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Identities of *Arthrobacter* spp. and *Arthrobacter*-Like Bacteria Encountered in Human Clinical Specimens[▽]

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After the initial description of *Arthrobacter* spp. isolated from clinical specimens in the mid-1990s, very few further reports on *Arthrobacter* spp. have appeared in the clinical microbiology literature. The aim of the present study was to elucidate the distribution of *Arthrobacter* spp. and *Arthrobacter*-like bacteria encountered in clinical specimens by studying 50 consecutively isolated or received strains of large-colony-forming, whiteish-grayish, non-cheese-like-smelling, nonfermentative gram-positive rods by applying phenotypic methods as well as 16S rRNA gene sequencing. We observed a very heterogenous distribution, with the 50 strains belonging to 20 different taxa and each of 13 strains as a single representative of its particular taxon. Thirty-eight strains represented true *Arthrobacter* strains, 7 strains belonged to the genus *Brevibacterium*, 2 were *Microbacterium* species, and each of 3 single strains was a member of the rarely encountered genera *Pseudoclavibacter*, *Leucobacter*, and *Brachybacterium*, respectively. *A. cumminsii* ($n = 14$) and *A. oxydans* ($n = 11$) were the most frequently found species. The present report describes the first three *A. aurescens* strains isolated from human clinical specimens. Comprehensive antimicrobial susceptibility data are given for the 38 *Arthrobacter* isolates.

Arthrobacter spp. belong to the heterogenous group of coryneform bacteria and had not been reported to have been isolated from human clinical specimens until the mid-1990s (7). This was somewhat surprising, because *Arthrobacter* spp. are the most frequently isolated coryneform bacteria when soil specimens are incubated aerobically, indicating that humans are unceasingly exposed to these bacteria. After the initial descriptions (7, 9), only a few other studies have been published regarding the appearance of *Arthrobacter* spp. in clinical specimens (1, 10, 11, 12, 23). Therefore, we have continued to collect *Arthrobacter* isolates either from our routine clinical laboratory procedures or as reference cultures sent to us. The present report outlines the data pertaining to 50 consecutive *Arthrobacter* or *Arthrobacter*-like strains collected by one of the authors (G. Funke). *Arthrobacter* and *Arthrobacter*-like strains were defined for the purposes of the present study as large-colony-forming, whiteish-grayish, non-cheese-like-smelling, nonfermentative gram-positive rods. The aim of the study was to investigate the identity of *Arthrobacter* and *Arthrobacter*-like strains by use of phenotypic and molecular genetic methods in order to finally reveal the true distribution within clinical *Arthrobacter* isolates. In addition, we determined MICs of a variety of antimicrobials against these bacteria, since limited data exist in the relevant literature on the antimicrobial susceptibility patterns of *Arthrobacter* spp. As a byproduct of our investigations, we describe two new bacterial species, namely, *Arthrobacter sanguinis* sp. nov. and *Brevibacterium ravenspurgense* sp. nov.

MATERIALS AND METHODS

Strains. The 50 strains examined in the present study were isolated in the routine clinical microbiology laboratories of Gärtner & Colleagues Laboratories, Ravensburg, Germany, or were referred to this institution by collaborating laboratories. None of the isolates had been included in any of our previous studies except strains 1369, NML 90-0364, NML 91-0435, NML 92-0385, NML 92-0394, NML 92-0600, and NML 93-0702 (7). The strains had been stored at −20°C in skim milk. For the investigations, strains were grown on Columbia sheep blood agar (SBA) plates (BD, Heidelberg, Germany) and passaged twice on SBA plates at 35°C in ambient air before use.

Phenotypic testing. The techniques applied have been outlined in detail before (8, 23). For assimilation tests of the nonfermenting gram-positive rods, we applied commercial AUX medium (bioMérieux, Marcy l'Etoile, France) to the API 50CH kit (bioMérieux) (5). Reading of the assimilation reactions was done at 48 and 120 h of incubation at 35°C.

Chemotaxonomic investigations. Analyses of cellular fatty acids and the diamino acid of the bacterial cell wall were performed as outlined before (8).

Molecular genetic investigations. Analysis of the complete 16S rRNA gene sequences was performed according to a published protocol (6). Almost complete (>1,350 bp) 16S rRNA gene sequences were determined for each clinical strain by aligning multiple overlapping sequences by use of the Lasergene 5 package (DNASTAR Inc., Madison, WI). Phylogenetic trees were constructed using the neighbor-joining method, included in the MEGA4 suite software (22), based on a comparison of approximately 1,350 nucleotides. Bootstrap values, expressed as percentages of 1,000 replications, are given at each branching point in the figures.

