Open Forum Infectious Diseases

MAJOR ARTICLE

Identification of *Streptomyces* spp. in a Clinical Sample: Always Contamination? Results of a French Retrospective Study

Emmanuelle Gras,[1,2] Emmanuelle Bergeron,[3,4] Mathilde Puges,[5] Maïlys Ducours,[5] Charlotte Leleux,[6] Lucie Amoureux,[7] Baptiste Jean,[8] Imane Bendjelloul,[9] François Camelena,[10,11] Rachel Chenouard,[12] Rafael Mahieu,[13] Olivier Lemenand,[14] Alexandre Toro,[15] Alain Lecoustumier,[16] Olivier Lortholary,[17,18] Véronica Rodriguez Nava,[3,4] and David Lebeaux,[1,2] for the French Streptomyces Study Group*

[1]Université Paris Cité, Paris, France, [2]Department of Microbiology, Antimicrobial Stewardship Team, European Hospital Georges Pompidou, Assistance Publique—Hôpitaux de Paris, Paris, France, [3]Research Group on Bacterial Opportunistic Pathogens and Environment, UMR CNRS5557, INRA1418 Ecologie Microbienne, French Observatory of Nocardiosis, Hospices Civils de Lyon, Lyon, France, [4]Université de Lyon 1, VetAgro Sup, Lyon, France, [5]Department of Infectious and Tropical Diseases, Teaching Hospital of Bordeaux, Bordeaux, France, [6]Infectious and Tropical Diseases Department, Teaching Hospital of Amiens, Amiens, France, [7]Department of Bacteriology, Teaching Hospital of Dijon, France, [8]Department of Infectious and Tropical Diseases, Teaching Hospital of Toulouse, Toulouse, France, [9]Laboratory of Bacteriology and Hygiene, Hospital of Cahors, Cahors, France, [10]Department of Bacteriology, Saint-Louis-Lariboisière Hospital, Assistance Publique—Hôpitaux de Paris, Paris, France, [11]Université Paris Cité, INSERM UMR 1137, IAME, Paris, France, [12]Department of Bacteriology and Hygiene, Teaching Hospital of Angers, Angers, France, [13]Department of Infectious Diseases, Teaching Hospital of Angers, University of Angers, Angers, France, [14]Department of Medical Biology, Hospital of Saint-Nazaire, Saint-Nazaire, France, [15]Department of Bacteriology, Hospital of Martigues, Martigues, France, [16]Department of Bacteriology, Hospital of Tarbes, Tarbes, France, [17]Necker-Enfants Malades University Hospital, Centre d'Infectiologie Necker-Pasteur, Assistance Publique—Hôpitaux de Paris, IHU Imagine, Paris, France, and [18]Institut Pasteur, CNRMA, CNRS, UMR 2000, Paris, France

Background. *Streptomyces* are environmental gram-positive bacilli that can cause ubiquitous mycetoma and, more rarely, invasive infections. We describe the clinical relevance of *Streptomyces* spp. identified in human samples and characteristics of patients with invasive *Streptomyces* infections.

Methods. We conducted a retrospective (2006–2017) study of *Streptomyces* isolates identified in clinical samples in French microbiology laboratories. *Streptomyces* genus was confirmed by a specific 16S rRNA polymerase chain reaction, and antibiotic susceptibility testing was performed by disk diffusion and trimethoprim-sulfamethoxazole minimum inhibitory concentration (E-test) if resistance was suspected. Patient characteristics, treatments, and outcomes were collected. Invasive infection was defined as a positive culture from a sterile site with signs of infection but without cutaneous inoculation.

Results. Of 137 *Streptomyces* isolates, all were susceptible to amikacin (113/113) and linezolid (112/112), and 92.9% to imipenem (105/113). Using disk diffusion, 50.9% (57/112) of isolates were susceptible to trimethoprim-sulfamethoxazole, but most of the apparently resistant isolates (25/36, 69.4%) tested by E-test were ultimately classified as susceptible. Clinical data were obtained for 63/137 (45.9%) isolates: 30 (47.6%) invasive infections, 8 (12.7%) primary cutaneous infections, 22 (34.9%) contaminations, 3 (4.7%) respiratory colonization. Patients with invasive infection were more frequently receiving corticosteroids than patients without invasive infection (11/30, 36.7%, vs 2/25, 8.0%; $P = .03$), and at 6-month follow-up, 14 of them were cured, 3 had relapsed, 4 were dead, and 9 were lost to follow-up.

Conclusions. Half of the clinical samples that grew *Streptomyces* were from patients with invasive infection. In that case, antimicrobial therapy should include 1 or 2 antibiotics among linezolid, amikacin, or imipenem.

Keywords. *Actinobacteria*; contamination; environment; invasive infection.

Streptomyces are filamentous gram-positive aerobic bacilli belonging to the *Actinobacteria* phylum that can be found in natural environments [1]. More than 650 species within the *Streptomyces* genus have currently been identified [2]. *Streptomyces* can cause actinomycetoma, a chronic subcutaneous infection resulting from repeated bacterial inoculation. Actinomycetoma has been described worldwide but has a higher prevalence in the "mycetoma belt" on both sides of the Tropic of Cancer, with >3 cases per 100 000 inhabitants in Mauritania [3]. *Streptomyces somaliensis* is the main species responsible for *Streptomyces* actinomycetoma and was identified in >10% of cases of mycetoma in a recent meta-analysis [3]. Actinomycetoma presents as a painless tumor often localized on the extremities [4, 5], but local extension may lead to severe complications when vital organs are invaded [6, 7].

Received 17 March 2022; editorial decision 24 May 2022; accepted 26 May 2022; published online 6 June 2022

*Collaborators listed in the Acknowledgments

Correspondence: Veronica Rodriguez Nava, PharmD, PhD, Research Group on Bacterial Opportunistic Pathogens and Environment, UMR CNRS5557, INRA1418 Ecologie Microbienne, French Observatory of Nocardiosis, Hospices Civils de Lyon, France, Université de Lyon 1, VetAgro Sup, 8 avenue Rockefeller, 69373 Lyon Cedex 08, France (veronica.rodriguez-nava@univ-lyon1.fr).

