Kerstersia gyiorum gen. nov., sp. nov., a novel *Alcaligenes faecalis*-like organism isolated from human clinical samples, and reclassification of *Alcaligenes denitrificans* Rger and Tan 1983 as *Achromobacter denitrificans* comb. nov.

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A polyphasic taxonomic study was performed on nine isolates recovered from various human clinical samples. Phenotypically, these isolates resembled *Alcaligenes faecalis*. Whole-cell protein analysis distinguished two different species, and this was confirmed by DNA–DNA hybridizations. Cellular fatty acid analysis and 16S rDNA sequence analysis indicated that these isolates were related to the genera *Alcaligenes*, *Bordetella*, *Achromobacter* and *Pigmentiphaga* and belonged to the family *Alcaligenaceae*. On the basis of the results of this study, the organisms were classified in a novel genus, *Kerstersia* gen. nov. This genus comprises one species, *Kerstersia gyiorum* sp. nov. (type strain LMG 5906^T = API 184-2-84^T = CCUG 47000^T), and several unnamed isolates. The DNA G + C content of members of the genus *Kerstersia* is between 61.5 and 62.9 mol%. On the basis of previously published DNA–DNA hybridization results and data from chemotaxonomic studies, it is proposed that *Alcaligenes denitrificans* Rger and Tan 1983 be reclassified as *Achromobacter denitrificans* comb. nov.

INTRODUCTION

The genera *Alcaligenes*, *Achromobacter*, *Bordetella* and *Pigmentiphaga* belong to the β -Proteobacteria and are grouped together in the family *Alcaligenaceae* (De Ley *et al.*, 1986; Yabuuchi *et al.*, 1998; Blmel *et al.*, 2001). The genus *Alcaligenes* has undergone considerable changes since its creation in 1919 and is now limited to *Alcaligenes faecalis* (the type species), *Alcaligenes latus* and *Alcaligenes defragrans* (Yabuuchi *et al.*, 1998; Foss *et al.*, 1998; Palleroni & Palleroni, 1978). *Alcaligenes faecalis* has been isolated from a wide variety of ecological niches including soil, water and various clinical samples (Kersters & De Ley, 1984). *Alcaligenes defragrans* strains have been isolated from soil and can use alkenoic monoterpenes as sole carbon source (Foss *et al.*, 1998). *Alcaligenes latus* was considered a species *insertae sedis* by Kersters & De Ley (1984), and recent data

have shown that this organism is closely related to the genera *Rubrivivax* and *Ideonella* and thus belongs to the *Comamonadaceae* (T. Coenye and P. Vandamme, unpublished data). The taxonomy of the genus *Alcaligenes* is closely intertwined with the taxonomy of the genus *Achromobacter*, and several *Alcaligenes* species have recently been reclassified as *Achromobacter* species (Yabuuchi *et al.*, 1998). The genus *Achromobacter* now contains three species [*Achromobacter xylosoxidans* (the type species), *Achromobacter piechaudii* and *Achromobacter ruhlandii*]. According to Yabuuchi *et al.* (1998), the species *Achromobacter xylosoxidans* encompasses two subspecies (*Achromobacter xylosoxidans* subsp. *denitrificans* and *Achromobacter xylosoxidans* subsp. *xylosoxidans*), but this contradicts previous work showing that there was sufficient evidence (including DNA relatedness studies) to allow the two taxa to be considered as distinct species (Vandamme *et al.*, 1996). Below, we refer to these taxa as *Achromobacter xylosoxidans* and *Alcaligenes denitrificans*, respectively. *Achromobacter xylosoxidans* is an opportunistic human pathogen capable of causing a wide variety of infections and is widespread in oligotrophic aquatic niches (Kersters & De Ley, 1984). *Alcaligenes denitrificans* strains are found in soil but can

The GenBank accession numbers for the 16S rRNA gene sequences of strains LMG 5890 and LMG 5906^T are respectively AY131212 and AY131213.