Identification. Strains were identified on the species level when the 16S rRNA gene sequence of the individual strain was >99.0% homologous to the type strain of a certain species (19) and when phenotypic testing did not indicate any aberrant reactions regarding the published data for this particular species.

Antimicrobial susceptibility testing. The CLSI standard for determination and interpretation of antimicrobial MICs for *Corynebacterium* spp. (2) was applied. Briefly, by use of a broth microdilution method, bacterial cells representing an inoculum equivalent to a 0.5 McFarland standard were grown in cation-adjusted Mueller-Hinton broth with lysed horse blood and incubated for up to 48 h. Reading of MICs was done by two independent researchers.

Nucleotide accession numbers. The GenBank accession numbers of the complete 16S rRNA gene sequences of all 50 clinical isolates included in the present study are given in Table 1. The GenBank accession number of the 16S rRNA gene sequence of the *Arthrobacter sanguinis* type strain is EU086805. The GenBank accession number of the 16S rRNA gene sequence of the *Brevibacterium ravenspurgense* type strain is EU086793.

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TABLE 1. Strains included in the present study

Strain collection no.	Patient age (yr), sex[a]	Clinical source	Identification	GenBank accession no.
20 (CCUG 56047)[b]	45, m	Wound swab	*Brevibacterium ravenspurgense* sp. nov.	EU086793
27	59, f	Sinus aspirate	*Brevibacterium otitidis*	EU086795
28	27, f	Deep wound	*Brevibacterium paucivorans*	EU086796
60	53, m	Wound swab	*Brevibacterium casei*	EU086802
84	2, m	Wound swab	*Arthrobacter aurescens*	EU086809
102	39, m	Wound secretion	*Microbacterium foliorum*	EU086781
115	89, f	Wound swab	*Arthrobacter oxydans*	EU086782
120	24, f	Wound swab	*Arthrobacter oxydans*	EU086783
124	34, f	Urine	*Arthrobacter cumminsii*	EU086784
148	78, f	Urine	*Arthrobacter aurescens*	EU086789
219	78, f	Wound swab	*Arthrobacter cumminsii*	EU086794
300	51, f	Blood culture	*Arthrobacter cumminsii*	EU086797
352	53, m	Tracheal secretion	*Arthrobacter cumminsii*	EU086798
379	46, m	Otitis externa	*Brevibacterium casei*	EU086799
448	62, m	Nasal swab	*Microbacterium oxydans*	EU086800
486	45, f	Urine	*Arthrobacter cumminsii*	EU086827
511	62, m	Wound swab	*Brachybacterium* sp.	EU086801
664	65, m	Urine	*Arthrobacter cumminsii*	EU086819
665	74, m	Aortic valve	*Pseudoclavibacter* sp.	EU086820
690	25, m	Blood culture	*Arthrobacter* sp. (uncultured soil bacterium clone AKAU3746)	EU086821
696	66, m	Wound swab	*Brevibacterium otitidis*	EU086822
720	83, m	Wound swab	*Arthrobacter cumminsii*	EU086803
740 (CCUG 44254)	45, f	Cervix	*Arthrobacter cumminsii*	EU086804
741 (CCUG 46407)	63, m	Human blood	*Arthrobacter sanguinis* sp. nov.	EU086805
742 (CCUG 29118)	82, f	Otitis externa	*Arthrobacter cumminsii*	EU086806
743 (CCUG 35230)	27, m	Human tibia	*Leucobacter* sp.	EU086807
744 (CCUG 46391)	88, f	Human blood	*Brevibacterium ravenspurgense* sp. nov.	EU086808
1361	36, f	Urine	*Arthrobacter cumminsii*	EU086785
1366	NK	Urine	*Arthrobacter albus*	EU086786
1369	NK	Vaginal swab	*Arthrobacter oxydans*	EU086823
1378	NK	Urine	*Arthrobacter cumminsii*	EU086787
1391	NK	Otitis externa	*Arthrobacter cumminsii*	EU086788
1515 (CCUG 33745)	47, m	Human blood	*Arthrobacter cumminsii*	EU086790
NML[c] 90-0364	NK	Eye	*Arthrobacter oxydans*	EU086810
NML 91-0435	NK	Eye	*Arthrobacter oxydans*	EU086824
NML 92-0385	NK	Blood culture	*Arthrobacter* sp. strain BS20	EU086825
NML 92-0394	NK	Human blood	*Arthrobacter oxydans*	EU086826
NML 92-0600	NK	Blood culture	*Arthrobacter oxydans*	EU086811
NML 93-0693	NK	NK	*Arthrobacter oxydans*	EU086791
NML 93-0702	NK	NK	*Arthrobacter oxydans*	EU086792
NML 93-0734	NK	NK	*Arthrobacter aurescens*	EU086812
NML 95-0018	NK	Hand wound	*Arthrobacter* sp. strain An16	EU086813
NML 95-0188	28, f	Blood culture	*Arthrobacter* sp. strain 19B	EU086814
NML 98-0077	NK	Urine	*Arthrobacter protophormiae*	EU086815
NML 99-0063	NK	Blood culture	*Arthrobacter albus*	EU086816
NML 99-0140	NK	Blood culture	*Arthrobacter cumminsii*	EU086817
NML 00-0248	NK	Blood culture	*Arthrobacter* sp. strain W8	EU086777
NML 01-0266	NK	Neck abscess	*Arthrobacter oryzae*	EU086778
NML 02-0288	NK	Blood culture	*Arthrobacter oxydans*	EU086779
NML 03-0063	NK	Lung swab at autopsy	*Arthrobacter oxydans*	EU086780

