDC G66	Eye; Wrexham, Wales, UK
89	CL53/80=CDC G67	Wound; Alsace, France
94	CL623/77=CDC G80	Blood culture; Worthing, UK
96	CL336/80=CDC G82	Potoroo; London, UK
99	CL540/78=CDC G99	Urine; Germany
162	CL477/81=Richard 3.81=CDC G351	Urine; Strasbourg, France
164	CL480/81=Richard 13.81=CDC G353	Urine; Strasbourg, France
165	CL481/81=Richard 14.81=CDC G354	Urine; Strasbourg, France
166	CL482/81=Richard 17.81=CDC G355	Urine; Strasbourg, France
167	CL483/81=Richard 18.81=CDC G356	Humidifier; Strasbourg, France
169	CL208/84=CCUG 12570=CDC G362	Unknown
170	CL113/83=Richard 5.82=CDC G363	Throat; Strasbourg, France
171	CL36/83=Richard 50.82=CDC G364	Urine; Strasbourg, France
172	CL35/83=Richard 49.82=CDC G365	Urine; Strasbourg, France; *F. meningosepticum* serotype N
173	CL 34/83=Richard 9.82=CDC G366	Vagina; Strasbourg, France; *F. meningosepticum* serotype N
174	CL31/83=Richard 4.82=CDC G367	Urine; Strasbourg, France
175	CL30/83=Richard 7.82=CDC G368	Urine; Strasbourg, France
176	CL29/83=Richard 3.82=CDC G369	Urine; Strasbourg, France
177	CL28/83=Richard 2.82=CDC G370	Urine; Strasbourg, France
178	CL300/82=Richard 17.82=CDC G371	Catheter; Grenoble, France
179	CL299/82=Richard 16.82=CDC G372	Catheter; Grenoble, France
180	CL172/82=Richard 1.82=CDC G373	Vagina; Bordeaux, France
181	CL170/82=Richard 24.81=CDC G374	Urine; Strasbourg, France
182	CL168/82=Richard 20.81=CDC G375	Indwelling catheter; Grenoble, France
184	CL67/82=Richard 24.81=CDC G377	Urine; Strasbourg, France
185	CL66/82=Richard 23.81=CDC G378	Urine; Strasbourg, France

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Strain	Other strain designation(s)	Source
186	CL64/82=Richard 21.81=CDC G379	Urine; Strasbourg, France
187	CL61/82=Richard 18.81=CDC G380	Blood; Grenoble, France
188	CL55/82=Richard 3.81=CDC G381	Wound; Leuven, Belgium
189	CL54/82=Richard 2.81=CDC G382	Sputum; Strasbourg, France
192	CL63/82=Richard 20.81=CDC G390	Vagina; Strasbourg, France
193	CL65/82=Richard 22.81=CDC G391	Urine; Strasbourg, France
194	CL171/82=Richard 26.81=CDC G392	Urine; Strasbourg, France
195	CL294/82=Richard 1.82=CDC G393	Vaginal discharge; Annecy, France
196	CL60/82=Richard 14.81=CDC G395	Urine; Strasbourg, France
199	CDC F6535	Vaginal discharge; USA
200	CDC F6236	Cat bite; USA
202	CDC F2617	Vagina; USA
205	CDC F4121	Blood; USA
216 CDC group IIh	CL715/92=CDC E8860	Wound; Alaska, USA
243	CDC G2308	Blood; Oxford, UK
93 group (12)		
93	CL311/80=A16/80=CDC G79	Calf; Midlothian, Scotland, UK
90	CL310/80=A15/80=CDC G68	Calf; Midlothian, Scotland, UK
100	CL604/80=McMeekin U31=CDC G100	Chiller water, poultry processing plant; Hobart, Australia
102	CL393/77=A49/77=CDC G102	Sputum; London, UK
206 CDC group IIe-like	CL721/92=CDC F9646	Brain biopsy; Pennsylvania, USA
218 CDC group IIe	CL717/92=CDC F7492	Eye lid; Tennessee, USA
219 CDC group IIe	CDC F3444	Sacral abscess; California, USA
223 CDC group IIe	CDC F5718	Tissue at base of meninges; North Carolina, USA
225 CDC group IIe?	CL718/92=CDC F1106	Genito-urinary tract; Washington, USA
226 CDC group IIe?	CL719/92=CDC F3670	Blood; Washington, USA
236 CDC group IIe	CL720/92=CDC F4391	Lung; Indiana, USA
237 CDC group IIe	CDC F6223	Blood; Georgia, USA
142 group (6)		
142^T	CL318/82^T=CDC $G229^T$=NCTC 13530^T=CCUG 60564^T	Sputum; Doncaster, UK
66	CL314/73=CDC F9968	Tongue swab; London, UK
79	CL144/74=CDC G42	Sputum; London, UK
146	CL229/85=CDC G233	Blood; Brighton, UK
147	CL126/81=CDC G234	Finger abscess; Melbourne, Australia
150	CL303/84=CDC G237	Sputum; London, UK
224 group (19)		
224 CDC group IIc	CDC F5649	Testicle; Iowa, USA
104	CL195/76	Blood culture; London, UK
105	CL184/75	Pleural aspirate; Sydney, Australia
107	CL187/75	Urine; Sydney, Australia
108	CL373/79	Blood culture; Zurich, Switzerland