Open Forum Infectious Diseases®

https://doi.org/10.1093/ofid/ofac271

In addition to these primarily cutaneous infections, *Streptomyces* can, rarely, be responsible for invasive infection, independent of any cutaneous trauma. Since 1951, 40 cases of invasive infection have been published, including a limited case series [8] (Supplementary Table 1). Most of these patients had underlying conditions, such as drug-induced immunosuppression, cancer, or hematological malignancies. The lungs were the most frequently involved organs, and chest imaging revealed nodules, lung consolidation, and interstitial changes [8–10]. The presence of foreign materials, a central venous catheter [11], and a prosthetic cardiac valve [12, 13] have also been reported. Identification of *Streptomyces* spp. in a clinical sample may also be related to environmental contamination during or after sample collection or to *Streptomyces* colonization of the patient's airways.

The main objective of this study was to describe the microbiological diversity of *Streptomyces* isolates identified in human samples and their antibiotic susceptibilities. Secondary objectives were to describe the clinical characteristics, therapeutic strategies, and outcomes of a large cohort of patients with invasive *Streptomyces* infection and compare these factors with those in patients with *Streptomyces* colonization or contamination.

METHODS

Study Design

We conducted a retrospective study in France, including data related to all *Streptomyces* isolates identified in a human clinical sample that had been sent during the study period (2006–2017) to the Observatoire Français des Nocardioses (OFN), a French laboratory studying *Actinobacteria* through species identification and antibiotic susceptibility testing (AST) [14]; to ensure completeness, we also reached out to a French national network of microbiologists working in >200 general hospitals (Collège de Bactériologie, Virologie et Hygiène) and contacted every French infectious disease specialist through the Infectio-Flash mailing list. No additional cases were identified with these 2 latter strategies, suggesting that the vast majority of *Streptomyces* isolates are sent to the OFN. Nonhuman samples were excluded (n = 12).

For each *Streptomyces* isolate, we asked the microbiologist or infectious disease specialist at the hospital that had sent the sample to complete a standardized clinical record form.

Microbiology

After receipt of the microbiological sample at the OFN, genus identification was performed by a *Streptomyces*-specific polymerase chain reaction (PCR) based on amplification and detection of the gene encoding the 16S rRNA. *Streptomyces* DNA was extracted from bacterial colonies by the boiling method using achromopeptidase (10 UμL-1, Sigma-Aldrich) [15]. AM42 (5= CAA GGG CAT CCA CCG T -3=) and AM44 (5=- CTT CGG GGT GAT CTG GGG ACT CAC -3=) primers were used to amplify a *Streptomyces* genus–specific 700-bp fragment of 16S rRNA [16]. Amplification was carried out using packaged PCR tubes (Ready-to-Go PCR beads, Amersham Biosciences, Orsay, France). Two hundred nanograms of DNA and primers, at a final concentration of 0.4 μM for each primer, was added, for a final volume of 25 μL of PCR mixture. Initial denaturation was then performed at 98°C for 5 minutes. After 30 cycles of denaturation at 95°C for 60 seconds, primer annealing at 45°C for 40 seconds, and extension at 72°C for 120 seconds, followed by postamplification extension at 72°C for 10 minutes, PCR products were purified using the E.Z.N.A. gel extraction kit (Omega Bio-Tek, Vaulx-en-Velin, France).

No guidelines are available for *Streptomyces* AST, but these bacteria share common features with *Nocardia*, including culture techniques. AST was therefore performed using the disk diffusion on cation-adjusted Mueller Hinton (CA-MH) agar plates, as previously described for *Nocardia* [14], upon request from the laboratory sending the sample. Inocula were prepared according to the Clinical Laboratory Standards Institute (CLSI) M24-A2 standard, and results were read after 72 hours of culture [14]. For each antibiotic disk, the diameter of the inhibition zone was recorded and compared with thresholds (Supplementary Table 2). *Escherichia coli* ATCC 25922, *Staphylococcus aureus* ATCC 25923, and *Nocardia asteroides* ATCC 19247 T were used as quality control organisms. Resistant or intermediate isolates were considered "nonsusceptible" to the antibiotic in question. For trimethoprim-sulfamethoxazole, plates were read at 80% of growth inhibition following the same criterion as that used for *Nocardia* genus [17]. If resistance to trimethoprim-sulfamethoxazole was suspected (inhibition zone <10 mm) using the disk diffusion method, susceptibility was also assessed using an E-test strip placed on Muller-Hinton blood agar plates and read at 72 hours (Figure 1). An isolate was considered susceptible using this method if the trimethoprim-sulfamethoxazole minimum inhibitory concentration (MIC) obtained was ≤2/38 mg/L and resistant if the MIC was ≥4/76 mg/L.

Clinical Data and Definitions

For each *Streptomyces* isolate, we tried to complete a standardized clinical record (Supplementary Methods) including the following:

Patient characteristics included age, sex, underlying diseases (eg, chronic pulmonary diseases, malignancies, or organ transplant before the date of sampling), and calculated Charlson Comorbidity Index score [18]. Patients with lung cancer were included in the chronic pulmonary disease category as this condition is frequently associated with local immunosuppression.