Protein profiles of the *Kerstersia* strains, fatty acid compositions of the strains studied and MIC values of the *Kerstersia* strains are available as supplementary material in IJSEM Online.

occasionally also be found in human clinical samples (Kerstens & De Ley, 1984). *Achromobacter piechaudii* has been isolated from soil and human clinical samples, including blood (Kiredjian *et al.*, 1986). *Achromobacter ruhlandii* is a soil commensal and is not known to be pathogenic to humans (Kerstens & De Ley, 1984). The genus *Bordetella* was described in 1952 (Moreno-López, 1952) and originally contained three species: *Bordetella pertussis* (the type species), *Bordetella parapertussis* and *Bordetella bronchiseptica*. *B. pertussis* is the causative agent of whooping cough, and *B. parapertussis* is closely related to *B. pertussis* but causes a milder form of disease (Pittman, 1984). *B. bronchiseptica* is traditionally isolated from the respiratory tract of wild and domestic animals, but this organism can occasionally cause respiratory tract infections in humans as well (Pittman, 1984). Several novel *Bordetella* species have been described since then: *Bordetella avium* (isolated from the respiratory tract of turkeys and other birds; Kersters *et al.*, 1984), *Bordetella holmesii* (formerly CDC group NO-2, isolated from human blood; Weyant *et al.*, 1995), *Bordetella hinzii* (isolated from the respiratory tract of animals and humans; Vandamme *et al.*, 1995), *Bordetella trematum* (isolated from various human clinical samples, including blood and infected ears; Vandamme *et al.*, 1996) and *Bordetella petrii* (isolated from an anaerobic bioreactor; von Wintzingerode *et al.*, 2001). Recently, a novel member of the family *Alcaligenaceae* was described, *Pigmentiphaga kullae* (Blümel *et al.*, 2001). This organism was isolated from soil and is capable of the aerobic degradation of azo dyes.

We performed a polyphasic taxonomic study to elucidate the taxonomic position of nine isolates, from human clinical samples, that phenotypically resembled *Alcaligenes faecalis*, and show that these isolates belong to a novel genus, for which we propose the name *Kerstesia*. On the basis of previously published data, we also propose the formal

reclassification of *Alcaligenes denitrificans* Rüger and Tan 1983 as *Achromobacter denitrificans* comb. nov.

METHODS

Bacterial strains and growth conditions. The *Kerstesia* strains used in this study are listed in Table 1. Reference strains of other taxa have been described previously (Vandamme *et al.*, 1995, 1996; Foss *et al.*, 1998; Yabuuchi *et al.*, 1998). All strains were grown aerobically on trypticase soy agar (BBL) at 37 °C unless otherwise indicated.

SDS-PAGE of whole-cell proteins. Strains were grown on trypticase soy agar for 48 h at 37 °C. Preparation of whole-cell proteins and SDS-PAGE were performed as described previously (Pot *et al.*, 1994). Densitometric analysis, normalization and interpolation of the protein profiles, as well as numerical analysis using Pearson's product-moment correlation coefficient, were performed using the GelCompar 4.2 software package (Applied Maths).

16S rDNA sequencing. DNA was prepared by heating one or two colonies at 95 °C for 15 min in 20 µl lysis buffer containing 0.25 % (w/v) SDS and 0.05 M NaOH. Following lysis, 180 µl distilled water was added to the lysate. The sequences of the 16S rRNA genes of strains LMG 5890 and LMG 5906^T were determined as described previously (Coenye *et al.*, 1999). Phylogenetic analyses and bootstrap analysis (1000 replicates) was performed using the KODON 1.5 software package (Applied Maths); a phylogenetic tree was constructed using the neighbour-joining method (Saitou & Nei, 1987).