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Strain	Other strain designation(s)	Source
109	CL205/78	Blood culture; Westcliff-on-Sea, UK
119	CL309/73	Blood culture; Birmingham, UK
120	CL524/73	Eye swab; Maidstone, UK
121	CL263/70=F144	Dialysis fluid; London, UK
155	CL445/80	Blood culture; London, UK
197	CL213/83	Eye swab; London, UK
201 CDC group IIc	CL709/92=CDC F8989	Eye; USA
203 CDC group IIh	CDC F4158	Eye; USA
214 CDC group IIh	CDC E7070	Blood; Florida, USA
227 CDC group IIc	CDC F1636	Blood; Miami, USA
229 CDC group IIc	CDC F3248	Wound; Texas, USA
232 CDC group IIc	CDC E6607	Urine; South Carolina, USA
239 CDC group IIc	CDC F283	Ear; Massachusetts, USA
241 CDC group IIc	CL710/92=CDC F7390	Leg ulcer; Rhode Island, USA
212 group (4)		
212 CDC group IIc	CL712/92=CDC F9257	Blood; Florida, USA
209 CDC group IIe	CL711/92=CDC F6666	Wound; Rhode Island, USA
213 CDC group IIe	CL713/92=CDC G134	Wound; California, USA
221 CDC group IIe	CDC F4031	CSF; Scotland, UK
217 group (2)		
217 CDC group IIi-like	CL716/92=CDC F859	Toe; Kansas, USA
233 CDC group IIi	CDC E6284	Urine; Hawaii, USA
231 group (2)		
231 CDC group IIi	CDC E8371	Urethra; Guam
215 CDC group IIi	CL714/92=CDC F715	Hand wound; Puerto Rico
255 group (6) ([*E. meningoseptica*] UB group II: 3)		
255	NCTC 11306=CIP 79.05=CDC G4075	Blood culture; Strasbourg, France; *F. meningosepticum* serotype H
111	CL496/75=CDC G146	Blood culture; Margate, UK
113	CL289/76=CDC G152	Unknown; Zurich, Switzerland
114	CL614/77=CDC G153	Urine; Dublin, Ireland
252	CL498/73=CDC G4072	Eye, keratitis; East Grinstead, UK
253	CL153/79=CDC G4073	Pyosalpinx; Worcester, UK
245 group (5) ([*E. meningoseptica*] UB group II: 1)		
245	ATCC 13254=NCTC 10585=CDC 422	Blood; Florida, USA; *F. meningosepticum* serotype B
246	ATCC 13255=NCTC 10586=CDC 3375	Spinal fluid and throat; South Carolina, USA; *F. meningosepticum* serotype C
247	Ursing & Bruun 267=CDC E6809	Blood; California, USA
248	Ursing & Bruun 265=CDC F3543	CSF; Florida, USA
249	Ursing & Bruun 266=CDC F3201	CSF; Kuwait
251 group (3) ([*E. meningoseptica*] UB group II:2)		
251	NCTC 11305=CIP 78.30=CDC G4071	Tracheal exudate; Strasbourg, France; *F. meningosepticum* serotype G

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Strain	Other strain designation(s)	Source
254	CL281/73=CDC G4074	Suction water; Reading, UK
258	Ursing & Bruun 238=CCUG 12664 =CDC G4121	Water; Sweden
Elizabethkingia meningoseptica UB group I (3)		
244^T	ATCC 13253^T=NCTC 10016^T=CDC 14^T	Spinal fluid; Massachusetts, USA
256	NCTC 13393=F8=E847=Greaves F2 =CDC G4076	Eye, conjunctivitis; Nottingham, UK
257	Ursing & Bruun 248=C. Richard 3.83 =CDC G4120	Urine; France
63 group (2)		
63^T	NCTC 11390^T=CDC KC1864^T=CCUG 60566^T=A140/68^T=F68^T	Milk bottle rinse, farm; Paisley, Scotland, UK
128	A139/68=CDC G197	As above
71 group (3)		
71	CL479/77=CDC F9973	Post-mortem lung; London, UK
69	CL12/79=A53/70=F131=CDC F9971	Sputum; Leicester, UK
148	CL542/79=CDC G235	Green lizard; London, UK
78 group (2)		
78^T	NCTC 13529^T=CCUG 60563^T=F91^T =CDC G41^T	Kidney abscess; Gloucester, UK
117	CL192/74=CDC G162	Urine; Newcastle-upon-Tyne, UK
92 group (2)		
92	ATCC 14234=F151=CL91/74=CDC G70	Unknown
97	CL94/78=Hayes P9/2=CDC G83	Pig carcass; unknown
95 group (2)		
95^T	CL88/78^T=Hayes B19/1^T=CDC G81^T =NCTC 13525^T=CCUG 60559^T	Beef; unknown
101	CL89/78=Hayes C8/1=CDC G101	Chicken; unknown
123 group (2)		
123	A86/68=CDC G186	Human unspecified; London, UK
153	CL278/82=CDC G240	Urine; Darlington, UK
125 group (2)		
125	A104/68=CDC G188	Milk swab, farm; Paisley, Scotland, UK
64	A103/68=CDC F9942	As above
137 group (2)		
137	CL636/74=CDC G208	Peritoneal pus; Preston, UK
204	CDC F4188	Wound; unknown
259 group (3) ([*E. meningoseptica*] UB group II: 4)		
259	Ursing & Bruun 2=H. Olsen 1=J100 =CDC G4122	Soil; Denmark
250	CL681/77=CDC G4070	Sputum; Melbourne, Australia
260	Ursing & Bruun 196=AB1572=CDC G4123	Lung (autopsy); Denmark