Clinical presentation at the time of sampling included general symptoms (fever, chills), organ-specific symptoms, and

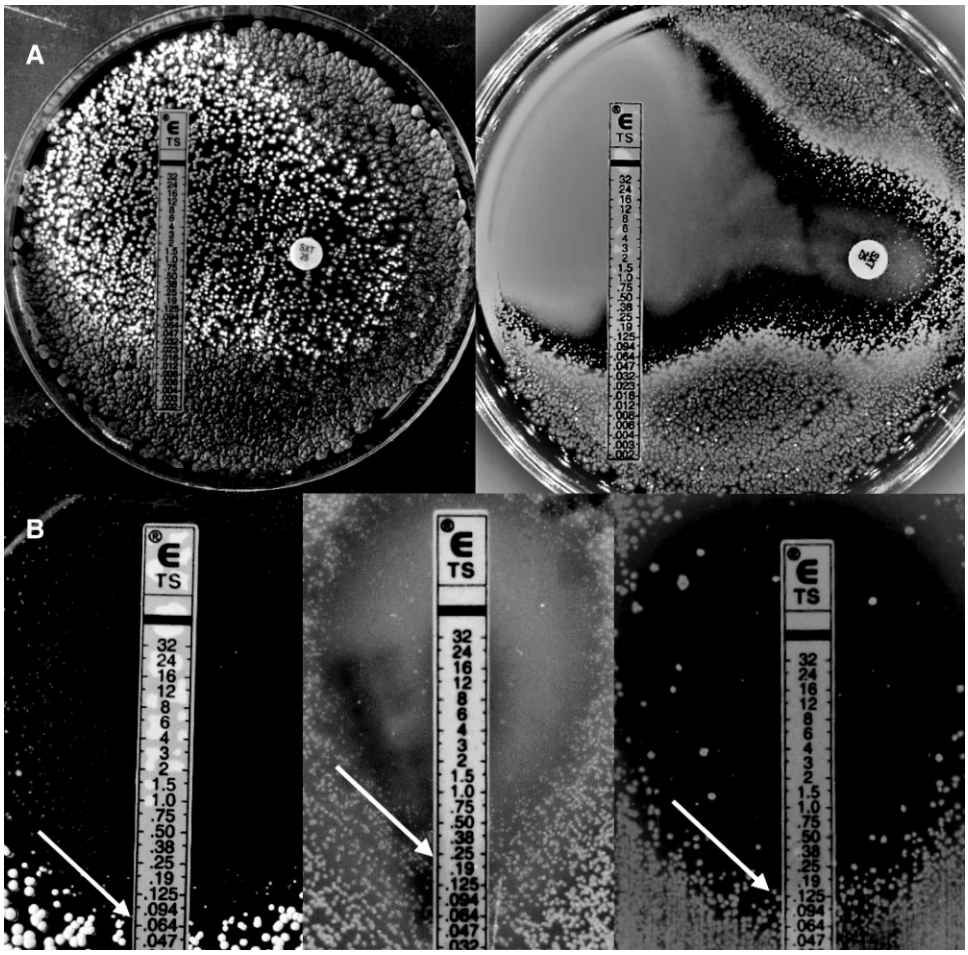

Figure 1. Interpretation of trimethoprim-sulfamethoxazole susceptibility testing using a combination of the disk diffusion method and E-test strip on Mueller-Hinton agar plate read after 72 hours of culture. A, Comparison of a resistant isolate (left picture) and a susceptible isolate (right picture). B, Reading of the trimethoprim-sulfamethoxazole MIC using the 80% inhibition criterion. From left to right, trimethoprim-sulfamethoxazole MICs were (white arrows) 0.094, 0.25, and 0.125. Abbreviation: MIC, minimum inhibitory concentration.

delay between symptom onset and diagnosis. Recorded blood markers included serum creatinine, leukocyte count, and C-reactive protein (CRP). Sample source and presence of other pathogens in the same sample were recorded. *Streptomyces* was considered a contaminant when present in a single sample with no clinical, radiological, or biological signs suggestive of infection. Colonization was defined when the *Streptomyces* isolate was isolated at least twice in a clinical sample from a patient with no sign of infection. Primary cutaneous *Streptomyces* infection was confirmed when cutaneous or subcutaneous infection followed direct inoculation from a wound, bite, or post-traumatic skin lesion. Invasive *Streptomyces* infection was diagnosed when there were signs of infection in an otherwise sterile site associated with a positive culture and no history of cutaneous inoculation. Disseminated infection was diagnosed when 2 noncontiguous organs were involved. Classifications were made by the physician completing the clinical record and 1 senior author (E.G.).

Therapeutic data included antibiotic regimen (duration and appropriateness [defined as at least 1 antibiotic with in vitro activity against the *Streptomyces* isolate]) and need for surgery. Relapse was defined as symptom recurrence after the end of treatment.

For *Streptomyces* isolates with no standardized clinical form completed, we extracted the patient's sex and age at the time of sampling, type of clinical sample, and AST of the *Streptomyces* isolate from the clinical microbiology laboratory database.

Statistical Analysis

Median (extreme variables) or mean (SD) values are used for continuous variables. Categorical variables are expressed as numbers and percentages. We compared 2 groups of isolates: "invasive infection" and "no infection." Because primary cutaneous *Streptomyces* infection following soil-contaminated wounds has already been comprehensively described in the literature and likely does not bear the same mechanism of

infection as invasive infection, we deliberately removed these cases from the analysis. Univariate analysis was conducted using the Fisher or chi-square test for categorical variables and the Student *t* test for continuous variables or the Wilcoxon-Mann-Whitney *U* test when a nonparametric test was required.

Ethics

The study was approved by the local ethics committee (Comité d'Éthique Necker-Enfants Malades reference 2018-DL10) and a national expert committee (Comité d'Expertise pour les Recherches, les Études et les Évaluations dans le Domaine de la Santé, reference TPS 95334) and was declared to the CNIL (Comité National de l'Informatique et des Libertés, reference MMS/OTB/AR1813447). Patients were informed of the present study by mail and could refuse to allow their data to be included in the study at any time. Patient confidentiality was ensured by anonymization of their clinical record.

RESULTS

Microbiological Diversity of *Streptomyces* Isolates In France

Between 2006 and 2017, *Streptomyces* was isolated from 137 samples at the OFN, which were included in the microbiological analysis (Table 1; Supplementary Table 3). The majority of the patients were men (91/131, 69.5%), and the median patient age (range) was 62 (47–75) years (Table 1). The lungs were the most frequent source of a positive culture (n = 70, 51.9%); *Streptomyces* were isolated from blood cultures or an indwelling device in 24 (17.8%) and 6 cases (4.4%), respectively (Table 1).

