**Streptococcus tigurinus**, a Novel Member of the *Streptococcus mitis* Group, Causes Invasive Infections

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We recently described the novel species *Streptococcus tigurinus* sp. nov. belonging to the *Streptococcus mitis* group. The type strain AZ_3aT of *S. tigurinus* was originally isolated from a patient with infective endocarditis. According to its phenotypic and molecular characteristics, *S. tigurinus* is most closely related to *Streptococcus mitis*, *Streptococcus pneumoniae*, *Streptococcus pseudopneumoniae*, *Streptococcus oralis*, and *Streptococcus infantis*. Accurate identification of *S. tigurinus* is facilitated by 16S rRNA gene analysis. We retrospectively analyzed our 16S rRNA gene molecular database, which contains sequences of all clinical samples obtained in our institute since 2003. We detected 17 16S rRNA gene sequences which were assigned to *S. tigurinus*, including sequences from the 3 *S. tigurinus* strains described previously. *S. tigurinus* originated from normally sterile body sites, such as blood, cerebrospinal fluid, or heart valves, of 14 patients and was initially detected by culture or broad-range 16S rRNA gene PCR, followed by sequencing. The 14 patients had serious invasive infections, i.e., infective endocarditis (*n* = 6), spondylodiscitis (*n* = 3), bacteremia (*n* = 2), meningitis (*n* = 1), prosthetic joint infection (*n* = 1), and thoracic empyema (*n* = 1). To evaluate the presence of *Streptococcus tigurinus* in the endogenous oral microbial flora, we screened saliva specimens of 31 volunteers. After selective growth, alpha-hemolytic growing colonies were analyzed by matrix-assisted laser desorption ionization–time of flight mass spectrometry (MALDI-TOF MS) and subsequent molecular methods. *S. tigurinus* was not identified among 608 strains analyzed. These data indicate that *S. tigurinus* is not widely distributed in the oral cavity. In conclusion, *S. tigurinus* is a novel agent of invasive infections, particularly infective endocarditis.

We recently described a novel species within the *Streptococcus mitis* group, *Streptococcus tigurinus* sp. nov. (21). Based on phenotypic and molecular analyses, *S. tigurinus* is most closely related to *Streptococcus mitis*, *Streptococcus pneumoniae*, *Streptococcus pseudopneumoniae*, *Streptococcus oralis*, and *Streptococcus infantis*. This novel species was not recognized in the past due to the limitations of conventional phenotypic test methods and of commercial systems (API 20 Strept and Vitek 2; bioMérieux, Marcy l’Etoile, France) as regards accurate identification of species within the *S. mitis* group (1, 3, 8, 11, 17). In addition, we have shown that *S. mitis* strain ATCC 15914 was initially misassigned when it was identified in 1977 (9); molecular analyses revealed the identification of strain ATCC 15914 as *S. tigurinus* (21). *S. tigurinus* colonies on sheep blood agar are alpha-hemolytic, smooth, and white to grayish with a diameter of 0.5 to 1 mm after incubation at 37°C with CO2 for 24 h (21). Analyses by Vitek 2 resulted in identification as *S. mitis/S. oralis*, and analyses by matrix-assisted laser desorption ionization–time of flight mass spectrometry (MALDI-TOF MS) revealed identification as *S. pneumoniae* with a score of ≥2.2 (21). However, the limited discriminative power of MALDI-TOF MS within the *S. mitis* group has been recognized previously by other authors (10, 18, 20). Hence, an identification result of *S. pneumoniae*, even with a score as high as ≥2.2, has to be interpreted with caution. Thus, analyses by commercial test systems, such as Vitek 2, or by MALDI-TOF MS are helpful for initial assignment to the *S. mitis* group, but genetic analyses are required for definitive assignment as *S. tigurinus*. We demonstrated a significant sequence demarcation within the 5′ part of the 16S rRNA gene (±500 bp) to the most closely related species, i.e., *S. mitis*, *S. pneumoniae*, *S. pseudopneumoniae*, and *S. oralis*, which allowed definite identification of *S. tigurinus* (21). This is remarkable because the discriminative power of the 5′ part of the 16S rRNA gene is not sufficient for accurate identification among these closely related species (1, 12).