[a] m, male; f, female; NK, not known.
[b] CCUG, Culture Collection University of Gothenburg, Gothenburg, Sweden.
[c] NML, National Microbiology Laboratory.

RESULTS

Table 1 lists all the data of the strains included in the present study. Patient data were available in 30 of 50 cases and showed that 16 patients were males and 14 females. The mean of the ages of the patients was 52.5 years (range, 2 to 89 years). Twelve strains each were isolated either from blood cultures or from wounds, and 8 strains came from urine samples. Five strains were isolated from primarily sterile tissues, and only one strain came from respiratory tract material.

We observed that the 50 strains included in the present study were representatives of 20 different taxa and that each of 13 strains was a single representative of a particular taxon. 16S rRNA gene homologies ranged from 99.03% to 99.93%, with a mean of 99.60%, excluding the two newly defined species (see below) and the members of the genera *Pseudoclavibacter*, *Leucobacter*, and *Brachybacterium* (see below). Thirty-eight strains represented true *Arthrobacter* species, 7 strains belonged to the genus *Brevibacterium*, 2 strains were microbacteria, and each of 3 single strains was a member of the genera *Pseudoclavibacter*, *Leucobacter*, or *Brachybacterium*, respectively. Within the arthrobacters,

TABLE 2. Phenotypic features differentiating *Arthrobacter sanguinis* sp. nov. from its closest phylogenetic neighbors

Feature	Reaction for:			
	A. sanguinis	*A. crystallo-poietes*	*A. cumminsii*	*A. globiformis*
Activity of:				
α-Galactosidase	+	−	−	−
β-Galactosidase	+	−	−	+
N-Acetyl-β-glucosaminidase	+	−	−	−
α-Glucosidase	+	+	−	+
α-Mannosidase	+	−	−	+
Utilization of[a]:				
Amygdaline	+	−	−	−
D-Arabitol	+	−	−	−
L-Arabinose	−	−	−	+
Cellobiose	+	+	−	−
Dulcitol	−	−	−	+
Galactose	+	−	−	+
N-Acetyl-glucosamine	+	−	−	+
Glycerol	+	−	−	+
Inositol	−	+	−	+
Maltose	+	−	−	+
Mannitol	+	+	−	+
Melezitose	−	−	−	+
Melibiose	+	−	−	+
Raffinose	+	−	−	+
Rhamnose	−	−	−	+
Sorbitol	+	−	−	+
Trehalose	+	−	−	+
Turanose	+	−	−	+

[a] Utilization reactions were tested as outlined in reference 5.

A. cumminsii ($n = 14$) and *A. oxydans* ($n = 11$) were the most frequently detected species and together represented half of the strains included in the present study. Three strains were identified as *A. aurescens*. Strain 690 was a representative of the so-far-noncultivated *Arthrobacter* bacterial clone AKAU3746, and strains NML 92-0385, NML 95-0018, and NML 95-0188 were representatives of *Arthrobacter* sp. strains BS20, An16, and 19B, respectively, none of which could be identified on the species level.