On AST, all tested isolates were susceptible to linezolid (112/112) and amikacin (113/113) (Figure 2A). Most isolates were susceptible to imipenem (92.9%, 105/113); 52.1% (37/71) were susceptible to meropenem. Trimethoprim-sulfamethoxazole susceptibility results depended on the method chosen: 50.8% (57/112) of the isolates were susceptible using the disk diffusion method on CA-MH agar plates, but 69.4% (25/36) were susceptible to trimethoprim-sulfamethoxazole when isolates previously classified as resistant were tested again using an E-test strip on MH-blood agar plates (Figure 2B). Twenty-one of the isolates identified as resistant using disk diffusion did not resume growth, so they could not be tested using the E-test. A majority of *Streptomyces* isolates were susceptible to tetracyclines, although most data were available for minocycline (94.7% susceptibility, n = 113) (Figure 2A). Isolates were susceptible to moxifloxacin and ciprofloxacin in 96.0% (72/75) and 76.1% (86/113) of cases, respectively.

Description of the Population, Characteristics, and Outcomes of Patients With Invasive Infection

For the 137 *Streptomyces* isolates, we obtained complete clinical data for 63 patients (39 men [61.9%]; median age [range], 62.5 [1–92] years) from 25 hospitals (Table 2). The mean (SD) Charlson Comorbidity Index score was 3.8 (2.8). Twenty-four of the patients (38.1%) had chronic lung disease, and 4 patients (6.3%) had received a lung transplant. Nineteen patients had drug-induced immunosuppression (30.6%), 10 of whom (52.6%) were receiving >1 immunosuppressive drug. Corticosteroids were the main immunosuppressive drugs used (n = 13, 20.9%); 9 patients (14.3%) had received a daily dose of >10 mg for >3 months (Table 2).

Invasive *Streptomyces* infection was diagnosed in 30 (47.6%) cases with no disseminated infection reported. Primary cutaneous infection, contamination, and respiratory colonization were diagnosed in 8, 22, and 3 patients, respectively.

Table 1. Characteristics of the 137 *Streptomyces* Isolates Received at the Observatoire Français des Nocardioses Between 2006 and 2017

Characteristics	137 *Streptomyces* Isolates
Patients' demographic data	
Age (n = 130), median [range], y	62 [47–75]
Male (n = 131), No. (%)	91 (69.5)
Country of origin, No. (%)	
France	132 (97.9)
Other[a]	3 (2.1)
Site and method of sampling (n = 135), No. (%)	
Lung	70 (51.9)
Sputum	16
Bronchial aspirate	28
Protected pulmonary sample[b]	20
Lung biopsy	4
Pleural fluid	2
Blood cultures	24 (17.8)
Skin and soft tissues	13 (9.7)
Skin biopsy	3
Post-traumatic skin lesions	6
Cutaneous abscess	4
Bone and joint	8 (5.9)
Bone biopsy	4
Open fracture	2
Other[c]	2
Ears, eyes, nose, and throat	6 (4.4)
External auditory canal	2
Nasal swab	2
Other[d]	2
Cerebrospinal fluid	2 (1.5)
Digestive tract	3 (2.2)
Gastric tube	2
Gall bladder stone	1
Lymph node biopsy	1 (0.7)
Heart and vessels[e]	2 (1.5)
Indwelling device[f]	6 (4.4)

Abbreviation: PICC, peripherally inserted central catheter.

[a]Other countries: Belgium, Senegal, Netherlands.

[b]Protected pulmonary sample: broncho-alveolar lavage, protected bronchial brush.

[c]Bone and joint other: joint fluid and hygroma aspirate.

[d]Ears, eyes, nose, and throat other: maxillary sinus and corneal abscess.

[e]Heart and vessels: aortic biopsy, cardiac valve.

[f]Indwelling device: 1 of each: PICC-line catheter, spinal cord neurostimulator cable, peridural catheter, humeral fixation, aortic prosthetic, peritoneal dialysis fluid.