With the description of *S. tigurinus*, we identified a novel pathogen associated with serious invasive infections. *S. tigurinus* was first documented as the causative agent in multiple blood cultures and aortic valve specimens of a patient with infective endocarditis (21). Other members of the *S. mitis* group, such as *S. mitis* and *S. oralis*, which are known agents of infective endocarditis (7, 17) and sepsis in neutropenic cancer patients (2, 15), are commensals of the oral flora. Therefore, the oral cavity is suspected to be the ecological niche of *S. tigurinus*.

The aim of our study was to determine the involvement of *S. tigurinus* in clinical infections and to assess the presence of *S. tigurinus* in the oral cavity. By retrospective analysis of our institute’s 16S rRNA gene sequence database obtained from clinical samples covering the years 2003 to 2012, we identified *S. tigurinus* as an emerging pathogen causing invasive infections.
MATERIALS AND METHODS

This study was approved by the ethics committee of the canton of Zurich, Switzerland. The study included a retrospective analysis of laboratory and clinical data and a prospective analysis of saliva specimens of volunteers.

**Bacterial strains.** A retrospective analysis covering the period 2003 to 2012 of the 16S rRNA gene sequence database (SmartGene, Zug, Switzerland) of the Institute of Medical Microbiology, University of Zurich, Switzerland, was performed. Bacterial strains were grown on Columbia agar plates containing 5% defibrinated sheep blood (bioMérieux) at 37°C under aerobic conditions.

**Patients.** Fourteen patients with *S. tigurinus* infections were hospitalized in Switzerland, and one patient in Germany. Clinical data were retrieved from the patients’ medical records and reviewed by infectious disease specialists. Eleven patients were male; the mean age was 47.4 years (range, 21 to 74). Infective endocarditis was defined according to established criteria (14).

**Analysis of 16S rRNA gene.** DNA was extracted from the cultures as follows. A loopful of bacteria was suspended in 500 μl 0.9% NaCl and incubated by shaking at 80°C for 10 min. After centrifugation, the pellet was resuspended in 200 μl of InstaGene Matrix (Bio-Rad Laboratories, Hercules, CA) and incubated at 56°C for 2 h and then at 95°C for 10 min. The mixture was centrifuged, and the supernatant was used as the template for PCR. An 800-bp fragment of the 16S rRNA gene was amplified with primers BAK11w (5′-AGTTTGATCMTGGCTCAG-3′; positions 10 to 27, *Escherichia coli* numbering) and BAK2 [5′-GGACTACHGGTATCTTAAT-3′; positions 806 to 787, *Escherichia coli* numbering]. The cycling parameters included an initial denaturation for 5 min at 95°C, 40 cycles of 1 min at 94°C, 1 min at 48°C, and 1 min at 72°C, and a final extension for 10 min at 72°C. Five microliters of the DNA extract was used for amplification in a total volume of 50 μl containing 1.25 U of AmpliTaq DNA polymerase LD (Applied Biosystems, Rotkreuz, Switzerland) and the appropriate buffer. Amplicons were purified with a QIAquick PCR purification kit (Qiagen AG, Hombrechtkon, Switzerland) and sequenced with the forward primer BAK11w using an automatic DNA sequencer (ABI Prism 310 genetic analyzer; Applied Biosystems). Broad-range 16S rRNA gene PCR was performed directly from clinical specimens as described earlier (4). 16S rRNA gene BLAST analysis was performed using SmartGene software (SmartGene, Zug, Switzerland).

16S rRNA gene sequences were stored in a Web-accessible database environment provided by SmartGene.

**Antibiotic susceptibility testing.** Antibiotic susceptibility testing was performed using susceptibility test discs (Becton, Dickinson, Germany, and I2a, Montpellier, France), and interpretation was done according to CLSI 2012 guidelines (6). For penicillin and high-level gentamicin resistance, MICs were determined using Etest strips (AB bioMérieux, Marcy l’Étoile, France). Susceptibility testing was performed on Mueller-Hinton agar supplemented with 5% sheep’s blood, using overnight cultures at 0.5 McFarland standard followed by incubation at 35 ± 2°C with 5% CO₂ for 20 to 24 h.

