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Phenotypic Description and Antimicrobial Susceptibilities of *Aerococcus sanguinicola* Isolates from Human Clinical Samples

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This report describes the clinical sources and phenotypic characterization of 16 isolates of *Aerococcus sanguinicola*. Sixteen conventional tests were used to describe and differentiate the 16 isolates of *A. sanguinicola* from 30 strains of *Aerococcus viridans*, 27 strains of *Aerococcus urinae*, and a single strain each of *Aerococcus christensenii* and *Aerococcus urinaehominis*. The phenotypic characterizations of the type strains for each species and 14 *A. sanguinicola* isolates were also compared in the two reference laboratories. *A. sanguinicola* are catalase-negative, vancomycin-susceptible, gram-positive cocci arranged in clusters and tetrads, as are all *Aerococcus* species except *A. christensenii* (which is arranged in short chains). All 16 isolates of *A. sanguinicola* were leucine aminopeptidase and pyrrolidonylarylamidase positive, which is unique to this species among the aerococci. All *A. sanguinicola* isolates grew in broth containing 6.5% NaCl, hydrolyzed hippurate, and were variable in the bile-esculin test. None of the isolates deaminated arginine or were Voges-Proskauer positive. The type strain of *A. sanguinicola* was isolated from a blood culture of a patient living in Denmark. Seven additional isolates were from patients living in Canada, all with urinary tract infections (six were female). Eight isolates were from patients living in five different states in the United States; five were from patients with urinary tract infections, and three were from blood cultures of one patient each with pneumonia, suspected endocarditis, and unknown clinical conditions. The antimicrobial susceptibility patterns were unremarkable; all isolates tested were susceptible to penicillin, amoxicillin, cefotaxime, cefuroxime, erythromycin, chloramphenicol, vancomycin, quinupristin-dalfopristin (Synercid), rifampin, linezolid, and tetracycline. Six of the 15 cultures were resistant to ciprofloxacin and levofloxacin, but all 15 strains were susceptible to sparfloxacin. High-level resistance was detected for meropenem (2 strains) and trimethoprim-sulfamethonazole (1 strain). Intermediate resistance was detected for trimethoprim-sulfamethoxazole (10 strains) and clindamycin (3 strains).

Aerococci are gram-positive cocci (GPC) that are arranged in tetrads and clusters. No chains are observed when Gram stains are prepared from growth in broth or agar cultures. Other genera that are GPC and catalase-negative that form clusters or tetrads (*Alloiococcus*, *Dolosigranulum*, *Gemella*, *Helcococcus*, *Pediococcus*, and *Tetragenococcus*) are differentiated from the *Aerococcus* species by a combination of phenotypic characteristics (9). Prior to 1992, only a single species, *Aerococcus viridans*, was included in the genus. Since 1992, four additional species have been described: *Aerococcus urinae*, *Aerococcus sanguinicola*, *Aerococcus christensenii*, and *Aerococcus urinaehominis*. Prior to the description of the new species, *Aerococcus viridans* was the only non-chain-forming GPC that was pyrrolidonylarylamidase (PYR) positive and leucine aminopeptidase (LAP) negative. The description of the new species *Aerococcus urinae* complicated the differentiation of the *Aerococcus* genus from the other non-chain-forming GPC because *A. urinae* cultures gave reactions in PYR and LAP opposite to those of *A. viridans* (1). With the description of additional species of aerococci, *A. sanguinicola* (14), *A. christensenii* (8), and *A. urinaehominis* (13), the differentiation of the members of the genus *Aerococcus* from the other non-chaining-forming GPC became even more difficult. The descriptions of the latter three species were made with a single strain each and based on the uniqueness of their 16S rRNA gene sequences. Phenotypic testing of a single strain is meager evidence for establishing schemes for the identification of bacterial species. It is the intent of this communication to establish a scheme for the differentiation and identification of *A. viridans*, *A. urinae*, and *A. sanguinicola*. We also wished to evaluate commercially available products distributed for the identification of GPC for the capacity to identify *A. sanguinicola*. Since none of these products have this species in their data banks, the correct identification would be "unacceptable profile (or identification)," "unidentified," or "no match." Identification of *A. sanguinicola* strains as any other bacterium would be an incorrect answer and a reflection of the nonspecific nature of the data bank. A third goal of this study was to determine if the antimicrobial susceptibility patterns of *A. sanguinicola* are different from those of related species.