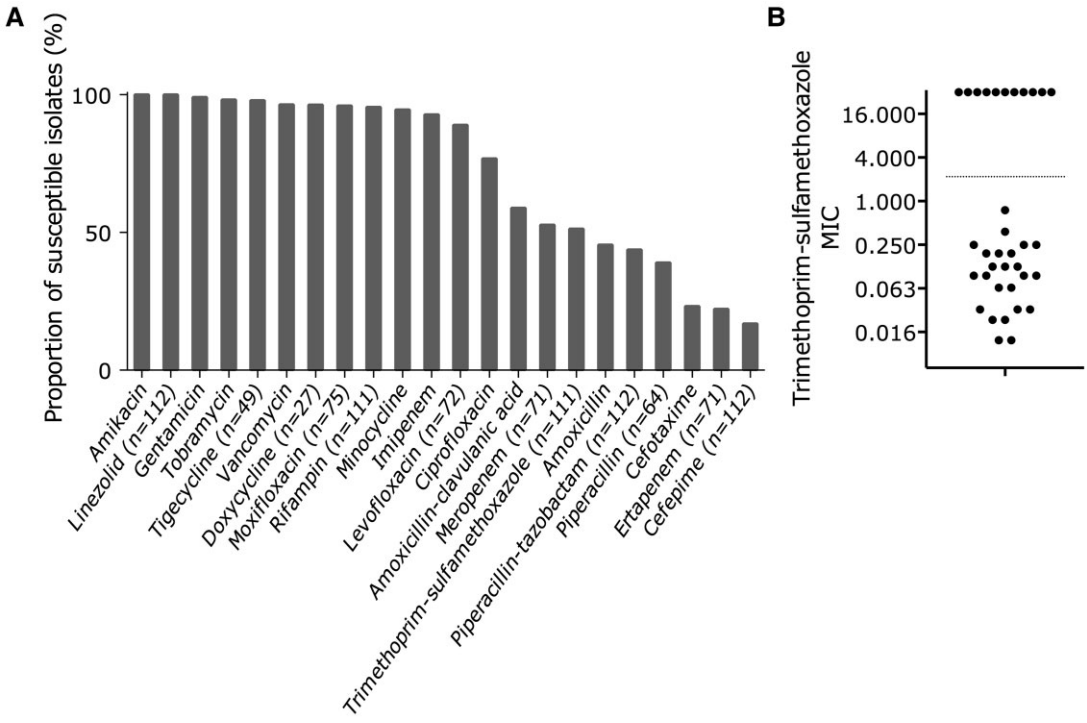

Figure 2. Susceptibility of 113 *Streptomyces* isolates to different antibiotics. A, Proportion of susceptible isolates (n = 113 or indicated between parentheses) using the disk diffusion on CA-MH agar plate method, read after 72 hours of culture. With this method, 50.8% (57/112) of the isolates were susceptible to trimethoprim-sulfamethoxazole. B, Results of the trimethoprim-sulfamethoxazole MIC measured by E-test strips on an agar plate. An isolate was considered susceptible if the trimethoprim-sulfamethoxazole MIC obtained was ≤2/38 mg/L (dotted line). Of note, among 55 isolates classified as being resistant using the disk diffusion method, only 36 resumed growth and could be tested using E-test strips. Isolates with trimethoprim-sulfamethoxazole MICs >32/608 are depicted as "32." Abbreviation: MIC, minimum inhibitory concentration.

Most of the cases of invasive *Streptomyces* infection were in men (n = 20, 66.7%), with a mean age (SD) of 60 (17) years, and a mean Charlson Comorbidity Index score (SD) of 3.6 (2.4) (Table 2). The main comorbidities were chronic lung disease (n = 14, 46.7%), solid tumor (n = 9, 30.0%), and solid organ transplantation (n = 4, 13.3%). Thirteen patients (n = 13, 43.3%) had drug-induced immunosuppression, including 11 patients who were receiving corticosteroid therapy (36.7%). For 17 (56.7%) patients, symptoms had started >7 days before diagnosis (mean [SD], 112 [187] days). The lungs were involved in 21 cases (66.6%), and 2 cases were related to an indwelling device (1 bioprosthetic cardiac valve and 1 aortic prosthesis). Dyspnea, cough, and sputum production were present in 12 (42.9%), 9 (30.0%), and 8 (26.7%) patients, respectively. Fatigue and fever were present in 9 (30.0%) and 6 (20.0%) patients, respectively. Among the 21 patients with pulmonary infection, the most frequent images on chest computed tomography (CT) scan were crazy paving (n = 7, 33.3%) and micronodules (n = 7, 33.3%). Eight patients (26.7%) were coinfected with other pathogens.

Twenty-eight patients received treatment (antibiotic only [n = 24], surgery only [n = 1], or antibiotic and surgery [n = 3]) for invasive *Streptomyces* infection (Supplementary Table 4). Antibiotic regimens included a beta-lactam (21/27, 77.7%), trimethoprim-sulfamethoxazole (12/27, 44.4%), amikacin (8/27, 29.6%), and fluoroquinolones (9/27, 33.3%). The antibiotic regimen was appropriate in 24/27 patients (88.8%). Twenty-one out of 27 patients (77.7%) received a multidrug regimen. The median antibiotic treatment duration (range) was 30.5 (7-365) days. At 6-month follow-up, 14 patients were cured, 3 had relapsed, 4 had died, and 9 were lost to follow-up. One patient relapsed at 8 months. The median follow-up period (range) was 12 (0–156) months.

Comparison of Patients With Invasive *Streptomyces* Infection and No Infection

There was no statistically significant difference in the proportion of patients with drug-induced immunosuppression among the patients with invasive infection and those with no infection (13/30 [43.3%] vs 6/25 [24.0%]; $P = .224$) (Table 2); patients with invasive infection were more frequently receiving corticosteroids (11/30 [36.7%] vs 2/25 [8.0%]; $P = .03$). There were no differences between groups in other causes of immunosuppression (solid organ transplant, solid cancer, hemopathy) or in chronic lung pathologies.

Table 2. Characteristics of the 63 Patients With Clinical Data and Comparison of Patients With *Streptomyces* Invasive Infections and Those Without Infection (Isolates Considered Contamination [n = 22] or Colonization [n = 3])

Characteristics	Total Population	Invasive Infection[a]	No Infection	*P*[a]
	n = 63	n = 30	n = 25	
Demography				
Age	59 (20)	60 (17)	60 (24)	.840
Male	39 (61.9)	20 (66.7)	13 (52.0)	.407
Comorbidities (n = 62)				
Chronic lung disease[c]	24 (38.7)	14 (46.7)	10 (40.0)	.823
Cardiovascular disease[d]	17 (27.4)	9 (30.0)	6 (24.0)	.847
Solid tumor[e]	16 (25.8)	9 (30.0)	7 (28.0)	.000
Diabetes (n = 61)	10 (16.4)	3 (10.3)	5 (20.8)	.499
Hematological malignancy[f]	9 (15.5)	6 (20.0)	3 (12.0)	.665
Solid organ transplant	5 (8.0)	4 (13.3)	1 (4.0)	.467
Moderate to severe kidney failure	2 (3.2)	1 (3.3)	1 (4.0)	.000
Charlson score	3.7 (2.8)	3.6 (2.4)	4.3 (3.3)	.407
Drug-induced immunosuppression (n = 62)	19 (30.6)	13 (43.3)	6 (24.0)	.224
Corticosteroids[g]	13 (20.9)	11 (36.7)	2 (8.0)	.030
Antimetabolites	8 (12.9)	6 (20.0)	2 (8.0)	.383
Antineoplastic chemotherapy	8 (12.9)	4 (13.3)	4 (16.0)	.000
Anticalcineurins/mTOR inhibitor	8 (12.9)	7 (23.3)	1 (4.0)	.101
Biological data				
CRP, mg/L	79 [86]	87.8 [92.6]	86.1 [86.3]	.952
Leukocyte count, Giga/L	8.9 [4.9]	7.9 [4.4]	9.8 [5.5]	.202
PMN, Giga/L	6.8 [4.2]	5.8 [3.5]	7.8 [4.6]	.136
Lymphocytes, Giga/L	1.5 [0.7]	1.3 [0.7]	1.6 [0.8]	.284
Creatinine, µmol/L	92 [62]	88 [44]	98 [87]	.731
Pulmonary samples				
Sputum culture	5 (7.9)	2 (6.7)	3 (12.0)	.830
Bronchial aspirate	15 (23.8)	8 (26.7)	7 (28.0)	.000
Bronchoalveolar lavage	9 (14.3)	8 (26.7)	1 (4.0)	.058
Direct examination of the pulmonary sample	18 (28.6)	7 (23.3)	7 (28.0)	.932
Coinfection	26 (41.3)	8 (26.7)	12 (48.0)	.225
Radiological findings				
Chest TDM	n = 32	n = 21	n = 10	
Micronodules	11 (34.3)	7 (33.3)	4 (40.0)	.735
Nodules	5 (15.6)	4 (19.1)	1 (10.0)	.467
Cavities	4 (12.5)	4 (19.1)	0 (0.0)	.844
Alveolar pattern	6 (18.75)	4 (19.1)	2 (20.0)	.844
Ground glass pattern	10 (31.2)	7 (33.3)	3 (15.0)	.463
Pleural effusion	2 (6.2)	1 (4.7)	1 (10.0)	.000
Normal	6 (18.75)	3 (14.3)	2 (20.0)	.000
Head imaging	n = 6	n = 4	n = 2	
Normal	4	2 (50.0)	1 (50.0)	.000
Other[h]	2	2 (50.0)	1 (50.0)	.000