**Oral cavity specimens.** Saliva samples were diluted with 0.85% NaCl and then plated on colistin-nalidixic acid agar. After incubation at 37°C with CO₂ for 24 h, alpha-hemolytic growing colonies were further analyzed. From each specimen, 20 morphologically different alpha-hemolytic colonies were picked and analyzed by MALDI-TOF MS after subcultivation. The analysis was performed by using a Microflex LT mass spectrometer (Bruker Daltonik GmbH, Bremen, Germany) using the MALDI BioTyper software package (version 3.0) with reference database version 3.1.2.0 (Bruker Daltonik GmbH). Sample preparation was done using the ethanol-formic acid extraction protocol according to the manufacturer’s instructions. For the identification of *S. tigurinus*, reference spectra of the strains *S. tigurinus* AZ_3a and *S. tigurinus* AZ_8 were added to the MALDI-TOF reference database. All putative strains displaying both a score of >2.2 to *S. tigurinus* and a score of <2.0 to *S. pneumoniae* in the reference database were suggestive of *S. tigurinus*. For confirmation, 16S rRNA gene analysis was performed as described above.

Nucleotide sequence accession numbers and strain deposition. Partial 16S rRNA gene sequences of cultured and uncultured *S. tigurinus* strains have been deposited in GenBank under the following accession numbers: strain AZ_1 (GenBank accession number JQ696859), AZ_2 (JQ696860), AZ_3b (JQ696868), AZ_4a (JQ696861), AZ_4b (JQ696869), AZ_5 (JQ696870), AZ_6 (JQ696862), AZ_7a (JQ696863), AZ_7b (JQ696871), AZ_8 (JQ696864), AZ_9 (JQ696872), AZ_10 (JQ696865), AZ_11 (JQ696866), AZ_12 (JQ696867), AZ_13a (JQ696873), AZ_13b (JQ696874), AZ_14 (JQ778987), and AZ_15 (JQ820471). The following *S. tigurinus* strains have been deposited in the Culture Collection of Switzerland (CCOS, Wadenswil, Switzerland): AZ_1 (culture number CCOS 683), AZ_2 (CCOS 675), AZ_4a (CCOS 676), AZ_6 (CCOS 681), AZ_7a (CCOS 667), AZ_8 (CCOS 678), AZ_10 (CCOS 679), AZ_11 (CCOS 682), AZ_12 (CCOS 680), and AZ_14 (CCOS 689).

RESULTS

Retrospective analysis of 16S rRNA gene sequence database. By retrospective analysis of our institution’s 16S rRNA gene sequence database, 17 sequences were identified which showed the highest 16S rRNA gene sequence similarities (99.4% to 100% identity) to *S. tigurinus* AZ_3a (JN004270). For all 17 sequences, BLAST search in GenBank revealed a sequence demarcation of ≥1.1% to the next validated species reference sequence. In general, a 16S rRNA gene sequence identity of ≥99.0% to a reference sequence of a classified species with a demarcation of >0.5% to the second classified species is considered to allow for assignment to species level (3). All 17 sequences, originally identified as *S. mitis* group, were assigned to *S. tigurinus*.

The 17 sequences were obtained from 14 patients and consisted of 12 *S. tigurinus* strains initially detected by culture methods, i.e., AZ_1, AZ_2, AZ_4a, AZ_6, AZ_7a, AZ_8, AZ_10, AZ_11, AZ_12, AZ_13a, AZ_13b, AZ_14, and 5 uncultured *S. tigurinus* bacteria detected by broad-range 16S rRNA gene PCR, i.e., AZ_4b, AZ_5, AZ_7b, AZ_9, and AZ_15. The *S. tigurinus* strains AZ_4a, AZ_7a, and AZ_10 have been described previously (21). The strains AZ_13a and AZ_13b were originally isolated at the Institute of Medical Microbiology, Friedrich-Schiller University of Jena, Germany. *S. tigurinus* bacteria identified from independent specimens of the same patient showed identical 16S rRNA gene sequences and are indicated with “a” and “b.” *S. tigurinus* strains AZ_1, AZ_2, AZ_4a, AZ_6, AZ_7a, AZ_8, AZ_10, AZ_12, AZ_13a, and AZ_14 were originally cultured from blood, strain AZ_11 from a vertebral-body biopsy specimen, and strain AZ_13b from an aortic valve specimen, respectively. In 2 patients with *S. tigurinus* isolated from blood, a sibling *S. tigurinus* bacterium was also recovered from a heart valve (AZ_4b) and cerebrospinal fluid specimen (AZ_7b) by sequence analysis after broad-range 16S rRNA gene PCR. In three patients, *S. tigurinus* was detected only by broad-range 16S rRNA gene PCR, i.e., AZ_5 from a periarticular prosthetic hip biopsy specimen and AZ_9 and AZ_15 from mitral valves.