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MATERIALS AND METHODS

Unidentified, catalase-negative, non-chain-forming GPC were retrieved from culture collections at the Centers for Disease Control and Prevention, Atlanta, Ga. (CDC), and the National Center for Streptococcus, Edmonton, Alberta,

TABLE 1. Comparison of conventional test results in two reference laboratories for the identification of *A. sanguinicola*[a]

Test	Result in indicated laboratory for:																									
	SS-1647		2119-98, 98R46		3938-99		453-00, 99R1186		454-00, 99R1705		458-00, 99R1186		3959-00, 00R805		848-01		1027-01		2335-01		2860-01		5091-01		910-02, R1747		912-02, R2447	
	CDC	NCS	CDC	NCS	CDC	NCS	CDC	NCS	CDC	NCS	CDC	NCS	CDC	NCS	CDC	NCS	CDC	NCS	CDC	NCS	CDC	NCS	CDC	NCS	CDC	NCS
PYR	+	+	+	+	+		+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
LAP	+	+	+	+	+		+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
BE	+	−	+	−	+		+	−	+	−	+	−	(+)	−	+	+	+	+	+	+	+	−	+	−	+	−	+	−
NaCl	+	+	+	+	+		+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Esc	+	+	+	−	+		+	+	+	−	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Arg	+	+	−	−	−		−	−	−	−	−	−	−	−	−	−	−	−	−	−	−	−	−	−	−	−	−	−
VP	−	−	−	−	−		−	−	−	−	−	−	−	−	−	−	−	−	−	−	−	−	−	−	−	−	−	−
Hip	+	+	+	+	+		+	+	+	+	+	+	+	+	+	+	+	+	−	+	+	+	+	+	+	+	+	+
Lac	−	−	−	−	−		−	+	−	−	+	+	−	+	+	−	+	−	−	−	−	+	+	+	+	+		
Mal	+	+	+		+		+				+	+	+		+		+		+		+		+		+			
Man	−	−	−	−	−		−	−	−	−	−	−	−	−	−	−	−	−	−	−	−	−	−	−	−	−	−	−
Rib	+	+	+	+	+		+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Suc	+	+	+	+	+		+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Tre	+	+	+	+	+		+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	−	+	+	+	+	+	+

[a] Abbreviations: PYR, pyrrolidonylaryl aminidase; LAP, leucine aminopeptidase; BE, bile-esculin reaction; NaCl, growth in broth containing 6.5% NaCl; Esc, hydrolysis of esculin; Arg, deamination of arginine; VP, Voges-Proskauer reaction; Hip, hydrolysis of hippurate; Lac, Mal, Man, Rib, Suc, and Tre, acidification of lactose, maltose, mannitol, ribose, sucrose, and trehalose broths, respectively. (+), 85% or more of the strains tested positive; −, 15% or fewer of the strains tested positive.

Canada (NCS). Cultures were received at these two national reference laboratories from state and provincial health departments from both countries. Reference and type strains for all *Aerococcus* species were obtained from the American Type Culture Collection (Manassas, Va.) or the Culture Collection of the University of Göteborg (Göteborg, Sweden).

Cultures were tested in both reference laboratories in a battery of conventional tests as previously described (9). Fourteen strains presumptively identified as *A. sanguinicola* were exchanged and tested in conventional tests and in commercial products designed to identify GPC.

All strains were tested for reaction in the AccuProbe *Enterococcus* culture identification test (GenProbe Inc., San Diego, Calif.). Five strains were sent to David Collins and Paul Lawson, University of Reading, Reading, United Kingdom, for 16S rRNA sequencing and to Enevold Falsen, University of Göteborg, for whole-cell protein analysis.