Preparation of high-molecular-mass DNA and DNA–DNA hybridization experiments. Preparation of high-molecular-mass DNA for DNA–DNA hybridization experiments and the determination of the degree of DNA–DNA binding by the initial renaturation rate method were performed as described previously (Vandamme *et al.*, 1992; De Ley *et al.*, 1970). Each value given is the mean of at least two hybridization experiments. The total DNA concentration was 65 µg ml⁻¹ and the optimal renaturation temperature in 2 × SSC (1 × SSC is 0.15 M NaCl plus 0.015 M sodium citrate, pH 7.0) was 79 °C. Alternatively, high-molecular-mass DNA was prepared as described by Pitcher *et al.* (1989), and DNA–DNA hybridizations were performed with photobiotin-labelled probes in microplate wells, as described by Ezaki *et al.* (1989), using an HTS7000 Bio

Table 1. Strains included in this study

Abbreviations: API, Appareils et Procédés d'Identification, Montalieu-Vercieu, France; CCUG, Culture Collection, University of Göteborg, Göteborg, Sweden; LMG, BCCM/LMG Bacteria Collection, Laboratorium voor Microbiologie, Gent, Belgium.

Strain	Other strain designation(s)	Depositor	Source
<i>Kerstesia gyiorum</i> gen. nov., sp. nov.			
LMG 15979	CCUG 34029	Public Health Laboratory, Göteborg, Sweden	Human faeces (85-year-old woman, Sweden, 1995)
LMG 15539	CCUG 24721, GLG 5504	G. L. Gilardi	Human leg wound (1986)
LMG 5905t1	API 183-2-84	D. Monget	Human leg wound
LMG 5895	API 150-2-84, CCUG 24726, GLG 4645	D. Monget	Human leg wound (1983)
LMG 5906 ^T	API 184-2-84 ^T , CCUG 47000 ^T	D. Monget	Human ankle wound
R-2516	BIJL977	G. Claeys	Human sputum (Belgium, 1997)
<i>Kerstesia</i> spp.			
LMG 5890	API 145-2-84, CCUG 46999	D. Monget	Human leg wound (USA, 1983)
LMG 5891	API 146-2-84, CCUG 46998	D. Monget	Human leg wound (USA, 1983)
LMG 5892	API 147-2-84, CCUG 24728, GLG 4530	D. Monget	Human leg wound (USA, 1983)

Assay Reader (Perkin-Elmer) for the fluorescence measurements. The hybridization temperature was 50 °C. Reciprocal experiments were performed for every pair of strains.

Determination of DNA base composition. DNA (prepared as described above) was enzymically degraded into nucleosides as described by Mesbah *et al.* (1989). The nucleoside mixture obtained was then separated by HPLC using a Waters SymmetryShield C8 column thermostatted at 37 °C. The solvent was 0.02 M $\text{NH}_4\text{H}_2\text{PO}_4$ (pH 4.0) with 1.5% acetonitrile. The detector used was a Waters model 484 UV-VIS absorbance detector set at 270 nm. Non-methylated λ phage DNA (Sigma) was used as the calibration reference.

Fatty acid methyl ester analysis. After an incubation period of 24 h at 35 °C, a loopful of well-grown cells was harvested and fatty acid methyl esters were prepared as described previously (Vandamme *et al.*, 1992), separated and then identified using the Sherlock Microbial Identification System (version 3.0; MIDI).

Phenotypic characterization. API galleries (API 50 CH, API 50AO and API 50AA; bioMérieux) were used to determine the assimilation of 147 organic compounds as sole carbon sources, as described previously (Kersters *et al.*, 1984). Classical phenotypic tests were performed as described by Vandamme *et al.* (1993). API 20NE tests were performed according to the recommendations of the manufacturer (bioMérieux). Strains LMG 5890, LMG 5891, LMG 5892, LMG 5905, LMG 5895 and LMG 5906^T were investigated with the API 50 galleries; strains LMG 5892, LMG 5895, LMG 15539 and LMG 15979 were investigated using classical phenotypic tests and API 20NE tests.