Data are presented as No. (%) or mean [SD].

[a]The 8 patients with primary cutaneous infection were not included in the statistical analysis.

[b]Comparison of invasive *Streptomyces* infections with no infection in univariate analysis using the Fisher or chi-square tests for categorical variables and the Student *t* test for continuous variables or the Wilcoxon-Mann-Whitney *U* test when a nonparametric test was required.

[c]Chronic lung diseases = 23 patients: chronic obstructive pulmonary disease = 9, bronchiectasis = 3, lung cancer = 4, asthma = 3, obstructive sleep apnea = 1, pulmonary hypertension = 1, cystic fibrosis = 1, Sweet syndrome = 1, mechanical ventilation for diaphragmatic hernia = 1, lung surgery for prior lung cancer = 1, evanescent lung = 1; 4 patients had >1 chronic pulmonary disease.

[d]Cardiovascular diseases: myocardial infarction, congestive heart failure, obliterating arteriopathy, stroke.

[e]Solid tumor: metastatic status = 4.

[f]Hematological malignancies: lymphoma = 3, leukemia = 6.

[g]Corticosteroids: >10 mg/d for 3 months = 9.

[h]Other: findings not related to *Streptomyces* invasive infection.

For pulmonary samples, bronchoalveolar lavage cultures were positive in 8/30 (26.7%) patients with invasive infection compared with 1/25 (4.0%) in the no infection group ($P = .058$). There was no statistically significant difference between the groups in the percentage of direct examinations that were positive (7/30 [23.3%] vs 7/25 [28.0%]; $P = .932$) (Table 2).

DISCUSSION

In this retrospective analysis of 137 *Streptomyces* isolates, mostly identified in lung samples, we observed that the bacteria were most frequently susceptible to amikacin, linezolid, and imipenem. There are no guidelines for AST for *Streptomyces,* so comparing our results with other published data is difficult.

Nevertheless, susceptibility to imipenem was more frequent among our French isolates than among 92 *Streptomyces* isolates analyzed by the Centers for Disease Control and Prevention (CDC; 92.8 vs 67.0%, respectively) [19]. Use of different AST techniques may be responsible for these discrepancies: Although the broth microdilution method is recommended for AST of *Actinobacteria* [17], this technique may overestimate the rate of imipenem resistance, possibly because of lack of imipenem stability in broth microdilution [14]. Similar difficulties can be met when testing trimethoprim-sulfamethoxazole: If resistance is suspected using 1 method, the CLSI suggests the use of another technique, such as an E-test [14, 17]. In our study, the disk diffusion method overestimated the rate of resistance to trimethoprim-sulfamethoxazole, as most apparently "resistant" isolates were reclassified as being susceptible when tested using the E-test strip method. Such difficulties have previously been reported for testing of susceptibility of *Nocardia* spp. to imipenem and trimethoprim-sulfamethoxazole [20–22]. Though incompletely explained, these difficulties should be kept in mind by clinical microbiologists when studying *Streptomyces* or *Nocardia* isolates.

Focusing on the 30 patients with a final diagnosis of invasive *Streptomyces* infection, we observed that the lungs were the most frequently involved organ (20/30 cases, 66.6%), as previously reported (15/30, 50.0%) [8–10, 23–26]. *Streptomyces* pneumonia was diagnosed mainly from bronchoalveolar lavage samples in our cases and in previous reports [9, 10, 23, 24]. Because *Streptomyces* is an environmental pathogen, when invasive *Streptomyces* pulmonary infection is considered, deep specimen should be preferred over other lung samples to reduce the risk of contamination. The main lung CT scan features of invasive infection were crazy paving and micronodules, which is consistent with the previous literature [8, 10, 23, 24]. In addition to the pulmonary infections, we also had 2 cases of prosthetic endovascular infection, as also reported in the literature [12, 13]. Conversely, we had no cases of catheter-related bloodstream infection, although this forms part of the clinical spectrum of invasive *Streptomyces* infection [8, 11, 27, 28]. However, we were unable to obtain a clinical report form for 12/24 (50.0%) of the patients with *Streptomyces* BSI, and it is possible that some of them had catheter-related infection.

We had no cases of disseminated infection. However, compared with routine practice for *Nocardia* infection, imaging workup [29] is not systematic, so we are unable to draw definitive conclusions regarding the presence of disseminated infection.