All strains were tested in the following commercially available identification systems: Rapid ID 32 Strep (ID 32), the API 20 Strep systems, and the Vitek gram-positive identification system. Cards were purchased from bioMerieux-Vitek (Hazelwood, Mo.) and tested on the Vitek system with analysis software version AMS-R07.01.

Rapid Pos ID panels were purchased from Dade MicroScan, Inc. (Sacramento, Calif.), and tested with the MicroScan WalkAway system, analysis software version 22.28. Cellular fatty acid analysis was performed with the Sherlock microbial identification (MIDI) system, software version 3.10, and the Clin40 library (Microbial ID, Inc., Newark, Del.). A BBL Crystal GP gram-positive identification system was purchased from Becton Dickinson Microbiology Systems, Sparks, Md. The codes generated by each system were sent to the manufacturer for species identification.

MICs were determined by methods described by the National Committee for Clinical Laboratory Standards (NCCLS) for testing streptococci other than *Streptococcus pneumoniae* (16). Seventeen antimicrobial agents with various concentrations were tested in customized panels (PML Microbiologicals, Wilsonville, Oreg.) by using the microdilution method in Mueller-Hinton broth supplemented with 3% lysed horse blood. See Table 5 for antibiotics and concentrations that were tested.

RESULTS

The comparison of conventional tests used in the CDC and NCS laboratories to characterize 14 strains of *A. sanguinicola* is shown in Table 1. Disagreement between laboratories was observed with the bile-esculin (BE) test: the CDC laboratory recorded all cultures as positive, but the NCS laboratory recorded only five cultures as positive. Also, the NCS laboratory observed that all cultures were positive for hydrolysis of hippurate, but the CDC laboratory observed that 1 of the 14 cultures was negative. Trehalose was fermented by all isolates in tests at the CDC laboratory, but 1 of 14 isolates was negative at the NCS laboratory. Reactions in the other eight tests matched perfectly between laboratories.

Since the cultures identified as *A. sanguinicola* had phenotypic characteristics similar to those of the enterococci, i.e., positive reactions in PYR, LAP, BE, and NaCl tests, we tested all cultures with the AccuProbe *Enterococcus* identification test. All strains were negative for *Enterococcus* identification.

The five cultures sent to P. Lawson and D. Collins in England had 16S rRNA sequences highly similar to that of the type strain of *A. sanguinicola.* The same five strains sent to E. Falsen in Sweden had whole-cell protein profiles highly similar to that of the type strain of *A. sanguinicola*. These results indicate that the phenotypic characteristics of the cultures generated by the CDC and NCS laboratories with conventional tests were representative of the species *A. sanguinicola*.

Since *A. viridans* is listed in the database for all six commercial systems, the correct identification should be *A. viridans* for the type strain and two very well characterized reference strains of *A. viridans*. The data from testing six systems with the

three strains are given in Table 2. ID 32 was tested in both CDC and NCS laboratories, and it identified all three strains as *A. viridans* but not in the same laboratory. The type strain of *A. viridans*, SS-1251, was erroneously identified as *Streptococcus acidominimus* at the NCS but correctly identified in the CDC laboratory. The Vitek system did not identify any of the three strains correctly. The MicroScan system identified all three strains as *A. viridans*. The MIDI system identified one strain, not the type strain of *A. viridans*, as *A. viridans*. The Crystal system correctly identified two reference strains but not the type strain. The API 20 Strep system identified all three strains correctly, but two of the strains were identified with low discrimination. Identification codes for *A. urinae* are included only in the Crystal system. The type strain for this species (SS-1320) was correctly identified by the Crystal system. Since identification codes for *A. urinae* are not included in the other five systems, the correct identification should be "unacceptable profile" or "unidentified." The ID 32 and Vitek systems gave these answers, but both the MicroScan and MIDI systems gave erroneous identifications as *Gemella morbillorum* and *Streptococcus pyogenes*, respectively (low similarity index). The API 20 Strep system incorrectly identified the type strain of *A. urinae* as *S. acidominimus*. Identification codes are not included in any of the data banks for *A. sanguinicola*, *A. christensenii*, or *A. urinaehominis*. Only 2 of 21 strains were correctly identified as "unacceptable profile" or "unidentified." Five of the 21 identifications were *A. viridans* at various percentages of likelihood (Table 2).