ATCC, American Type Culture Collection, Manassas, VA, USA; CDC, Centers for Disease Control, Atlanta, GA, USA. UB, Ursing and Bruun.

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Table 2

DNA–DNA hybridization used to classify strains of *C. indologenes, C. gleum, C. balustinum, C. indoltheticum* and *Empedobacter brevis*

For all DNA–DNA hybridization results (Tables 2, 3, 4, 5, 6, 8 and 9), 55 °C was the temperature for optimal reassociation and 70 °C was the stringent condition. Once reassociation was completed, the melting temperature (T_m) of the hybrid DNA was measured, and the columns labelled D (divergence) show the reduction in T_m between the related DNA sequences compared with the T_m of reassociated homologous sequences, due to the increased number of unpaired base pairs in heterologous sequences. Values are relative binding ratios, with homologous binding considered as 100%.

Source of unlabelled DNA	DNA–DNA relatedness (%) with labelled DNA from strain:														
	C. indologenes 50^T			*C. gleum* 51^T			*C. balustinum* 62^T			*C. indoltheticum* 106^T			*E. brevis* 55^T		
	55 °C	D	70 °C	55 °C	D	70 °C	55 °C	D	70 °C	55 °C	D	70 °C	55 °C	D	70 °C
C. indologenes															
50^T	100	0	100	50			19			16			9		
65	89	0.5	96	52									6		
67	89			48			13			17			10		
68	89	0	87	50									6		
70	88	0	86	50									6		
74	87	0	90	44									7		
75	85	0	80	44									6		
76	87	0	86	51									8		
77	84	0	82	48									5		
80	85	0.5	84	47									7		
116	87	0.5	86	59									6		
124	87	0.5	91	47									5		
141	87	0.5	83	33									7		
144	88	0.5	89	40									7		
145	82	0.5	87	50									5		
157	87	0.5	81	52	12								8		
C. gleum															
51^T	31			100	0	100	13			16			6		
59	41			80	5	68	15			14			11		
C. balustinum															
62^T	26			32			100	0	100	67	8.5	39	13		

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Source of unlabelled DNA	DNA–DNA relatedness (%) with labelled DNA from strain:														
	C. indologenes 50^T			*C. gleum* 51^T			*C. balustinum* 62^T			*C. indoltheticum* 106^T			*E. brevis* 55^T		
	55 °C	D	70 °C	55 °C	D	70 °C	55 °C	D	70 °C	55 °C	D	70 °C	55 °C	D	70 °C
C. indoltheticum															
106^T	35			32			49			100	0	100	7		
110	26			35			48			76	2.5	74	5		
140	23			13			46			82	3	77	6		
Empedobacter brevis															
54													82	0.5	
55^T	9			11			4			4			100	0	100
81													79	1	
83													75	0.5	
87													80	0.5	
91													80	1	
129													71	0.5	
159													78	1	
160													78	1	
161													77	1	
168													72	1	
183													69	1	
190													71	1	
191													71	1	
211													75	0.5	
123 group (*C. ureilyticum*)															
123	40			52			13			16			5		
153	29			31			5			13			5		
125 group (*C. shigense*)															
64	32			40			16			21			6		
125	33			42			16			23			7		
137 group (*F. lindanitolerans*)															
137	7			33			1			3			4		
204	5			5			4			8			5		