A majority of the cases of invasive infection reported in the literature involved immunosuppressed patients. Most of these patients had systemic immunosuppression such as cancer [8, 11, 28], hemopathy [8], or drug-induced immunosuppression [27]. Other patients with lung involvement had local immunosuppression, either as a result of COPD or inhaled corticosteroids [9, 25]. By contrast with other reports, in our series, only half of the patients with invasive infection had ongoing, systemic, or local immunosuppression. Only corticosteroids were associated with invasive infection. Nonetheless, we cannot exclude that, given the small number of patients receiving systemic immunosuppressive drugs other than corticosteroids, our analysis was underpowered to detect a significantly higher rate of immunosuppression in the invasive infection group. Of note, no patient in the invasive infection group had undergone bone marrow transplant, and, interestingly, diabetes was not associated with invasive infection.

When identifying ramified gram-positive bacilli in a clinical sample from a patient with suspected ongoing infection, a multidrug regimen active against *Nocardia, Streptomyces,* and other gram-positive bacilli may be considered. In our series, linezolid, imipenem, and amikacin covered almost 100% of the *Streptomyces* isolates and would be active against most *Nocardia* isolates as well [14].

Treatment duration relies on the clinical resolution of symptoms, the organ involved, and associated complications. Overall, our data suggest that shorter antimicrobial duration may be necessary compared with treatment of *Nocardia* infections, with few relapses in our patients (13.6%) despite an average treatment duration of just 30 days. In the literature, a treatment duration of about 6 months was reported for most patients [9, 10, 30], although 4 patients had a 6-week course of treatment [12, 13, 24, 28]. All patients with known follow-up were cured, except for 3 [8, 11, 27].

We compared patients with invasive infection with those with no infection to identify factors that could help the physician to distinguish these 2 scenarios. In contrast to the previous literature, in our study a positive direct microscopic examination did not differ in these 2 subgroups. A previous report from Kapadia and coworkers suggested that clinical specimen with a positive direct Gram staining were more frequently *Streptomyces* infection (rather than contamination or colonization) [8]. However, direct examination may be difficult, as illustrated by a case report in which a negative initial direct

examination was later corrected after culture positivity for *Streptomyces* [19]. In this comparison, we decided to exclude primary cutaneous *Streptomyces* infections as we assumed that their mechanism of infection is different than invasive infection. Furthermore, the clinical diagnosis of primary cutaneous infections is less challenging considering the available literature in this field, although microbiological diagnosis of actinomycetoma remains difficult.

Our study has several limitations. As with any retrospective study, memory biases were present. To limit classification biases, cases were discussed between the physician completing the clinical record and the principal investigator when needed. Given the rareness of the infection, a prospective study to counteract the mentioned biases cannot be reasonably considered.

In conclusion, we report the largest series to date of 30 invasive *Streptomyces* infections, along with antibiotic susceptibilities of 113 *Streptomyces* isolates. When such an infection is considered, first-line antimicrobial therapy should include linezolid, imipenem, or amikacin, to which the isolates were most likely to be susceptible. Optimal treatment duration could be shorter than for *Nocardia* infection, but further data are needed to determine the appropriate management of invasive *Streptomyces* infection in terms of antibiotic choice(s) and duration. In addition, when the disk diffusion method is used for AST and trimethoprim-sulfamethoxazole resistance is suspected, a second attempt should be made using the E-test method with a lecture criterion set at 80% of inhibition to confirm this observation.

Supplementary Data

Supplementary materials are available at *Open Forum Infectious Diseases* online. Consisting of data provided by the authors to benefit the reader, the posted materials are not copyedited and are the sole responsibility of the authors, so questions or comments should be addressed to the corresponding author.

Acknowledgments

The authors would like to thank Fatima Zohra DJELOUAT for their help with data collection. The authors would like to thank Karen Pickett for her editorial assistance.

French* Streptomyces *Study Group (Collaborators). Anne Vachée, Department of Medical Biology, Hospital of Roubaix, Roubaix, France; Véronique Blanc, Department of Medical Biology, Hospital of Antibes, Antibes, France; Pascal Beuret, Department of Intensive Care Unit, Hospital of Roanne, Roanne, France; Olivier Moquet, Department of Biology, Hospital of Nevers, Nevers, France; Huong Porcheret, Department of Biology, Robert Ballanger Hospital, Aulnay-sous-Bois, France; Stéphane Lo, Bacteriology Department, Bichat Claude Bernard University Hospital, Assistance Publique—Hôpitaux de Paris, Paris, France, and INSERM, IAME UMR 1137, University of Paris, Paris, France; Raphaël Lepeule, Unité Transversale de Traitement des Infections, Département de Prévention, Diagnostic et Traitement des Infections, Henri Mondor Hospital, Assistance Publique—Hôpitaux de Paris, Créteil, France; Florence Huguet, Department of Radiation Oncology, Tenon Hospital, Assistance Publique—Hôpitaux de Paris, Paris, France; Vincent Bunel, Department of Pneumology B and Transplant, Bichat Hospital, Assistance Publique—Hôpitaux de Paris, Paris, France; Sandrine Picot, Department of Bacteriology, Hospital of Sud Réunion, Saint-Louis, France; Mustapha Terki, Department of Microbiology, Hospital of Gonesse, Gonesse, France.

Financial support. David Lebeaux was supported by the following grant: Bourse Junior 2015–Société de Pathologie Infectieuse de Langue Française (SPILF).

Potential conflicts of interest. All authors: no reported conflicts of interest. All authors have submitted the ICMJE Form for Disclosure of Potential Conflicts of Interest. Conflicts that the editors consider relevant to the content of the manuscript have been disclosed.