Ten cultures of *A. sanguinicola* were tested in all commercial rapid identification systems (Table 3). Since this species is not included in the databases for these systems, the correct identification should be "unacceptable profile," "unidentified," or "no match." Identifications provided by the manufacturer for the profile codes provided by the CDC and NCS laboratories differed somewhat for ID 32. Nine of 10 cultures were correctly identified as "unacceptable profile" by the CDC laboratory (one was identified as *A. viridans*), but only five of the cultures were correctly identified from the data provided by the NCS laboratory (four were identified as *A. viridans*, three as "doubtful profile," and one as *S. acidominimus* with low discrimination). The Vitek system correctly reported four cultures of *A. sanguinicola* as "unidentified" but incorrectly identified one as *A. viridans* and five as other species or genera of bacteria (Table 3). The MicroScan system correctly reported two cultures of *A. sanguinicola* as unidentified but incorrectly identified two as *A. viridans* and six as other species or genera of bacteria. The MIDI system failed to correctly report any of the 10 cultures as unidentified: 3 cultures were identified as *A. viridans*, and 7 were identified as other bacterial species or genera, and all similarity indices were <0.5 (Table 3). The Crystal system correctly identified eight cultures of *A. sanguinicola* as "unacceptable identification" and the other two as *Corynebacterium renale*. The API Strep system identified 8 of the 10 cultures of *A. sanguinicola* as *A. viridans* and one each as *Streptococcus bovis* and *S. acidominimus*.

Table 4 lists the conventional tests that can be used to identify the five species of *Aerococcus*, *Dolosigranulum pigrum*, *Helcococcus kunzii*, and *Tetragenococcus halophilus*. Identification of *A. christensenii*, *A. urinaehominis*, and *T. halophilus* should be approached with caution, since biochemical data for all of

TABLE 2. Identification of type strains of *Aerococcus* species by commercial identification systems

Organism	ID with[a]:						
	ID 32		Vitek	MicroScan	MIDI[b]	Crystal	Api 20 Strep
	NCS	CDC					
SS-1251, *A. viridans* ATCC 11563	*S. acidominimus* (86%)	*A. viridans* (97.9%) (G)	Unidentified	*A. viridans* (78.5%)	No match	Unacceptable ID	*A. viridans* (69.2) (LD)
SS-1677, *A. viridans* ATCC 700406	*A. viridans* (99.9%)	Unacceptable profile	*Streptococcus uberis* (82%)	*A. viridans* (99.99%)	*Pedobacter* (0.560)	*A. viridans* (99.96%)	*A. viridans* (71.1) (VG genus)
SS-1280, *A. viridans* ATCC 10400	*A. viridans* (97.6%) (DP)	Unacceptable profile	Unidentified	*A. viridans* (99.99%)	*A. viridans* (0.560)	*A. viridans* (99.99%)	*A. viridans* (99.4) (VG)
SS-1320, *A. urinae* NCFB 2893	Unacceptable profile	Unacceptable profile	Unidentified	*G. morbillorum* (99.9%)	*S. pyogenes* (0.283)	*A. urinae* (96.88%)	*S. acidominimus* (99.0) (VG)
SS-1647, *A. sanguinicola*	*S. acidominimus*; *A. viridans* (LD)	*S. acidominimus* (69.2%); *A. viridans* (30.6%) (LD)	*S. acidominimus* (82%)	*G. morbillorum* (87%)	*S. equinus* (0.220)	*Staphylococcuswarneri* (95.04%)	*A. viridans* (48.1) (LD)
SS-1564, *A. christensenii*	*S. acidominimus* (90%)	Unacceptable profile	*S. sanguinis* (90%)	*S. sanguinis* (99.99%)	*Arcanobacteriicum hemolyticum* (0.254)	*Streptococcus vestibularis* (99.99%)	*S. sanguinis* (93.7) (G)
SS-1678, *A. urinaehominis*	*A. viridans* (53.9%); *S. acidominimus* (46%) (LD)	*A. viridans* (53.5%); *S. acidominimus* (46%) (DP)	*G. morbillorum* (92%)	Unidentified	*A. viridans* (0.110)	*Staphylococcus haemolyticus* (93%)	*A. viridans* (96.6) (G)