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Source of unlabelled DNA	DNA–DNA relatedness (%) with labelled DNA from strain:														
	C. indologenes 50^T			*C. gleum* 51^T			*C. balustinum* 62^T			*C. indoltheticum* 106^T			*E. brevis* 55^T		
	55 °C	D	70 °C	55 °C	D	70 °C	55 °C	D	70 °C	55 °C	D	70 °C	55 °C	D	70 °C
142 group (*C. bernardetii* sp. nov.)															
66	42			48			18			21			5		
79	34			45			15			14			5		
142^T	40			48			16			14			6		
146	37			47			14			13			5		
147	42			50			15			14			5		
150	44			60			13			13			8		
212 group (*C. taklimakanense* comb. nov.)															
209	11			9						16			5		
212	9			13			8			17			4		
213	9			13			13			14			4		
221	8			14			9			14			4		
217 group (*S. lactis*)															
217	3			6			3			3			4		
233	2			4			2			3			3		
224 group (*C. hominis*)															
104	14			16			10			9			9		
105	16			18			12			14			11		
107	13			16			10			8			7		
108	32			23			13			8			7		
109	14			15			11			10			6		
119	23			27			14						7		
120	12			21			7			7			7		
121	12			17			8			7			4		
155	20			38			10			10			7		
197	12			11			10			16			7		
201	11			7			10			17			8		
203	10			9			11			18			15		
214	11			16			11			17			5		

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Source of unlabelled DNA	DNA–DNA relatedness (%) with labelled DNA from strain:														
	C. indologenes 50^T			*C. gleum* 51^T			*C. balustinum* 62^T			*C. indoltheticum* 106^T			*E. brevis* 55^T		
	55 °C	D	70 °C	55 °C	D	70 °C	55 °C	D	70 °C	55 °C	D	70 °C	55 °C	D	70 °C
224	11			20			12			21			5		
227	11			17			10			19			4		
229	12			21			10			20			4		
232	15			14			12			19			4		
239	13			14			11			19			5		
241	9			14			9			19			3		
231 group (*S. daejeonense*)															
215	3			7			3			4			6		
231	4			4			2			5			2		
255 group (‘*E. meningoseptica* UB group II: 3)															
111				36			12			11			11		
113	15			21			7			5			6		
114	16			22			9			5			7		
58 group (*W. falsenii* genomovar 2)															
53	9			9			6			6			41		
56	12			16			6			3			45		
57	11			15			6			4			48		
58	7			9			6			5			40		
82	9			9			6			4			53	5	
84	4			7			4			4			42		
85	4			6			4			4			42		
88	36			37			16			14			28		
89	5			6			4			9			46	7	
94	5			6			4			3			42		
96	2			7			4			4			47		
99	9			9			4			3			47		
162	10			20			4			9			42		
164	9			10			4			9			49		
165	9			9			6			12			49		

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Source of unlabelled DNA	DNA–DNA relatedness (%) with labelled DNA from strain:														
	C. indologenes 50^T			*C. gleum* 51^T			*C. balustinum* 62^T			*C. indoltheticum* 106^T			*E. brevis* 55^T		
	55 °C	D	70 °C	55 °C	D	70 °C	55 °C	D	70 °C	55 °C	D	70 °C	55 °C	D	70 °C
166	7			8			4			10			42		
167	9			46			4			6			34		
169	6			8			3			8			38		
170	5			6			2			6			41		
171	16			8			5			8			38		
172	7			8			3			8			33		
173	8			8			3			8			41		
174	4			7			4			7			38		
175	5			7			2			8			34		
176	6			4			2			8			33		
177	5			7			3			8			41		
178	4			4			3			7			41		
179	4			6			3			9			40		
180	4			5			2			8			35		
181	4			6			3			7			36		
182	5			6			3			8			42		
184	5			6			6			8			50		
185	5			6			7			7			38		
186	4			5			5			7			35		
187	5			9			6			7			37		
188	6			4			6			9			39		
189	4			3			4			7			33		
192	4			5			5			6			36		
193	5			3			9			7			38		
194	5			4			6			7			40		
195	7			5			8			8			39		
196	4			2			5			7			36		
199	12			6			7			8			38		
200	7			4			5			7			37		

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Source of unlabelled DNA	DNA–DNA relatedness (%) with labelled DNA from strain:														
	C. indologenes 50^T			*C. gleum* 51^T			*C. balustinum* 62^T			*C. indoltheticum* 106^T			*E. brevis* 55^T		
	55 °C	D	70 °C	55 °C	D	70 °C	55 °C	D	70 °C	55 °C	D	70 °C	55 °C	D	70 °C
202	11			2			6			7			6		
205	5			3			8			6			39		
216	4			5			5			6			37		
63 group (*C. lactis* sp. nov.)															
63^T	56	11	30	44			21			19			9		
128	51			48			14			13			5		
71 group (‘*C. gleum*-like species 1’)															
69	40			65	9.5		15			17			6		
71	36			59	9.5		13			18			5		
148	45			43			5			14			5		
78 group (*C. nakagawai* sp. nov.)															
78^T	39			47			16			18			6		
117	49			57			21			11			6		
92 group (‘*E. brevis*-like species 1’)															
92	8			7			4			5			45		
97	7			9			4			4			34		
93 group (*C. anthropi*)															
90	11			15			9			11			10		
93	12			13			8			10			6		
100	14			12			9			5			42		
102	15			15			12			24			12		
206	9			7			11			19			8		
218	9			14						16			5		
219	10			15			10			17			4		
223	9			14			9			21			4		
225	9			11			10			20			4		
226	10			14			10			21			4		
236	12			7			10			19			4		
237	12			13			10			19			4		