Antimicrobial susceptibility testing. MIC values towards levofloxacin, ciprofloxacin, ofloxacin, sparfloxacin, erythromycin, roxithromycin, clarithromycin, azithromycin, cefotaxim, ceftiofur and rifampicin were determined for eight strains (R-2516 was not included in the analysis) by using the agar dilution method conforming to the guidelines of the NCCLS (1995). Strains were grown on Mueller–Hinton agar (BRL) for 16–20 h at 35 °C.

RESULTS AND DISCUSSION

Phylogenetic position and structure of the genus *Kerstersia*

The reproducibility of SDS-PAGE of whole-cell proteins was checked by preparing protein extracts in duplicate; the level of correlation between the patterns was more than 93% (data not shown). After numerical analysis and visual comparison of the profiles, three clusters could be delineated, while isolate LMG 5890 and reference strains of *Bordetella*, *Alcaligenes* and *Achromobacter* species occupied separate positions in the dendrogram (Fig. 1). A first cluster contained LMG 5891 and LMG 5892. Strains LMG 15539, R-2516 and LMG 15979 formed a second cluster. Finally, LMG 5895, LMG 5905 and LMG 5906^T constituted a third cluster. Visual comparison of the patterns indicated that LMG 5890 was most similar to cluster I isolates LMG 5891 and LMG 5892 and that isolates from clusters II and III formed a second group. This discrepancy between numerical analysis and visual comparison was due to slight but reproducible variation in the position of a high-intensity band within the profile, which had a profound effect on numerical analysis (data not shown). The complete 16S rDNA sequences of isolates LMG 5890 and LMG 5906^T were

Fig. 1. Dendrogram derived from the UPGMA linkage of correlation coefficients between the protein patterns of the strains studied. The correlation coefficient is expressed as percentage similarity for convenience.

determined and compared with available 16S rDNA sequences of other β -Proteobacteria (Fig. 2). The 16S rDNA sequences of LMG 5890 and LMG 5906^T were very similar to each other (98.3%). The levels of similarity towards *Bordetella* species (93.5–96.4%), *Achromobacter* species (91.6–95.1%), *Alcaligenes* species (92.7–94.8%) and *P. kullae* (92.9–93.5%) were lower. Bootstrap analysis indicated that strains LMG 5890 and LMG 5906^T formed a stable phylogenetic group (the bootstrap value was 100%). DNA was prepared from isolates LMG 5890, LMG 5906^T and LMG 15539 and from reference strains of *Alcaligenes faecalis*, *Alcaligenes denitrificans*, *B. hinzii*, *Achromobacter xylosoxidans*, *Achromobacter ruhlandii* and *Achromobacter piechaudii*. DNA–DNA binding values are shown in Table 2. The G+C contents of isolates LMG 5890, LMG 5906^T and LMG 15539 were respectively 61.5, 62.9 and 62.7 mol%. The cellular fatty acid compositions of all *Kerstersia* isolates were also determined (shown in Supplementary Table A in IJSEM Online). The *Kerstersia* isolates formed a homogeneous group: the predominant fatty acids were 16:0, 17:0 cyclo, 18:1 ω 7c and summed feature 2 (comprising 14:0 3-OH, 16:1 iso I, an unidentified fatty acid with an equivalent chain-length of 10.928 and/or 12:0 ALDE). All strains examined showed catalase activity but no oxidase, arginine dihydrolase, lysine decarboxylase, ornithine decarboxylase, β -galactosidase, gelatinase, amylase, urease or DNase activity. No reduction of nitrate or nitrite, hydrolysis of aesculin or production of acid or H_2S from triple-sugar-iron agar was observed. All strains examined were capable of growth between 28 and 42 °C and could grow with NaCl concentrations ranging from 0 to 4.5%. Growth in 6% NaCl and resistance to penicillin were strain dependent. All strains examined assimilated the following substrates: acetate, propionate, butyrate, n-valerate,

Fig. 2. Phylogenetic tree based on 16S rDNA sequences showing the position of *Kerstersia* gen. nov. The scale bar represents 10% sequence dissimilarity.