By analyzing the electropherogram of the 16S rRNA gene sequence of *S. tigurinus* strain AZ_1, double peaks were observed by sequencing with the forward primer BAK11w, indicating the presence of different 16S rRNA gene copies. In sequencing with the reverse primer BAK2, an unambiguous electropherogram signal was obtained, consistent with a single copy. Detailed analysis of the electropherograms showed a frameshift mutation of two base pairs within the first 100 nucleotides of the 5′ part of the 16S rRNA gene, which resulted in a double-peak electropherogram.
when sequencing with the forward primer. Other sequences did not show a double-peak electropherogram.

**Patient clinical features.** In total, we identified 15 patients with isolation of *S. tigurinus*, including the 4 patients of the original *S. tigurinus* species description (21). The patient characteristics are summarized in Table 1. All patients had serious infections. Seven patients were diagnosed with definite infective endocarditis according to the modified Duke criteria (14). *S. tigurinus* was isolated from multiple blood cultures in five patients; in three of these patients, the pathogen was also isolated from surgically resected heart valve specimens. In two patients with infective endocarditis, *S. tigurinus* was detected only in heart valve specimens, presumably due to antibiotic treatment before blood culture sampling. In one patient with bacterial meningitis, *S. tigurinus* was found in multiple blood cultures and in cerebrospinal fluid. *S. tigurinus* was associated with spondylodiscitis (*n* = 3), prostatic joint infection (*n* = 1), and thoracic empyema (*n* = 1). Two patients had bacteremia alone. In 11 patients, *S. tigurinus* was the only organism detected in the specimens. Four patients had mixed infections, i.e., two patients with *S. tigurinus* and *Streptococcus salivarius* group bacteria, one patient with *S. tigurinus*, *S. salivarius* group bacteria, and a streptococcal species which was most closely related to *Streptococcus parasanguinis*, and one patient with *S. tigurinus* and *Staphylococcus aureus*. Most of the mixed infections occurred in patients with intravenous drug use. Both immunocompromised and immunocompetent patients had *S. tigurinus* infections. All patients recovered after appropriate antimicrobial therapy, with (*n* = 7) or without surgery (*n* = 8).

**Antimicrobial susceptibility profile.** The antibiotic susceptibility profile was determined for the cultured *S. tigurinus* strains (*n* = 13). All strains were susceptible to penicillin, and none of the strains displayed high-level gentamicin resistance (Table 2). For tetracycline, the strains AZ_6, AZ_7a, and AZ_14 displayed reduced susceptibility or resistance. No inductive clindamycin resistance was detected.

**Frequency of *S. tigurinus* in causing endocarditis.** Microbiological reports from the Institute of Medical Microbiology from 2003 to 2012 were consistent with infective endocarditis were investigated. In total, we identified 48 cases of infective endocarditis caused by viridans group streptococci, 7 (14.5%) of which were caused by *S. tigurinus*. Thirteen (27%) endocarditis cases were caused by *S. mitis*/*S. oralis*, 11 (23%) by *Streptococcus sanguinis*, 6 (12.5%) by *Streptococcus gordonii*, and the remaining 11 (23%) endocarditis cases were caused by other viridans group streptococci.