Strain 741 was phylogenetically distinct from all of the other valid *Arthrobacter* species and is, therefore, described as a new species, *Arthrobacter sanguinis* sp. nov. (see below). 16S rRNA gene homologies with all other 51 presently valid *Arthrobacter* spp. ranged from 91.8 to 94.7%, with *A. crystallopoietes* being its closest known phylogenetic neighbor. Table 2 gives the results of biochemical reactions differentiating *A. sanguinis* sp. nov. from its closest phylogenetic neighbors, with 16S rRNA gene homology values between 94.5 and 94.7%. Cellular fatty acids were of the branched type, with $C_{15:0ai}$ and $C_{17:0ai}$ predominating, and lysine was the diamino acid of the cell wall, which was compatible with an assignment of the unknown bacterium to the genus *Arthrobacter*. Figure 1 shows the phylogenetic position of *A. sanguinis* with respect to its neighbors, demonstrating that *A. sanguinis* represents a unique deep branch within the genus *Arthrobacter*.

Of the seven *Brevibacterium* strains, two turned out to be *B. casei*, two were *B. otitidis*, and one was *B. paucivorans*. Two further strains could not be assigned to any of the established brevibacterial species. Phylogenetic analyses revealed that the 16S rRNA genes from these two isolates shared homology of only between 93.3 and 95.2% with those of the 19 other presently defined *Brevibacterium* species. Cellular fatty acids were of the branched type, with $C_{15:0ai}$ and $C_{17:0ai}$ predominating, and *meso*-diamino pimelic acid was detected as the diamino acid of the cell wall, which was compatible with an assignment to the genus *Brevibacterium*. The two unknown strains showed 99.7% (1,444 of 1,448 bp) 16S rRNA gene homology and were defined as a new species, *Brevibacterium ravenspurgense* sp. nov. Their phylogenetic relationship with all other presently defined brevibacteria is depicted in Fig. 2.

Two strains turned out to be *Microbacterium oxydans* and *M. foliorum*, respectively, and did not, in these two instances, exhibit yellow pigmentation. One strain each was a member of the genera *Pseudoclavibacter*, *Leucobacter*, and *Brachybacterium*. Those three isolates could not be identified on the species level, since the phenotypic and genetic data of other strains belonging to these genera are presently not comprehensive enough to allow unambiguous assignment to a particular species.

Antimicrobial susceptibility testing of the 38 *Arthrobacter* strains showed that 90.5% of the results fell into the susceptible category, 3.9% into the intermediate category, and 5.5% into the resistant category (Table 3). All antimicrobials tested, except ciprofloxacin and erythromycin, demonstrated good activities against *Arthrobacter* spp.

DISCUSSION

The present report represents the most comprehensive study of *Arthrobacter* and *Arthrobacter*-like strains isolated from clinical specimens published to date. Previous studies had included a maximum of 15 *Arthrobacter* strains (7, 9, 23). That the previous reports examined a relatively small number of strains is not surprising, since, in our experience, arthrobacters are two to three times less frequently isolated from clinical specimens than other nonfermenting coryneforms such as microbacteria or brevibacteria (G. Funke, unpublished observation). Despite the large number of *Arthrobacter* isolates, one limitation of our present study is that the clinical data of the patients were limited, but this lack is very often present in studies coming from reference centers.

The most frequently found *Arthrobacter* species in the present series was *A. cumminsii*. That same result had been published in another report a decade ago (9), and other authors later confirmed this observation (23). *A. oxydans* represented more than 20% of the clinical strains in the present study. In the study of Wauters et al. (23), *A. oxydans* represented 2 of 5 clinical *Arthrobacter* strains. *A. oxydans* has been, at least in some studies, the most frequently found *Arthrobacter* species in specimens from soil (17), which might have been the source from which our clinical strains originated.

The third most frequently found *Arthrobacter* species was *A. aurescens*, which has not been reported before as being isolated from human clinical specimens. *A. aurescens* can be differentiated from the closely related *A. nitroguajacolicus* (99.7% 16S rRNA gene homology) by a negative sucrose utilization reaction (13); such a result was seen for all three clinical strains included in the present study.