Table 2. DNA–DNA binding values of all strains examined

Asterisks indicate values obtained using the initial renaturation rate method; other values were obtained using the microplate method.

Strain	DNA–DNA binding (%) with strain:		
	1	2	3
1. <i>K. gyiorum</i> LMG 5906 ^T	100		
2. <i>K. gyiorum</i> LMG 15539	91*	100	
3. <i>Kerstersia</i> sp. LMG 5890	26*/38	28*	100
4. <i>Alcaligenes faecalis</i> LMG 1229 ^T	1	–	2
5. <i>B. hinzii</i> LMG 13501 ^T	8	–	6
6. <i>Achromobacter xylosoxidans</i> LMG 1863 ^T	9	–	8
7. <i>Achromobacter piechaudii</i> LMG 1873 ^T	17	–	6
8. <i>Achromobacter ruhlandii</i> LMG 1866 ^T	9	–	4
9. <i>Achromobacter denitrificans</i> LMG 1231 ^T	6	–	9

n-caproate, heptanoate, pelargonate, caprate, succinate, fumarate, DL-lactate, DL-lactate plus methionine, DL-3-hydroxybutyrate, D-malate, L-malate, pyruvate, citrate, phenylacetate, *p*-hydroxybenzoate, D- α -alanine, L- α -alanine, L-phenylalanine, L-tyrosine, L-aspartate, L-glutamate, L-ornithine, L-proline, DL-4-aminobutyrate, 2-aminobenzoate, 4-aminobenzoate, amylamine and DL-kynurenine. None of the strains examined assimilated erythritol, D- or L-arabinose, ribose, D- or L-xylose, adonitol, methyl β -D-xyloside, galactose, D-glucose, D-fructose, D-mannose, L-sorbose, L-rhamnose, dulcitol, inositol, mannitol, sorbitol, methyl α -D-mannoside, methyl α -D-glucoside, *N*-acetylglucosamine, amygdalin, arbutin, aesculin, salicin, maltose, lactose, melibiose, sucrose, trehalose, inulin, D-melezitose, raffinose, starch, glycogen, xylitol, D-turanose, D-lyxose, D-tagatose, D- or L-fucose, D- or L-arabitol, 2- or 3-ketogluconate, oxalate, malonate, malate, glycolate, DL-glycerate, D- or L-tartrate, *meso*-tartrate, benzoate, *o*- or *m*-hydroxybenzoate, D- or L-mandelate, phthalate, isophthalate, terephthalate, glycine, L-serine, L-histidine,

D-tryptophan, trigonelline, L-lysine, L-citrulline, L-arginine, betaine, creatine, DL-3-aminobutyrate, DL-5-aminovaleate, 3-aminobenzoate, urea, acetamide, sarcosine, ethylamine, ethanolamine, benzylamine, diaminobutane, spermine, histamine or glucosamine. Assimilation of glycerol, gluconate, cellobiose, β -gentiobiose, isobutyrate, isovalerate, caprylate, glutarate, adipate, pimelate, suberate, azelate, sebacate, levulinate, 2-ketoglutarate, citraconate, itaconate, mesaconate, aconitate, L-leucine, L-isoleucine, DL-2-aminobutyrate, L-norleucine, L-valine, DL-norvaline, L-threonine, L-cysteine, L-methionine, L-tryptophan, β -alanine, tryptamine and butylamine was strain dependent. The range of MIC values and the MIC₅₀ and MIC₉₀ values of the strains are shown in Supplementary Table B in IJSEM Online.