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Source of unlabelled DNA	DNA–DNA relatedness (%) with labelled DNA from strain:														
	C. indologenes 50^T			*C. gleum* 51^T			*C. balustinum* 62^T			*C. indoltheticum* 106^T			*E. brevis* 55^T		
	55 °C	D	70 °C	55 °C	D	70 °C	55 °C	D	70 °C	55 °C	D	70 °C	55 °C	D	70 °C
95 group (*C. carnis* sp. nov.)															
95^T	19			14			8			10			8		
101	17			15			7			7			11		

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Table 3

DNA–DNA hybridization used to classify strains of groups 58, 93, 142, 224 and 212

Source of unlabelled DNA	DNA–DNA relatedness (%) with labelled DNA from strain:														
	58			93			142^T			224			212		
	55 °C	D	70 °C	55 °C	D	70 °C	55 °C	D	70 °C	55 °C	D	70 °C	55 °C	D	70 °C
58 group (*W. falsenii* genomovar 2)															
53	85	3	77												
56	83	2.5	78												
57	87	1	87												
58	100	0	100	9			6			18			7		
82	83	3	73												
84	77	3.5	72												
85	73	4.5	65												
88	77	4.5	60												
89	80	2.5	78												
94	87	2.5	85												
96	78	3	77												
99	79	2.5	74												
162	75	3.5	68												
164	86	2.5	82												
165	98	4.5	83												
166	87	2.5	86												
167	96	0.5	100												
169	83	2	82												
170	56	1.5	62				39			12					
171	88	1	89												
172	92	0.5	95												
173	100	1	100												
174	81	2.5	80												
175	81	3	77												
176	78	3.5	74												

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Source of unlabelled DNA	DNA–DNA relatedness (%) with labelled DNA from strain:														
	58			93			142^T			224			212		
	55 °C	D	70 °C	55 °C	D	70 °C	55 °C	D	70 °C	55 °C	D	70 °C	55 °C	D	70 °C
177	85	3	80												
178	92	2	90												
179	98	2	96												
180	84	2.5	82												
181	88	2.5	86												
182	100	0	100												
184	100	0.5	100												
185	100	0.5	100												
186	84	2	83												
187	100	0	100												
188	87	1.5	85												
189	79	1.5	77												
192	95	0	98												
193	81	2	80												
194	95	0.5	98												
195	77	3	74												
196	79	3	76												
199	94	1.5	88												
200	82	2	73												
202	73	2.5	69												
205	87	3	80												
216	78	2	69												
243	86	1.5	84												
93 group (*C. anthropi*)															
90	23			100	0	100									
93	18			100	0	100	5			33			33		
100	16			65	4.5	51	14			35					
102	17			72	1	71									
206	13			78	1	77									

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Source of unlabelled DNA	DNA–DNA relatedness (%) with labelled DNA from strain:														
	58			93			142^T			224			212		
	55 °C	D	70 °C	55 °C	D	70 °C	55 °C	D	70 °C	55 °C	D	70 °C	55 °C	D	70 °C
218	8			73	1	70									
219	6			77	1	74									
223	5			69	1.5	66									
225	4			72	1	74									
226	8			67	1	65									
236	5			76	1.5	70									
237	8			70	1	70									
142 group (*C. bernardetii* sp. nov.)															
66	30			25			87	1	85	30					
79	32			23			87	1	85	30					
142^T	13			27			100	0	100	31			10		
146	13			23			86	0	87	15					
147	11			22			89	2	84	23					
150	15			30			86	1	84	23					
224 group (*C. hominis*)															
104	24			29						81	2	77			
105	21			26						76	3	68			
107	18			29						83	3.5	78			
108	11			28						84	2.5	78			
109	11			25						69	2.5	64			
119	10			25						68	3.5	58			
120	15			30						76	3.5	63			
121	9			27						74	3	66			
155	12			29						82	2.5	79			
197	10			26						70	3	67			
201	19			26						67	3	66			
203	13			43	3.5	29				73	4.6	66			
214	7			27						87	2.5	87			
224	7			51	3	46	11			100	0	100	27		

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Source of unlabelled DNA	DNA–DNA relatedness (%) with labelled DNA from strain:														
	58			93			142^T			224			212		
	55 °C	D	70 °C	55 °C	D	70 °C	55 °C	D	70 °C	55 °C	D	70 °C	55 °C	D	70 °C
227	5			35						85	2.5	81			
229	11			33						78	2.5	75			
232	5			28						86	2	85			
239	7			33						79	2.5	78			
241	5			21						74	2.5	64			
212 group (*C. taklimakanense* comb. nov.)															
209	6			34									87	1.5	77
212	7			33			10			27			100	0	100
213	5			31									83	0.5	66
221	8			35									82	2	66
123 group (*C. ureilyticum*)															
123	14			28			30			29			23		
153	12			21			40			28			17		
125 group (*C. shigense*)															
64	29			25											
125	29			28			30			28			18		
137 group (*F. lindanitolerans*)															
137	6			9			7			15			7		
204	7			17									8		
217 group (*S. lactis*)															
217	7			9			2			4			12		
233	4			7									5		
231 group (*S. daejeonense*)															
215	6			8									12		
231	2			8			3			16			4		
245 group ([*E. meningoseptica*] UB group II: 1)															
245							9								
246							6								
247							10								