[a] Unless otherwise noted, data in parentheses indicate probability of identification provided by the manufacturer (percentages) and/or a qualitative result specific to the given test (abbreviations). Abbreviations: ID, identification; G, good identification; LD, low discrimination; DP, doubtful profile; VG, very good identification.

[b] Where a value is given in parentheses, it is the similarity index. A value of >0.5 is considered a good identification.

TABLE 3. Identification of clinical isolates of *A. sanguinicola* by commercial identification systems

Strain ID	ID with:[a]						
	ID-32		Vitek	MicroScan	MIDI[b]	Crystal	API-20 Strep
	NCS	CDC					
2119-98, 98R46	*A. viridans* (93%) (DP)	Unacceptable profile	*Streptococcus intermedius* (54%)	*Streptococcus milleri* group (99.9%)	*Streptococcus equinus* (0.257)	Unacceptable ID	*A. viridans* (81.6) (LD)
3938-99	*A. viridans* (99.9%)	Unacceptable profile	Unidentified	*Streptococcus salivarius* group (72%)	*S. equinus* (0.333)	Unacceptable ID	*A. viridans* (61.9) (LD)
453-00, 99R1186	Unacceptable profile	Unacceptable profile	Unidentified	*S. sanguinis* (98%)	*S. pyogenes* (0.158)	*C. renale* (87.49%)	*S. acidominimus* (69.1) (LD)
454-00, 99R1705	*A. viridans* (93%) (DP)	Unacceptable profile	Unidentified	*A. viridans* (97%)	*S. equinus* (0.139)	Unacceptable ID	*A. viridans* (81.6) (LD)
458-00, 00R1514	Unacceptable profile	Unacceptable profile	*Erysipelothrix rhusiopathiae* (99%)	*G. morbillorum* (99.9%)	*S. bovis* (0.355)	Unacceptable ID	*A. viridans* (58.9) (LD)
3959-00, 00R805	*S. acidominimus* (54%) (LD)	Unacceptable profile	*Erysipelothrix rhusiopathiae* (99%)	*S. milleri* group (72%)	*A. viridans* (0.182)	*C. renale* (87.49%)	*A. viridans* (90.5) (G)
848-01	Unacceptable profile	Unacceptable profile	*Aerococcus* (99%)	*Streptococcus mitis* group (50%)	*S. bovis* (0.148)	Unacceptable ID	*A. viridans* (8.9) (LD)
1027-01	Unacceptable profile	Unacceptable profile	*Streptococcus constellatus* (74%)	*A. viridans* (97%)	*S. equinus* (0.436)	Unacceptable ID	*A. viridans* (81.6) (LD)
2335-01	Unacceptable profile	*A. viridans* (93%) (DP)	Unidentified	Unidentified	*A. viridans* (0.139)	Unacceptable ID	*S. bovis* II (89.2) (LD)
2860-01	*A. viridans* (93%) (DP)	Unacceptable profile	*S. equinus* (85%)	Unidentified	*A. viridans* (0.242)	Unacceptable ID	*A. viridans* (81.6) (LD)

[a] Unless otherwise noted, data in parentheses indicate probability of identification provided by the manufacturer (percentages) and/or a qualitative result specific to the given test (abbreviations). Abbreviations: ID, identification; DP, doubtful profile; LD, low discrimination; G, good identification.
[b] Where a value is given in parentheses, it is a similarity index. A value of >0.5 is considered a good identification.

these species are based on the description of one or two isolates. The data listed in this table for *A. viridans* and *A. urinae* were taken from the results for GPC identified within the past 7 years in the *Streptococcus* laboratory at the CDC. Sixteen cultures were confirmed as *A. sanguinicola* by phenotypic characteristics determined by conventional testing. Only one has not been tested in both laboratories.