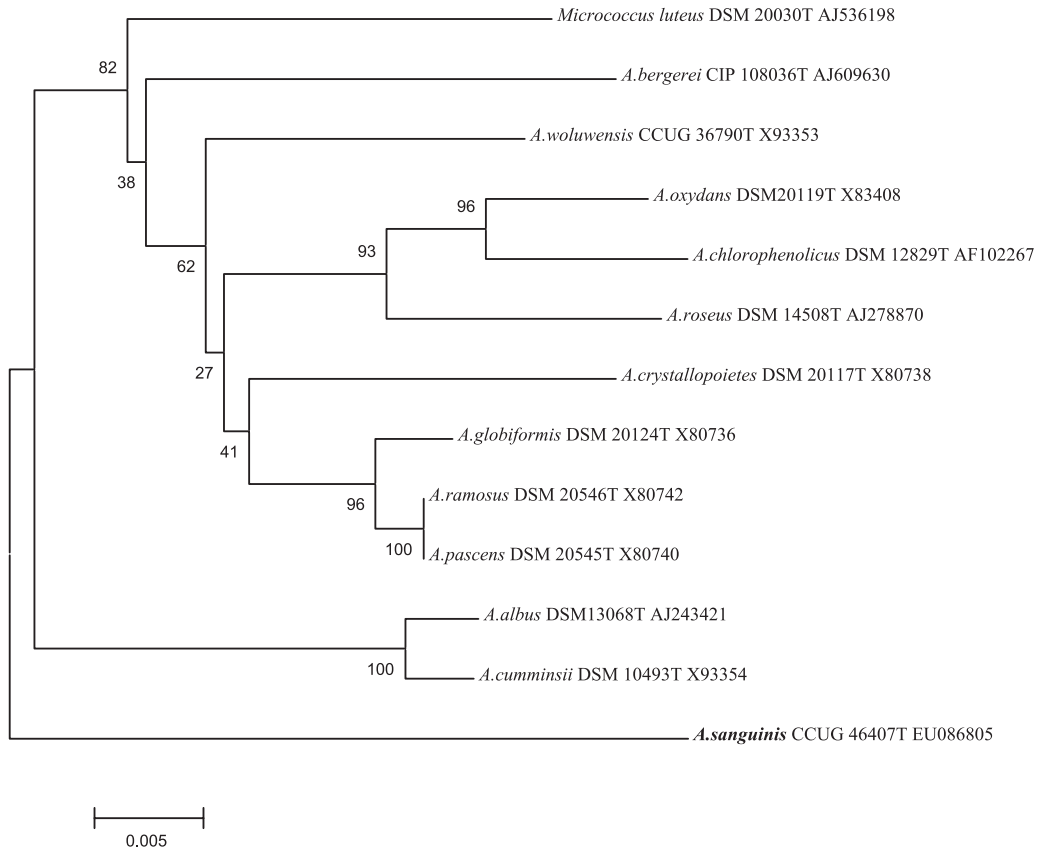

FIG. 1. Phylogenetic tree based on 16S rRNA gene sequences, showing the position of *Arthrobacter sanguinis* sp. nov. within its closest phylogenetic neighbors. *Micrococcus luteus* was used as an outlier. The bar represents percent substitutions.

We observed an enormous heterogeneity within the 50 studied large-colony-forming, whitish-grayish, non-cheese-like-smelling, nonfermentative gram-positive rods, with each of 13 strains representing a single strain belonging to a particular taxon. Strain 690 represented an isolate of the previously noncultured *Arthrobacter* clone AKAU3746; whether this strain is a representative of a new *Arthrobacter* species remains to be elucidated by extensive quantitative DNA-DNA hybridization studies, since strain 690 did not have a 16S rRNA gene divergence percentage of greater than 3% compared to that of its most closely defined phylogenetic neighbor in the present study. In contrast, the 16S rRNA gene divergence value for strain 741 was greater than 5% for all of the *Arthrobacter* spp., which clearly demonstrated that this strain deserves recognition as an individual species (19, 20) (for species description, see below).