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Source of unlabelled DNA	DNA–DNA relatedness (%) with labelled DNA from strain:														
	58			93			142^{T}			224			212		
	55 °C	D	70 °C	55 °C	D	70 °C	55 °C	D	70 °C	55 °C	D	70 °C	55 °C	D	70 °C
248							13								
249							14								
251 group ([*E. meningoseptica*] UB group II: 2)															
251							7								
254							18								
258							14								
255 group ([*E. meningoseptica*] UB group II: 3)															
111	29			15									14		
113	14			19									16		
114	11			26			13			27			16		
252							7								
253							6								
255							12								
259 group ([*E. meningoseptica*] UB group II: 4)															
250							8								
259							6								
260							5								
63 group (*C. lactis* sp. nov.)															
63^{T}	29			18			36			31			20		
128	13			26			44			40			18		
71 group (‘*C. gleum*-like species 1’)															
69	40			22						29			24		
71	37			21			38			28			19		
148	15			24						23			20		
78 group (*C. nakagawai* sp. nov.)															
78^{T}	35			20			50			28			19		
117	12			24			52			36			20		

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Source of unlabelled DNA	DNA–DNA relatedness (%) with labelled DNA from strain:														
	58			93			142^T			224			212		
	55 °C	D	70 °C	55 °C	D	70 °C	55 °C	D	70 °C	55 °C	D	70 °C	55 °C	D	70 °C
92 group (‘*E. brevis*-like species 1’)															
92	49	13	26	10			3			18			7		
97	53	12.5	28	58	0	67	26			28			16		
95 group (*C. carnis* sp. nov.)															
95^T	14			34			12			29			24		
101	19			33			8			27			27		
244 group (*E. meningoseptica* UB group I)															
244							17								
256							15								
257							13								

See legend to Table 2 for further details.

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Table 4

DNA–DNA hybridization used to classify strains of groups 217, 231, 255, 245 and 251

Source of unlabelled DNA	DNA–DNA relatedness (%) with labelled DNA from strain:														
	217			231			255			245			251		
	55 °C	D	70 °C	55 °C	D	70 °C	55 °C	D	70 °C	55 °C	D	70 °C	55 °C	D	70 °C
217 group (*S. lactis*)															
217	100	0	100	21			15			3					
233	80	3	76	26											
231 group (*S. daejeonense*)															
215	28			88	3	85									
231	29			100	0	100	2			3					
255 group ([*E. meningoseptica*] UB group II: 3)															
111							81								
113							81								
114	4						82	1	78	68	5.5	47			
252							100	0.5	100	65	6.5	38			
253							100	0.5	100	63	6.5	49			
255							100	0	100	65	6	43	71	4	52
245 group ([*E. meningoseptica*] UB group II: 1)															
245							65	6	37	100	0	100	61	6	41
246										95	1	89			
247										90	1	77			
248										83	1	73			
249										90	1	85			
251 group ([*E. meningoseptica*] UB group II: 2)															
251							71	4	52	61	6	41	100	0	100
254										67	5.5	51	95	1.5	100
258										63	6	49	93	0	96
123 group (*C. ureilyticum*)															
123	3						17			13			15		
153	3						12			13			18		

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Source of unlabelled DNA	DNA–DNA relatedness (%) with labelled DNA from strain:														
	217			231			255			245			251		
	55 °C	D	70 °C	55 °C	D	70 °C	55 °C	D	70 °C	55 °C	D	70 °C	55 °C	D	70 °C
259 group ([*E. meningoseptica*] UB group II: 4)															
250							75	5	46	66	6.5	39	75	5	50
259							64	5.5	36	55	7.5	33	64	5	45
260							62	5.5	36	61	10	34	65	5	47
63 group (*C. lactis* sp. nov.)															
63^{T}	2						13			13					15
128	4						14			13			20		
71 group (‘*C. gleum*-like species 1’)															
69	1														
71	2						13			13			12		
148	4														
78 group (*C. nakagawai* sp. nov.)															
78^{T}	2						17			15					17
117	2						14			18			18		
92 group (‘*E. brevis*-like species 1’)															
92	4						5			6			7		
97	1						8			7			10		
95 group (*C. carnis* sp. nov.)															
95^{T}	4						8			7					8
101	6						7			6			9		
244 group (*E. meningoseptica* UB group I)															
244							38			50	6.5	43	42		
137 group (*F. lindanitolerans*)															
137	4						6			4			6		
125 group (*C. shigense*)															
125	4						15			15			18		

See legend to Table 2 for further details.