**Screening for *S. tigurinus* in the oral cavity.** Paraffin-stimulated saliva specimens of 31 volunteers (mean age of 27.8 years, range of 19 to 49 years; 16 male) were investigated for the presence of *S. tigurinus*. From the initial 620 alpha-hemolytic bacterial colonies isolated from the saliva of 31 persons, 12 strains did not grow after subcultivation, and therefore, 608 strains were analyzed by MALDI-TOF MS. Using MALDI-TOF screening criteria, 26 of the 608 strains, obtained from 17 persons, were suggestive for *S. tigurinus*. However, subsequent 16S rRNA gene analyses showed that none of the strains could be assigned to *S. tigurinus*; a single strain was closely related to *S. tigurinus AZ_3a* (JN004270), with a sequence similarity of 98.9%. Because the MALDI-TOF screening criteria, i.e., a score of >2.2 to *S. tigurinus* and a score of ≥2.0 to *S. pneumoniae* by molecular methods. Even with these relaxed criteria, however, no *S. tigurinus* strains were detected.

For the most recent endocarditis patients with isolation of *S. tigurinus* (patients 14 and 15) (Table 1), we had the opportunity to test saliva specimens for the presence of *S. tigurinus*. For patient 14, the saliva specimen was obtained at day 13 of antibiotic therapy. None of the 20 strains investigated was suggestive for *S. tigurinus*. For patient 15, the analysis of the saliva specimen obtained at day 15 of antibiotic therapy revealed 2 strains suggestive for *S. tigurinus*, but they were not confirmed by molecular methods.

**DISCUSSION**

Using our institute’s molecular database containing all 16S rRNA gene sequences obtained from clinical samples over a 10-year period, we showed the involvement of the newly described *S. tigurinus* species in serious clinical infections. Including the first recognized cases with *S. tigurinus* (21), we identified, in total, 15 patients from whom *S. tigurinus* was either isolated by initial culture methods or identified by broad-range 16S rRNA gene PCR. Most patients had infective endocarditis, but *S. tigurinus* was also identified as the causative agent in patients with spondylodiscitis, meningitis, prostatic joint infection, thoracic empyema, and bacteremia. *S. tigurinus* infection was documented in both immunocompromised and immunocompetent patients of various ages and with a wide range of underlying conditions. Given the limited number of patients, however, a specific risk factor profile for the development of invasive infections with *S. tigurinus* could not be established. Data regarding the dental and mucosal health of the patients were limited by the retrospective study design.

Most patients with *S. tigurinus* infection had endocarditis. The frequency of *S. tigurinus* in causing infective endocarditis compared to the frequencies of other viridans group streptococci seems to be remarkably high, since 14.5% of all putative endocarditis cases associated with viridans group streptococci as observed at our institute during 2003 to 2012 were caused by *S. tigurinus*. The number of patients with infective endocarditis might even be higher due to missing information. The causative agents in 11 of the 13 endocarditis cases caused by *S. mitis*/*S. oralis* were identified only by conventional phenotypic methods, such as Vitek and API 20 Strep (bioMérieux). Therefore, it is possible that a substantial proportion of these bacteria actually represent *S. tigurinus* if molecular analyses had been performed.