Our two *A. albus* strains are only the third and fourth strains of this species that have appeared in the literature (23). We can confirm that the susceptibility to desferrioxamine concentrations of 1,000 μg per disk may allow the separation between *A. cumminsii* (susceptible) and *A. albus* (resistant) (23); of the 14 *A. cumminsii* strains tested in the present study, only strain 219 was resistant to desferrioxamine. Although it is acknowledged that our number of *A. albus* strains (plus the 16S rRNA gene data from the type strain of *A. albus*) was limited, the following 16S rRNA gene signature nucleotides seem to allow a differentiation between the closely related *A. cumminsii* and *A. albus* (based on *A. cumminsii* numbering using the type strain sequence X93354): at position 157, a C in *A. cumminsii* versus a nucleotide deletion in *A. albus*; between positions 161 and 162, a deletion in *A. cumminsii* versus a T in *A. albus*; at position 163, G versus A; at position 504, G versus A; at position 988, C versus T; and at position 1096, T versus G.

The susceptibility patterns of the 38 *Arthrobacter* strains were similar to the data reported before (7, 9), with nearly all isolates exhibiting susceptibility to β-lactams, doxycycline, gentamicin, linezolid, rifampin, and vancomycin. Only the MICs of gentamicin tended to be lower with the broth microdilution method used in the present investigations compared to the results seen with the agar dilution method used in a previous study (7). We did not detect multiresistant *Arthrobacter* isolates, which contrasts with results reported before for *A. woluwensis* (1, 7).

The two *B. casei* strains found in the present study did not exhibit the distinctive cheese-like smell usually detected in *B. casei* (5). So far, only four *B. otitidis* strains have been described in the literature (3, 16, 25) and only seven strains have been described for *B. paucivorans* (24). The two strains representing an unknown *Brevibacterium* (i.e., *B. ravenspurgense*) had a distinctive sticky colony consistency which has not been observed for other true brevibacteria except for some *B. paucivorans* strains. *B. ravenspurgense* can be differentiated from *B. paucivorans* by the following reactions: strains of *B. ravenspurgense* are positive for pyrazinamidase and esterase (C4)