References

1. Barka EA, Vatsa P, Sanchez L, et al. Taxonomy, physiology, and natural products of *Actinobacteria*. Microbiol Mol Biol Rev **2016**; 80:1–43.
2. Euzeby J. List of bacterial names with standing in nomenclature: a folder available on the internet. Int J Syst Bacteriol **1997**; 47:590–2.
3. van de Sande WWJ. Global burden of human mycetoma: a systematic review and meta-analysis. PLoS Negl Trop Dis **2013**; 7:e2550.
4. Viguier M, Lafaurie M. Images in clinical medicine. Actinomycetoma. N Engl J Med **2015**; 372:264.
5. Mestre T, Vieira R, Coutinho J. Mycetoma of the foot—diagnosis of the etiologic agent and surgical treatment. Am J Trop Med Hyg **2015**; 93:1–2.
6. Szarf G, Obac AR, Puchnick A, et al. Mycetoma involving the heart. Circulation **2013**; 128:e159–61.
7. Cameron K, Bannon K, Mittal V. Abdominal wall mass. Fungal mycetoma. JAMA Surg **2014**; 149:743–4.
8. Kapadia M, Rolston KVI, Han XY. Invasive *Steptomyces* infections: six cases and literature review. Am J Clin Pathol **2007**; 127:619–24.
9. Kofteridis DP, Maraki S, Scoulica E, Tsioutis C, Maltezakis G, Gikas A. *Streptomyces* pneumonia in an immunocompetent patient: a case report and literature review. Diagn Microbiol Infect Dis **2007**; 59:459–62.
10. Riviere E, Neau D, Roux X, et al. Pulmonary *Streptomyces* infection in patient with sarcoidosis, France, 2012. Emerg Infect Dis **2012**; 18:1907–9.
11. Carey J, Motyl M, Perlman DC. Catheter-related bacteremia due to *Streptomyces* in a patient receiving holistic infusions. Emerg Infect Dis **2001**; 7:1043–5.
12. Mossad SB, Tomford JW, Stewart R, Ratliff NB, Hall GS. Case report of *Steptomyces* endocarditis of a prosthetic aortic valve. J Clin Microbiol **1995**; 33: 3335–7.
13. Shehatha JS, Taha AY. Early-onset *Steptomyces* endocarditis in a prosthetic aortic valve. Asian Cardiovasc Thorac Ann **2017**; 25:137–9.
14. Lebeaux D, Bergeron E, Berthet J, et al. Antibiotic susceptibility testing and species identification of *Nocardia* isolates: a retrospective analysis of data from a French expert laboratory 2010–2015. Clin Microbiol Infect **2019**; 25:489–95.
15. Rodríguez-Nava V, Couble A, Devulder G, Flandrois JP, Boiron P, Laurent F. Use of PCR-restriction enzyme pattern analysis and sequencing database for hsp65 gene-based identification of *Nocardia* species. J Clin Microbiol **2006**; 44:536–46.
16. Lebeaux D, Freund R, van Delden C, et al. Outcome and treatment of nocardiosis after solid organ transplantation: new insights from a European study. Clin Infect Dis **2017**; 64:1396–405.
17. Woods GL, Brown-Elliott BA, Conville PS, et al. Susceptibility Testing of Mycobacteria, Nocardiae, and Other Aerobic Actinomycetes. 2nd ed. Clinical and Laboratory Standards Institute; **2011**.
18. Charlson ME, Pompei P, Ales KL, MacKenzie CR. A new method of classifying prognostic comorbidity in longitudinal studies: development and validation. J Chronic Dis **1987**; 40:373–83.
19. Rose CE, Brown JM, Fisher JF. Brain abscess caused by *Steptomyces* infection following penetration trauma: case report and results of susceptibility analysis of 92 isolates of *Steptomyces* species submitted to the CDC from 2000 to 2004. J Clin Microbiol **2008**; 46:821–3.
20. Schlaberg R, Fisher MA, Hanson KE. Susceptibility profiles of *Nocardia* isolates based on current taxonomy. Antimicrob Agents Chemother **2014**; 58:795–800.
21. Valdezate S, Garrido N, Carrasco G, et al. Epidemiology and susceptibility to antimicrobial agents of the main *Nocardia* species in Spain. J Antimicrob Chemother **2017**; 72:754–61.
22. Larruskain J, Idigoras P, Marimón JM, Pérez-Trallero E. Susceptibility of 186 *Nocardia* sp. isolates to 20 antimicrobial agents. Antimicrob Agents Chemother **2011**; 55:2995–8.
23. Canouï E, Ingen-Housz-Oro S, Ortonne N, et al. [Hemophagocytic lymphohistiocytosis with granulomatosis and diffuse T-cell infiltration associated with disseminated nocardiosis and pulmonary infection due to *Steptomyces* spp]. Rev Med Interne **2019**; 40:457–61.
24. Ariza-Prota MA, Pando-Sandoval A, Fole-Vázquez D, García-Clemente M, Budiño T, Casan P. Community-acquired bacteremic *Steptomyces atratus* pneumonia in an immunocompetent adult: a case report. J Med Case Rep **2015**; 9:262.

25. Manteca A, Pelaez AI, del Mar Garcia-Suarez M, Hidalgo E, del Busto B. Mendez FJ. A rare case of lung coinfection by *Streptomyces cinereoruber* and *Haemophilus influenzae* in a patient with severe chronic obstructive pulmonary disease: characterization at species level using molecular techniques. Diagn Microbiol Infect Dis **2008**; 60:307–11.
26. Dunne EF, Burman WJ, Wilson ML. *Streptomyces* pneumonia in a patient with human immunodeficiency virus infection: case report and review of the literature on invasive *Steptomyces* infections. Clin Infect Dis **1998**; 27:93–6.
27. Ekkelenkamp MB, de Jong W, Hustinx W, Thijsen S. *Streptomyces thermovulgaris* bacteremia in Crohn's disease patient. Emerg Infect Dis **2004**; 10:1883–5.
28. Moss WJ, Sager JA, Dick JD, Ruff A. *Streptomyces bikiniensis* bacteremia. Emerg Infect Dis **2003**; 9:273–4.
29. Corsini Campioli C, Castillo Almeida NE, O'Horo JC, et al. Clinical presentation, management, and outcomes of patients with brain abscess due to *Nocardia* species. Open Forum Infect Dis **2021**; 8:XXX–XX.
30. Datta P, Arora S, Jain R, Chander J, van de Sande W. Secondary peritonitis caused by *Steptomyces viridis*. J Clin Microbiol **2012**; 50:1813–4.