The oral microbial flora was assumed to represent a main reservoir of *S. tigurinus*, similar to the case for other viridans group streptococci. However, screening a population of 31 volunteers for the presence of *S. tigurinus* in the saliva did not reveal any *S. tigurinus* strains. In two patients with infective endocarditis caused by *S. tigurinus*, we were able to evaluate saliva specimens during the first 15 days of antibiotic therapy. *S. tigurinus* was not detected in these specimens. *S. tigurinus* seems rarely to be present as part of the oral flora. Given the limited discriminatory power of MALDI-TOF MS within the *S. mitis* group (10, 18, 20) and the limited capacity to analyze all strains by molecular methods, however, we might have underestimated the presence of *S. tigurinus* in the oral microbial flora. A BLAST search in GenBank revealed that *S. tigurinus* is likely to be present in the human oral cavity, since a number of 16S rRNA gene sequences deriving from the human mouth showed high sequence similarities to the *S. tigurinus* type strain sequence (JN004270), allowing accurate assignment (16).
<table>
<thead>
<tr>
<th>Patient</th>
<th>Age (yr), sex</th>
<th>S. tigurinus (strain/uncultured)</th>
<th>Site of isolation (no. of positive blood cultures)</th>
<th>Immunosuppression</th>
<th>Comorbidity(ies)</th>
<th>Oral health status</th>
<th>Antibiotic treatment</th>
<th>Diagnosis</th>
<th>Outcome</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>47, M</td>
<td>AZ_1(^b)</td>
<td>Blood (4)(^c)</td>
<td></td>
<td>Systemic lupus erythematosus, long-term corticosteroid therapy</td>
<td>ND</td>
<td>AMC and RIF</td>
<td>Bacteremia</td>
<td>Recovered</td>
</tr>
<tr>
<td>2</td>
<td>58, M</td>
<td>AZ_2(^b)</td>
<td>Blood (6)(^c)</td>
<td></td>
<td>Prosthetic aortic valve</td>
<td>ND</td>
<td>VAN, CIP, and RIF, then PEN and GEN, and then CRO</td>
<td>Spondylodiscitis, bacteremia</td>
<td>Recovered</td>
</tr>
<tr>
<td>3</td>
<td>74, F</td>
<td>AZ_3a(^b), AZ_3b(^d)</td>
<td>Blood (6)(^c) and aortic valve</td>
<td></td>
<td>Aortic stenosis</td>
<td>Dental procedure 2 mo earlier</td>
<td>PEN and GEN and then CRO</td>
<td>Endocarditis</td>
<td>Recovered</td>
</tr>
<tr>
<td>4</td>
<td>37, F</td>
<td>AZ_4a(^b), AZ_4b(^d)</td>
<td>Blood (6)(^c) and mitral valve</td>
<td></td>
<td>No abnormalities detected</td>
<td>No abnormalities detected</td>
<td>PEN and GEN and then CRO</td>
<td>Endocarditis</td>
<td>Recovered</td>
</tr>
<tr>
<td>5</td>
<td>65, M</td>
<td>AZ_5(^d)</td>
<td>Periarticular hip biopsy(^c)</td>
<td>Heavy alcohol use</td>
<td>Possible cirsosis of the liver, atrial fibrillation</td>
<td>Extraction of several carious teeth during antibiotic therapy</td>
<td>CRO then CTX</td>
<td>Infection of prosthetic hip joint</td>
<td>Recovered</td>
</tr>
<tr>
<td>6</td>
<td>35, M</td>
<td>AZ_6(^b)</td>
<td>Blood (4)(^c)</td>
<td></td>
<td>Intraavenous drug use</td>
<td>No abnormalities detected</td>
<td>AMC, then ERTA, and then AMC</td>
<td>Thoracic empyema, bacteremia</td>
<td>Recovered</td>
</tr>
<tr>
<td>7</td>
<td>30, M</td>
<td>AZ_7a(^b), AZ_7b(^d)</td>
<td>Blood (4)(^c) and cerebrospinal fluid</td>
<td></td>
<td>Cerebrospinal fluid leak after operation for hypophysal tumor</td>
<td>No abnormalities detected</td>
<td>PEN and GEN and then CRO</td>
<td>Endocarditis</td>
<td>Recovered</td>
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<tr>
<td>8</td>
<td>64, M</td>
<td>AZ_8(^b)</td>
<td>Blood (8)(^c)</td>
<td></td>
<td>No</td>
<td>No abnormalities detected</td>
<td>AMC and GEN and then PEN</td>
<td>Endocarditis</td>
<td>Recovered</td>
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<tr>
<td>9</td>
<td>65, M</td>
<td>AZ_9(^d)</td>
<td>Mitral valve(^c)</td>
<td></td>
<td>No</td>
<td>No abnormalities detected</td>
<td>CIP and then AMC and GEN</td>
<td>Endocarditis</td>
<td>Recovered</td>
</tr>
<tr>
<td>10</td>
<td>29, F</td>
<td>AZ_10(^d)</td>
<td>Blood (2)(^f)</td>
<td></td>
<td>Intraavenous drug use</td>
<td>No abnormalities detected</td>
<td>CRO and then AMC and GEN</td>
<td>Spondylodiscitis, bacteremia</td>
<td>Recovered</td>
</tr>
<tr>
<td>11</td>
<td>32, M</td>
<td>AZ_11(^b)</td>
<td>Vertebral body biopsy(^g)</td>
<td></td>
<td>Intraavenous drug use</td>
<td>No abnormalities detected</td>
<td>PEN and GEN and then PEN</td>
<td>Endocarditis</td>
<td>Recovered</td>
</tr>
<tr>
<td>12</td>
<td>50, M</td>
<td>AZ_12(^b)</td>
<td>Blood (1)(^f)</td>
<td>HIV infection</td>
<td>Myocardial infarction</td>
<td>No abnormalities detected</td>
<td>AMX and CRO and AMC</td>
<td>Bacteremia</td>
<td>Recovered</td>
</tr>
<tr>
<td>13</td>
<td>47, M</td>
<td>AZ_13a(^b), AZ_13b(^b)</td>
<td>Blood (5)(^c)</td>
<td></td>
<td>No</td>
<td>No abnormalities detected</td>
<td>PEN and GEN and then PEN</td>
<td>Endocarditis, local encephalitis</td>
<td>Recovered</td>
</tr>
<tr>
<td>14</td>
<td>57, M</td>
<td>AZ_14(^d)</td>
<td>Blood (8)(^c)</td>
<td>Hypertrophic obstructive cardiomyopathy</td>
<td>No abnormalities detected</td>
<td>No abnormalities detected</td>
<td>Healthy teeth, mild gingivitis</td>
<td>Endocarditis</td>
<td>Recovered</td>
</tr>
<tr>
<td>15</td>
<td>21, F</td>
<td>AZ_15(^d)</td>
<td>Mitral valve(^c)</td>
<td></td>
<td>No</td>
<td>No abnormalities detected</td>
<td>Healthy teeth, mild gingivitis</td>
<td>Endocarditis</td>
<td>Recovered</td>
</tr>
</tbody>
</table>