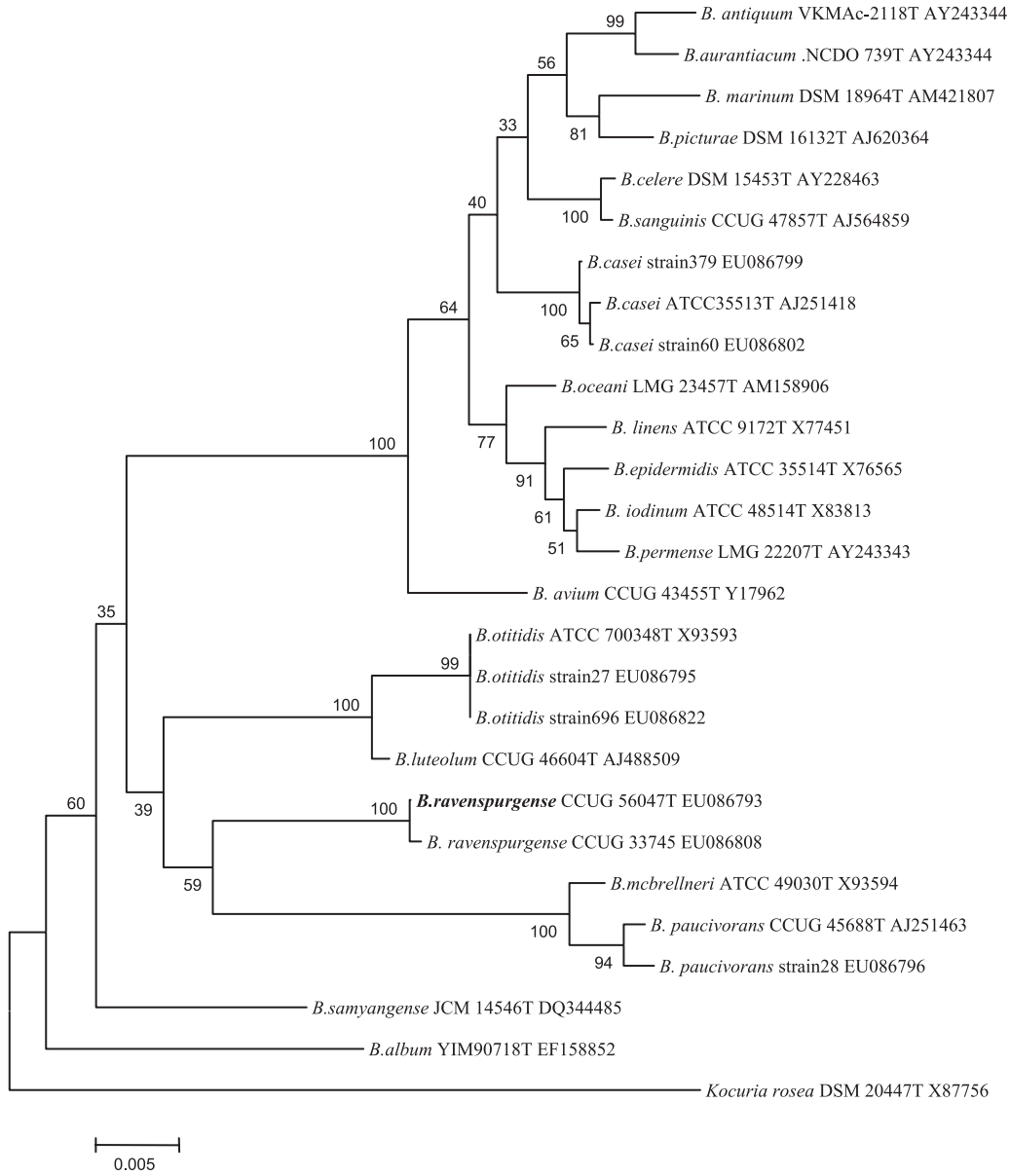

FIG. 2. Phylogenetic tree of the genus *Brevibacterium* based on 16S rRNA gene sequences showing the position of *Brevibacterium ravenspurgense* sp. nov. as well as of the other *Brevibacterium* strains from the present study. *Kocuria rosea* was used as an outlier. The bar represents percent substitutions.

whereas *B. paucivorans* strains are not; in contrast, *B. paucivorans* strains are variable with respect to *N*-acetyl-β-glucosaminidase reactivity whereas the two strains of *B. ravenspurgense* described in the present report were negative for this particular reaction. Like *B. mcbrellneri* and *B. paucivorans* (16, 24), the two unknown *Brevibacterium* strains did not utilize any of the carbohydrates in the test system used whereas the majority of the other brevibacteria are quite reactive in this test system (5). Interestingly, we did not observe gelatinase activity for the unknown *Brevibacterium* strains whereas nearly all other brevibacteria express this enzyme activity (5, 24).

When nonfermenting microbacteria do not exhibit yellow pigmentation, they can be easily confused with *Arthrobacter* spp. In the present series, 4% of the strains represented nonfermenting *Microbacterium* strains. It is noteworthy that two former *Arthrobacter* species ("*Arthrobacter flavescens*" and "*Arthrobacter terregens*") have been reclassified as *Microbacterium* species (see http://www.bacterio.cict.fr/a/arthrobacter.html), indicating the close phenotypic relationship between these two genera. Furthermore, it should be mentioned that *Arthrobacter* and *Microbacterium* are the two largest genera within the nonfermenting coryneform bacteria, with each presently comprising more than 50 valid species.

This paper reports only the fourth *Pseudoclavibacter* strain isolated from humans. The type species of the genus had been previously designated "*Brevibacterium helvolum*," and the genus *Zimmermannella* is a later synonym of *Pseudoclavibacter* (14, 15). Up to now, *Leucobacter* strains have not been re-

TABLE 3. Antimicrobial susceptibility patterns of *Arthrobacter* strains ($n = 38$)[a]

Antimicrobial agent	MIC (μg/ml)			No. (%) of isolates in indicated susceptibility category		
	Range	50%	90%	Susceptible	Intermediate	Resistant
Cefotaxime	≤0.03 to 8	0.12	1	36 (95)	1 (3)	1 (3)
Ciprofloxacin	0.12 to 16	2	4	16 (42)	13 (34)	9 (24)
Doxycycline	≤0.06 to 16	0.12	1	37 (97)	0 (0)	1 (3)
Erythromycin	0.03 to >32	0.12	32	32 (84)	0 (0)	6 (16)
Gentamicin	0.25 to 16	1	4	36 (95)	1 (3)	1 (3)
Linezolid	0.12 to 2	1	2	38 (100)	0 (0)	0 (0)
Meropenem	≤0.03 to 2	0.25	1	38 (100)	0 (0)	0 (0)
Penicillin	≤0.03 to >64	0.12	0.5	36 (95)	0 (0)	2 (5)
Rifampin	≤0.015 to 4	≤0.015	0.25	37 (97)	0 (0)	1 (3)
Vancomycin	0.25 to 2	0.5	1	38 (100)	0 (0)	0 (0)

[a] The *Arthrobacter* strains listed in Table 1 were tested.

ported to have been isolated from human clinical material (14, 18, 21). In the relevant literature, only two *Brachybacterium* strains isolated from humans have appeared so far, both of which exhibited fermentative metabolism (4).