Seven cultures were obtained from Canadian patients (six females and one male) with urinary tract infections; all but one of these patients were ≥79 years old. Eight cultures were obtained from patients residing in the United States: five (two males and three females, all more than 69 years of age) had urinary tract infections, and three had positive blood cultures, one with sepsis, one with suspected endocarditis, and the other without a diagnosis. The remaining isolate was the type strain isolated from a blood culture of a patient living in Denmark (14).

Table 5 lists the antimicrobial susceptibilities of 15 strains of *A. sanguinicola* to 17 different drugs. One culture failed to grow in the MIC panels. All cultures were susceptible to penicillin, amoxicillin, cefotaxime, cefuroxime, erythromycin, chloramphenicol, vancomycin, quinupristin-dalfopristin (Synercid), rifampin, linezolid, and tetracycline. Most of the susceptibility values were equal to or less than the lowest dilution of drug tested. Six of the 15 cultures were resistant to ciprofloxacin and levofloxacin, but all 15 strains were susceptible to sparfloxacin. Intermediate or high resistance to meropenem, clindamycin, trimethoprim-sulfamethoxazole and chloramphenicol was detected in a few strains (one to three cultures for each drug). The MIC interpretation as susceptible or resistant was obtained by using the NCCLS criteria for streptococci other than *S. pneumoniae* except for trimethoprim-sulfamethoxazole (SXT) and rifampin; criteria for *S. pneumoniae* were used for these two drugs (16).

DISCUSSION

Whole-cell protein analysis and 16S rRNA gene sequencing were performed for 5 of the 16 strains to validate the phenotypic description. The 14 conventional tests listed in Table 4 along with the Gram stain (cellular arrangement) and catalase test can be used to phenotypically identify *A. sanguinicola*. These tests have proven to be reproducible in two laboratories, so that the same identification should be attainable in any laboratory performing the same tests. Other species, such as *A. viridans*, *A. urinae*, *D. pigrum*, and *H. kunzii*, can also be identified with the same tests. The other catalase-negative, non-chain-forming GPC, such as *Pediococcus*, *Gemella*, and *Alloiococcus*, can be selected out from this identification scheme with only a few additional tests, such as those described in reference 7. Briefly, the pediococci are the only catalase-negative, non-chain-forming GPC that are vancomycin resistant, and the *Gemella* species are the only catalase-negative, non-chain-forming GPC that fail to grow in 6.5% NaCl. Alloiococci fail to grow on sheep blood agar plates and have been found only in ear fluids. *A. sanguinicola* is not related to the subspecies of *A. urinae* described by Christensen et al. (7). The phenotypic characteristics of each of the subspecies of *A. urinae* and *A. sanguinicola* are distinct.

It is disappointing that the commercially available systems

TABLE 4. Phenotypic characteristics of catalase negative, non-chain-forming GPC[a]

Test	Result[b] (% of strains) for:							
	A. viridans (30)[d]	*A. urinae* (27)	*A. sanguinicola* (16)	*A. christensenii* (1)[c]	*A. urinaehominis* (1)[c]	*D. pigrum* (26)	*H. kunzii* (21)	*T. halophilus* (2)[c]
Lactose	v (66)	− (0)	v (36)	−	−	− (0)	− (0)	−
Maltose	v (77)	− (7)	+ (100)	−	+	− (0)	− (0)	v
Mannitol	v (30)	+ (93)	− (6)	−	−	− (0)	− (0)	v
Ribose	v (58)	+ (89)	+ (100)	−	v	− (0)	− (0)	+
Sucrose	+ (87)	+ (100)	+ (100)	−	+	− (0)	− (0)	+
Trehalose	+ (87)	− (0)	+ (100)	−	−	− (0)	− (0)	+
Arginine	− (0)	− (0)	− (6)[e]	−	−	− (0)	− (0)	+
Esculin	+ (90)	v (11)	+ (100)	−	+	+ (92)	+ (100)	+
PYR	+ (100)	− (0)	+ (100)	−	−	+ (96)	+ (100)	+
LAP	− (0)	+ (100)	+ (100)	+	−	+ (100)	− (0)	+
BE	v (80)	− (0)	+ (100)[f]	−	−	− (0)	− (0)	+
NaCI	+ (100)	+ (100)	+ (100)	−	+	+ (100)	+ (100)	+
Hippurate	v (75)	+ (93)	+ (94)	+	+	− (0)	− (0)	−
Voges-Proskauer	− (10)	− (0)	− (0)	+	−	− (0)	− (0)	+