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Table 5

DNA–DNA hybridization used to classify strains of groups 244, 63, 71, 78 and 92

Source of unlabelled DNA	DNA–DNA relatedness (%) with labelled DNA from strain:														
	244^T			63^T			71			78^T			92		
	55 °C	D	70 °C	55 °C	D	70 °C	55 °C	D	70 °C	55 °C	D	70 °C	55 °C	D	70 °C
244 group (*E. meningoseptica* UB group I)															
244^T	100	0	100	20									5		
256	87	0	91												
257	82	0	82												
63 group (*C. lactis* sp. nov.)															
63^T	20			100	0	100	48			38			4		
128	23			100	1	100	54			39					
71 group (‘*C. gleum*-like species 1’)															
69							96	1.5	100	35					
71	16			36			100	0	100	34			10		
148							97	0.5	100	43					
78 group (*C. nakagawai* sp. nov.)															
78^T	16			37			43	11.5		100	0	100	4		
117	16			39			55	14		69	4	59			
92 group (‘*E. brevis*-like species 1’)															
92	5			4			10			4			100	0	100
97	7			17			4			10			95	2	88
123 group (*C. ureilyticum*)															
123	18			36			51	13.5		49			4		
153	25			37			51	10		29			8		
125 group (*C. shigense*)															
64															
125	15			31			52			30			4		
137 group (*F. lindanitolerans*)															
137	7			8			12			5			3		
204										5					

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Source of unlabelled DNA	DNA–DNA relatedness (%) with labelled DNA from strain:														
	244^T			63^T			71			78^T			92		
	55 °C	D	70 °C	55 °C	D	70 °C	55 °C	D	70 °C	55 °C	D	70 °C	55 °C	D	70 °C
259 group ([*E. meningoseptica*] UB group II: 4)															
250	52														
259	54			13									6		
260	50														
95 group (*C. carnis* sp. nov.)															
95^T	12			11			24			10			15		
101	9			9			28			9			2		

See legend to Table 2 for further details.

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Table 6

DNA–DNA hybridization used to classify strains of groups 95, 123, 125, 137 and 259

Source of unlabelled DNA	DNA–DNA relatedness (%) with labelled DNA from strain:														
	95^T			123			125			137			259		
	55 °C	D	70 °C	55 °C	D	70 °C	55 °C	D	70 °C	55 °C	D	70 °C	55 °C	D	70 °C
95 group (*C. carnis* sp. nov.)															
95^T	100	0	100	19			21			7			8		
101	83						20								
123 group (*C. ureilyticum*)															
123	16			100	0	100	32			13					
153	13			85	0		40								
125 group (*C. shigense*)															
64							100	0	100						
125	22			35			100	0	100	13					
137 group (*F. lindanitolerans*)															
137	7			13			13			100	0	100	4		69
204							7			90	1	85			100
259 group ([*E. meningoseptica*] UB group II: 4)															
250													90	4	79
259	8												100	0	
260													85	1.5	

See legend to Table 2 for further details.

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Table 7

Summary of disposition of the various groups studied

Group in Table 1	Species name in publication	Representative strain	Provisional CDC group(s)
58 group (48 strains)	*W. falsenii* genomovar 2	CDC KC1862	None (47), IIh (1)
63 group (2)	*C. lactis* sp. nov.	CDC KC1864^T	None (2)
71 group (3)	'*C. gleum*-like species 1'	CDC F9973	None (3)
78 group (2)	*C. nakagawai* sp. nov.	CDC G41^T	None (2)
92 group (2)	'*E. brevis*-like species 1'	CDC G70	None (2)
93 group (12)	*C. anthropi*	CDC G79	None (4), IIe (5), IIe? (2), IIe-like (1)
95 group (2)	*C. carnis* sp. nov.	CDC G81^T	None (2)
123 group (2)	*C. ureilyticum*	CDC G186	None (2)
125 group (2)	*C. shigense*	CDC G188	None (2)
137 group (2)	*F. lindanitolerans*	CDC G208	None (2)
142 group (6)	*C. bernardetii* sp. nov.	CDC G229^T	None (6)
212 group (4)	*C. taklimakanense* comb. nov.	CDC F9257	IIc (1), IIe (3)
217 group (2)	*S. lactis*	CDC F859	IIi (1), IIi-like (1)
224 group (19)	*C. hominis*	CDC F5649	None (10), IIc (7), IIh (2)
231 group (2)	*S. daejeonense*	CDC E8371	IIi (2)
245 group (5) ([*E. meningoseptica*] UB group II: 1)	*E. meningoseptica* genomosp. 1	CDC 422	None (5)
251 group (3) ([*E. meningoseptica*] UB group II: 2)	*E. meningoseptica* genomosp. 2	CDC G4071	None (3)
255 group (6) ([*E. meningoseptica*] UB group II: 3)	*E. meningoseptica* genomosp. 3	CDC G4075	None (6)
259 group (3) ([*E. meningoseptica*] UB group II: 4)	*E. meningoseptica* genomosp. 4	CDC G4122	None (3)
C. balustinum (1)			None (1)
C. gleum (2)			None (2)
C. indologenes (16)			None (16)
C. indoltheticum (3)			None (3)
244 group (3) (*E. meningoseptica* UB group I)			None (3)
Empedobacter brevis (15)			None (15)
Distinct groups, each represented by a single strain (15)			None (9), IIc (1), IIc? (1), IIe? (1), IIh (1), IIh? (1), IIi (1)