Isolates LMG 5906^T and LMG 5890 (representatives of the two major protein electrophoretic groups) formed a single and stable phylogenetic lineage. The closest related genera are *Bordetella*, *Achromobacter*, *Alcaligenes* and *Pigmentiphaga*, but 16S rDNA sequence similarities with these genera

were below 96.4, 95.1, 94.8 and 93.5 %, respectively. The stability of the cluster (indicated by a bootstrap value of 100 %) and the relatively low similarity values towards the closest related genera warrant the inclusion of these isolates in a novel genus, for which we propose the name *Kerstesia*. Numerical analysis and visual comparison of the protein profiles suggested that there were two different genomic groups present within the genus *Kerstesia*. Since the similarity of the 16S rDNA of representative isolates of the two protein electrophoretic groups was above 98.3 %, DNA–DNA hybridizations were performed to clarify their relatedness (Stackebrandt & Goebel, 1994). The results from these DNA–DNA hybridization experiments confirmed that the two major protein electrophoretic groups (one containing the strains belonging to cluster I and one containing the strains belonging to clusters II and III) formed two separate genomic species. Biochemically, the two genomic species were indistinguishable. At this time, we propose the formal binomial name *Kerstesia gyiorum* gen. nov., sp. nov. for the isolates belonging to protein electrophoretic clusters II and III. Pending the availability of differential biochemical characteristics, LMG 5890, LMG 5891 and LMG 5892 are classified as *Kerstesia* spp.

Identification of members of the genus *Kerstesia*

Several previous studies have shown that species of the genera *Alcaligenes*, *Achromobacter* and *Bordetella* can be identified using SDS-PAGE of whole-cell proteins (Vancanneyt *et al.*, 1995; Vandamme *et al.*, 1995, 1996). This was confirmed in the present study. In addition, our data also indicated that SDS-PAGE of whole-cell proteins can be used to distinguish the two genomic groups present in the genus *Kerstesia* from each other and from related taxa. Using whole-cell fatty acid analysis, *Kerstesia* can be

separated from other members of the *Alcaligenaceae* by the absence of 12:0 2-OH and by the large relative amounts of 18:1 ω 7c. Biochemically, members of the genus *Kerstesia* are difficult to separate from other members of the *Alcaligenaceae*. In contrast to *Kerstesia* spp., most *Achromobacter* strains are oxidase-positive and assimilate aesculin and meso-tartrate, while most *Bordetella* strains are oxidase-positive and do not assimilate caprate. *P. kullae* does not assimilate phenylacetate but does assimilate adipate. Phenotypic characteristics useful for the differentiation of *K. gyiorum* from related taxa are shown in Table 3.

Taxonomic status of *Alcaligenes denitrificans* Rger and Tan 1983

According to Yabuuchi *et al.* (1998), *Alcaligenes denitrificans* should be classified as a subspecies of *Achromobacter xylosoxidans*, based on intermediate DNA–DNA binding values between the two type strains and the results of 16S rDNA sequence analysis. However, previous work (Kiredjian *et al.*, 1981; Vandamme *et al.*, 1995, 1996) has shown clearly that strains of the two subspecies of *Achromobacter xylosoxidans* can be easily differentiated by DNA–DNA relatedness studies, whole-cell protein and fatty acid analysis as well as by phenotypic characteristics. These conclusions are again substantiated in the present study. It is therefore justified to reclassify *Alcaligenes denitrificans* Rger and Tan 1983 as *Achromobacter denitrificans* comb. nov.

Description of *Achromobacter denitrificans* comb. nov.

The description of *Achromobacter denitrificans* is the same as the description given by Kersters & De Ley (1984) for *Alcaligenes xylosoxidans* subsp. *denitrificans*.