[a] All *Pediococcus* species are vancomycin resistant, differentiating them from all other species listed above and below. All *Gemella* species fail to grow in broth containing 6.5 NaCl, which differentiates these species from all other genera and species listed above and below. All *Alloiococcus otitis* strains to date have been isolated from inner ear infections, and they fail to grow in thioglycollate broth.
[b] v, variable reaction (10 to 85% positive); +, ≥86% positive; −, <9% positive.
[c] Reactions for *A. christensenii*, *A. urinaehominis*, and *Tetragenococcus halophilus* from CDC lab with conventional testing of only the type strains.
[d] Numbers in parentheses after the species names are numbers of isolates.
[e] Only the type strain of *A. sanguinicola* is positive in arginine.
[f] Only 42% of 12 strains tested were positive in the BE test in the NCS laboratory.

are not consistently capable of identifying reference strains of *A. viridans*. It is apparent that not all systems have used the same reference strains to document their database. It is also apparent that some of the systems are not restrictive enough to accurately identify *Aerococcus* species. It is probable that the identification was not accurate for the strains used to establish the databases, which results in erroneous matches in the database. Among the commercially available rapid identification systems, the ID 32 and Crystal systems would be the most convenient to improve, since they gave the correct response of "no identification"; adding profile numbers to the databases would easily provide for accurate identification for an appreciable number of strains. The other rapid systems will require significant retooling of their databases because of the number of erroneous identifications.

A. sanguinicola is found in human infections and is frequently identified in clinical samples from elderly female patients with urinary tract infections. *A. sanguinicola is* similar to *A. urinae* in clinical implication, as both were first described as causes of urinary tract infections (6, 24) and then later found to be associated with bacteremia, sepsis, and endocarditis (4, 12). *A. sanguinicola* has been isolated from blood cultures of patients with sepsis and endocarditis (this report). *A. viridans*, the first *Aerococcus* species described, has been isolated from pa-

TABLE 5. MICs of 17 antimicrobial agents for 15 strains of *A. sanguinicola*

Antimicrobial drug (cancer range, μg/ml)	No. of strains for which MIC (μg/ml) of antimicrobial agent was:										No. of strains classified as S/I/R[a]
	0.03	0.06	0.12	0.25	0.5	1.0	2.0	4.0	8.0	16	
Penicillin (0.03–16.0)	15	0	0	0	0	0	0	0	0	0	15/0/0
Amoxicillin (0.03–8.0)	15	0	0	0	0	0	0	0	0		15/0/0
Cefotaxime (0.06–8.0)		4	6	5	0	0	0	0	0		15/0/0
Cefuroxime (0.25–4.0)				7	5	3	0	0			15/0/0
Meropenem (0.06–2.0)		13	0	0	0	1	1	0			13/0/2
Erythromycin (0.03–64.0)	2	3	8	2	0	0	0	0	0	0	15/0/0
Clindamycin (0.03–4.0)	2	0	4	6	3	0	0	0			12/3/0
Trimethoprim (0.12–8.0)			0	1	3	9	1	1	0		13/2/0
Levofloxacin (0.5–16)					1	8	0	1	1	4	9/1/5
Sparfloxacin (1.0–2.0)						15	0				15/0/0
Ciprofloxacin (2.0–8.0)						9	1	5			9/1/5
Chloramphenicol (2.0–16.0)							13	2	0		13/2/0
Vancomycin (0.12–2.0)			5	10	0	0	0				15/0/0
Quinupristin-dalfopristin (1.0–2.0)						15	0				15/0/0
Rifampin (2.0)							15				15/0/0
Linezolid (2.0–4.0)							15	0			15/0/0
Tetracycline (2.0–8.0)							15	0	0		15/0/0