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Table 8

DNA relatedness of groups 95, 231, 21 7 and 58 identified in this study and type strains of more recently described taxa

Source of unlabelled DNA	DNA–DNA relatedness (%) with labelled DNA from strain:														
	S. jeonii JCM 12382^T			*S. daejeonense* CCUG 52468^T			*S. mizuiaii* NCTC 12149^T			*W. fahenii* CCUG 51536^T			*W. fahenii* CCUG 51537*		
	55 °C	D	70 °C	55 °C	D	70 °C	55 °C	D	70 °C	55 °C	D	70 °C	55 °C	D	70 °C
95^T (95 group)	20	13.0	5	ND	–	–	ND	–	–	ND	–	–	ND	–	–
231 (231 group)	ND	–	–	80	1.5	84	ND	–	–	ND	–	–	ND	–	–
217 (217 group)	ND	–	–	ND	–	–	21	15.0	2	ND	–	–	ND	–	–
58 (58 group)	ND	–	–	ND	–	–	ND	–	–	85	3.5	80	95	0.5	90

See legend to Table 2 for further details, ND, Not done.

*Genomovar 2.

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Table 9

DNA relatedness of groups 212, 224, 123 and 125 identified in this study and type strains of more recently described taxa

Source of unlabelled DNA	DNA–DNA relatedness (%) with labelled DNA from strain:											
	C. haifense DSM 19056^T			*C. hominis* CCUG 52711^T			*C. joostei* CCUG 46665^T			*C. shigense* NCIMB 14047^T		
	55 °C	D	70 °C	55 °C	D	70 °C	55 °C	D	70 °C	55 °C	D	70 °C
212 (212 group)	18	15.5	4	ND	–	–	ND	–	–	ND	–	–
224 (224 group)	ND	–	–	96	2.5	95	ND	–	–	ND	–	–
123 (123 group)	ND	–	–	ND	–	–	55	10.0	21	ND	–	–
125 (125 group)	ND	–	–	ND	–	–	ND	–	–	77	7.0	57

See legend to Table 2 for further details. ND, Not done.

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Table 10

Phenotypic characteristics useful for the differentiation of *C. bernardetii* sp. nov. (142 group), *C. lactis* sp. nov. (63 group) and *C. nakagawai* sp. nov. (78 group) from their nearest neighbours

Taxa: 1, *C. bernardetii* sp. nov.; 2, *C. lactis* sp. nov.; 3, *C. nakagawai* sp. nov.; 4, C. aquifrigidense NCTC 13488^{T}; 5, C. jejuense NCTC 13492^{T}; 6, C. joostei NCTC 13454^{T}.

Characteristic	1	2	3	4	5	6
Acid in ASS medium from fructose	+	+	−	−	−	+
Growth at/on:						
5 °C	−	d	−	−	+	+
37 °C	+	+	+	+	−	+
Cetrimide agar	−	−	d	−	+	−
MacConkey agar	+	+	−	+	+	+
Hydrolysis of:						
Starch	d	−	+	−	+	−
Tween 20	d	+	+	−	+	+
Tween 80	d	+	d	−	+	+
Tyrosine	+	d	+	−	+	+
Urease production	d	−	−	−	−	+
Christensen’s citrate	d	−	d	+	+	+

+, All strains tested positive; d, strains give different results; −, all strains tested negative.

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Table 11

Phenotypic characteristics useful for the differentiation of *C. carnis* sp. nov. (95 group) from its nearest neighbours

Taxa: 1, *C. carnis* sp. nov.; 2, *Chryseobacterium* (*Sejongia*) *antarcticum* NCTC 13489^T; 3, *Chryseobacterium* (*Sejongia*) *jeonii* NCTC 13459^T; 4, *Chryseobacterium* (*Sejongia*) *marinum* (data from Lee *et al.*, 2007). Data are from this study unless indicated. +, All strains positive; −, all strains negative; ND, no data available. Kämpfer *et al.* (2009a) proposed that *Sejongia* species be transferred to *Chryseobacterium*, a proposal supported in this study.

Characteristic	1	2	3	4
Acid in ASS medium from:				
Glucose	+	−	−	ND
Maltose	+	−	−	ND
Growth at/on:				
37 °C	+	−	−	−
MacConkey agar	+	−	−	ND
β-Hydroxybutyrate	+	−	−	ND
Hydrolysis of:				
Aesculin	−	+	+	+
Starch	+	+	−	ND
Tween 20	+	+	−	ND
Nitrate reduction	−	+	−	ND
Oxidase production	+	+	+	−
Casein digestion	+	−	−	ND
DNase production	+	−	−	ND
Gelatin liquefaction	+	−	+	ND

* Positive after 5 days of incubation.