\(^a\) Abbreviations: CAD, coronary artery disease; ND, not determined; AMC, amoxicillin-clavulanic acid; AMX, amoxicillin; CTX, cefotaxime; CIP, ciprofloxacin; CRO, ceftriaxone; ERTA, ertapenem; GEN, gentamicin; MTZ, metronidazole; PEN, penicillin; RIF, rifampin; VAN, vancomycin.

\(^b\) Cultural strain.

\(^c\) S. tigurinus was the only organism detected in the specimens.

\(^d\) Uncultured, detected by broad-range 16S rRNA gene PCR analysis.

\(^e\) Staphylococcus aureus was detected in the same blood cultures.

\(^f\) Staphylococcus salivarius group bacteria and Streptococcus sp. bacteria most closely related to Streptococcus parasanguinis were detected in the same blood cultures.

\(^g\) Streptococcus salivarius group bacteria were detected in the same specimen.
This is consistent with the mucosal presence of streptococcal species in general (17). In future, oral microbiome projects will provide a more detailed analysis of the composition of the oral microbiota and the presence of S. tigurinus.

To date, we have detected S. tigurinus as the causative agent of invasive infections in patients from Switzerland and Germany, and it is likely that other bacterial agents previously reported as belonging to the S. mitis group actually represent S. tigurinus. This is most likely due to limitations of species identification within the S. mitis group by conventional means. In general, sequences of strains classified only by biochemical phenotypic test methods frequently result in deposition in GenBank with an inaccurate assignment (5), e.g., the erroneous assignment of strain ATCC 15914 as S. mitis (13, 19, 21).

Our data clearly document that S. tigurinus is a significant human pathogen. The observation that all patients from whom S. tigurinus was isolated had severe invasive infections (Table 1) suggests the importance of conducting further studies to better characterize the potential pathogenic role of S. tigurinus in invasive infections, particularly infective endocarditis. A prerequisite is the accurate identification of viridans group streptococci by molecular methods. Further work also should focus on the natural habitat of and the potential for colonization with S. tigurinus.

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REFERENCES