[a] S, I, and R, susceptible, intermediate, and resistant, determined by using the NCCLS interpretive criteria for streptococci other than *S. pneumoniae* except for SXT and rifampin, for which the standards for *S. pneumoniae* were used.

tients with meningitis (15, 17), endocarditis (10, 18, 23), and bacteremia in granulocytopenia (11) and from human immunodeficiency virus-positive patients (19).

Whether it is clinically relevant to differentiate between the *Aerococcus* species remains to be determined.

Further investigation of the antimicrobial susceptibilities of the different species is required to resolve some of the apparent discrepancies in the literature. Penicillin resistance among strains of *A. viridans* has been reported (2, 4, 24). In an earlier study in this laboratory, it was found that nearly half the cultures of *A. viridans* were either highly or intermediately resistant to penicillin (2). One should interpret all published susceptibility reports for aerococci, including this report, with caution. The NCCLS does not have antimicrobial susceptibility criteria for the aerococci. Whether the interpretive criteria for streptococci other than *S. pneumoniae* or those for *S. pneumoniae* are valid for this genus can be questioned. Most reports describe single isolates or very small numbers of isolates, which indicates that *A. viridans* has various degrees of susceptibility to penicillin (10, 12, 15, 17–23). One report showed that 3 of 29 isolates were resistant (MIC of 0.25) to penicillin (11). Christensen and colleagues proposed using penicillin resistance as a potential way of differentiating between *A. viridans* (resistant) and *A. urinae* (susceptible) (5). The description of antimicrobial susceptibilities of *A. urinae* reported that these strains are susceptible to penicillin (4, 5, 6, 12, 24). We found that all 15 isolates of *A. sanguinicola* were susceptible to all beta-lactam antimicrobials.

The NCS has observed that all aerococci tested to date produce a zone of inhibition around a penicillin 10-unit disk (on blood agar plates), although the zone size may vary. Interestingly, an area of enhanced alpha hemolysis at the edge of the inhibitory zone may be observed for both *A. sanguinicola* and *A. viridans*, while *A. urinae* fails to produce this halo effect. This observation may provide a useful identification tool but does not imply susceptibility to penicillin. In 1989, Buu-Hoi and colleagues (3) reported that several strains of *A. viridans* had acquired the *ermB* gene of *Enterococcus faecalis* to confer erythromycin resistance and that other strains had acquired the *tetM* gene to code tetracycline resistance. We did not detect resistance to either erythromycin or tetracycline in the *A. sanguinicola* strains we investigated. One report of a single strain of *A. urinae* indicated that this isolate was susceptible to erythromycin (20). We found that 1 of the 15 strains of *A. sanguinicola* was resistant and 10 of the 15 strains were intermediately resistant to SXT. Kern and Vanek (11) reported that 19 of the 29 strains of *A. viridans* were resistant to SXT, while other reports (15, 24) indicated that all their strains were susceptible to SXT. Although there are no reports of SXT susceptibility studies for strains of *A. urinae*, several studies have reported sulfonamide resistance (4, 6, 12). Kern and Vanek (11) reported that 19 of the 29 strains of *A. viridans* in their studies were resistant to ofloxacin. We detected resistance to ciprofloxacin and levofloxacin but not sparfloxacin among the strains of *A. sanguinicola* in this study. We did not find any reported data on quinolone resistance in *A. urinae*. These data indicate that antimicrobial susceptibility studies should be performed on aerococci isolated from sterile sites as is currently done with the viridans group streptococci. It does not appear that antimicrobial resistance is a serious problem among the different species of aerococci at this time.

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