Table 3. Phenotypic characteristics useful for the differentiation of *K. gyiorum* from related taxa

Species: 1, *K. gyiorum*; 2, *Alcaligenes faecalis*; 3, *Alcaligenes defragrans*; 4, *Achromobacter denitrificans*; 5, *Achromobacter ruhlandii*; 6, *Achromobacter xylosoxidans*; 7, *Achromobacter piechaudii*; 8, *B. pertussis*; 9, *B. parapertussis*; 10, *B. bronchiseptica*; 11, *B. avium*; 12, *B. trematum*; 13, *B. hinzi*; 14, *B. holmesii*; 15, *B. petrii*; 16, *P. kullae*. +, Positive; –, negative; v, strain dependent; ND, not determined. Data for *Alcaligenes*, *Achromobacter* and *Bordetella* species were taken from Kersters & De Ley (1984), Pittman (1984), Vandamme *et al.* (1995, 1996), Weyant *et al.* (1995), Yabuuchi *et al.* (1998), Foss *et al.* (1998) and von Wintzingerode *et al.* (2001); data for *P. kullae* were taken from Blmel *et al.* (2001).

Characteristic	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16
Growth at 42 C	+	+	–	+	+	+	+	–	–	+	+	+	+	–	ND	+
Oxidase activity	–	+	ND	+	+	+	+	+	+	+	+	–	+	+	+	+
Assimilation of:																
D-Glucose	–	–	–	–	+	+	–	–	–	–	–	–	–	–	–	–
D-Xylose	–	–	–	–	ND	+	–	–	–	–	–	–	–	–	–	–
Caprate	+	+	ND	+	+	+	–	–	–	v	–	–	+	–	–	ND
Adipate	–	–	–	+	+	+	+	–	–	+	+	+	+	–	+	+
L-Malate	+	+	+	+	+	+	+	–	–	v	+	+	+	–	+	ND
Phenylacetate	+	v	ND	+	+	+	+	–	–	+	+	+	+	–	–	–
Aesculin	–	–	ND	+	ND	+	–	–	–	–	–	+	+	–	–	ND

Description of *Kerstersia* gen. nov.

Kerstersia (Kers.ter's'i.a. N.L. fem. n. *Kerstersia* in honour of K. Kersters, an eminent Belgian microbiologist, for his contributions to polyphasic taxonomy and to the introduction of computerized protein gel electrophoresis).

Gram-negative, small (1–2 µm long), coccoid cells that occur as single units, in pairs or in short chains. Motility is strain dependent. On nutrient agar, colonies are flat or slightly convex with smooth margins with a colour ranging from white to light brown. Catalase activity, but not oxidase, urease or β-galactosidase activity, is observed. Growth is observed at 28 and 42 °C. Additional biochemical characteristics are given above (see Results). MIC values for a number of antibiotics are shown in Supplementary Table B in IJSEM Online. The following fatty acid components are present: 14:0, 14:0 2-OH, 16:0, 17:0 cyclo, 18:0, 18:1ω7c, 19:0 cyclo ω8c, summed feature 2 and summed feature 3. The G + C content is 61.5–62.9 mol%. Strains of this genus have been isolated from various human clinical samples. The type species is *Kerstersia gyiorum*.

Description of *Kerstersia gyiorum* sp. nov.

Kerstersia gyiorum (gy.i.o'rum. Gr. n. *gyion* limb; N.L. gen. n. *gyiorum* from the limbs, referring to the fact that the majority of strains were isolated from human leg wounds).

The description is the same as for the genus. Additional characteristics are the assimilation of caprylate and the absence of assimilation of isobutyrate, isovalerate, glutarate, adipate, pimelate, suberate, azelate, sebacate, levulinate, 2-ketoglutarate, citraconate, itaconate, mesaconate, aconitate, L-leucine, L-isoleucine, L-valine, DL-norvaline, L-threonine, L-cysteine and L-methionine. The G + C content is 62.7–62.9 mol%. The type strain, LMG 5906^T (=API 184-2-84^T=CCUG 47000^T), was isolated from a human ankle wound. Characteristics for the type strain are the same as described above for the species. In addition, the type strain can use L-norleucine. The G + C content of the type strain is 62.9 mol%. All *K. gyiorum* strains reported in this study have been deposited in the BCCM/LMG Bacteria Collection (Laboratorium voor Microbiologie, Gent, Belgium) and the CCUG (Department of Clinical Bacteriology, Göteborg, Sweden).

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