For the routine clinical laboratory, we recommend the use of molecular identification techniques (e.g., full-length 16S rRNA gene sequencing) in order to identify clinically relevant large-colony-forming, whitish-grayish, non-cheese-like-smelling, nonfermentative gram-positive rods because of the great degree of heterogeneity within this group of bacteria as shown in the present study.

***Arthrobacter sanguinis* sp. nov.** *Arthrobacter sanguinis* (san′gui.nis. L. masc. gen. n. *sanguinis* of blood, indicating that the bacterium was isolated from a blood culture).

The cells are coryneform bacteria without irregular branching, and spores are not formed. The organism is obligately aerobic. The colonies are whitish-grayish, slightly convex, of creamy texture, and up to 2 mm in diameter after 24 h of incubation at 35°C on Columbia SBA plates. Activities of the following enzymes are detected: catalase, acid phosphatase, alkaline phosphatase, esterase (C4), esterase lipase (C8), α-galactosidase, β-galactosidase, gelatinase, *N*-acetyl-β-glucosaminidase, α-glucosidase, leucine arylamidase, α-mannosidase, pyrazinamidase, and trypsin. Activities of α-chymotrypsin, cystine arylamidase, α-fucosidase, β-glucosidase, β-glucuronidase, lipase (C14), nitrate reductase, naphthol-AS-BI-phosphohydrolase, urease, and valine arylamidase are not observed. The bacterium is capable of utilizing *N*-acetylglucosamine, amygdalin, D-arabitol, cellobiose, fructose, galactose, gentiobiose, glucose, glycerol, maltose, mannitol, mannose, melibiose, potassium gluconate, potassium 2-ketogluconate, raffinose, sucrose, sorbitol, trehalose, and turanose as carbon sources. The type strain did not utilize adonitol, D-arabinose, L-arabinose, L-arabitol, arbutin, dulcitol, erythritol, fucose, methyl-α-D-glucopyranoside, glycogen, inositol, inulin, potassium 5-ketogluconate, lactose, lyxose, methyl-α-D-mannopyranoside, melezitose, rhamnose, ribose, salicin, sorbose, starch, tagatose, methyl-β-D-xylopyranoside, xylitol, or xylose. Lysine is the diamino acid of the peptidoglycan, and $C_{15:0ai}$ and $C_{17:0ai}$ are the predominant cellular acid acids. The type strain is CCUG 46407 and has been deposited in the Culture of the University of Gothenburg, Sweden, and as strain DSM 21259 in the German Collection of Microorganisms and Cell Cultures.

***Brevibacterium ravenspurgense* sp. nov.** *Brevibacterium ravenspurgense* (ra.vens.pur.gen′se. N.L. adj. from Ravenspurgum, Latin name of the town of Ravensburg, Germany, where the type strain of this species was isolated).

Cells are coryneform bacteria without irregular branching, and spores are not formed. The organism is obligately aerobic. The colonies are whitish-grayish, slightly convex, have a sticky consistency, and are up to 2 mm in diameter after 24 h of incubation at 35°C on Columbia SBA plates. Activities of the following enzymes are detected: catalase, esterase (C4), esterase lipase (C8), leucine arylamidase, naphthol-AS-BI-phosphohydrolase, and pyrazinamidase. Activities of the following enzymes could not be observed: acid phosphatase, α-chymotrypsin, α-fucosidase, α-galactosidase, β-galactosidase, gelatinase, *N*-acetyl-β-glucosaminidase, α-glucosidase, β-glucosidase, β-glucuronidase, lipase (C14), α-mannosidase, nitrate reductase, pyrrolidonyl arylamidase, urease, and valine arylamidase. Activities of alkaline phosphatase and trypsin are variable. The organism does not utilize carbohydrates in the system described by Funke and Carlotti (5). *meso*-Diamino pimelic acid is the diamino acid of the peptidoglycan, and $C_{15:0ai}$ and $C_{17:0ai}$ are the predominant cellular fatty acids. The type strain is CCUG 56047 and has been deposited in the Culture of the University of Gothenburg, Sweden, and as strain DSM 21258 in the German Collection of Microorganisms and Cell Cultures.

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