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Nontuberculous mycobacteria in patients of a specialty hospital

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ABSTRACT

The incidence and clinical characteristics of NTM diseases in Brazil remain relatively unknown. The present study describes the diagnosis of NTM isolates, the clinical presentation and treatment outcomes. We analyzed NTM isolates in patients of a tertiary hospital in the Southeast region of Brazil, from January 2008 to July 2019. The ATS/IDSA criteria for diagnosis and treatment of these patients was applied. *Mycobacterium kansasii* were identified in 13/113 (11.5%) patients. In 59/113 (52.2%) patients who met the ATS criteria for disease, 29/59 (49.1%) received treatment, and 22/29 (75.8%) were cured. The major species identified was *M. kansasii*. The most frequent symptoms among the treated patients were dyspnea and cough, and the proportion of cured patients was high.

KEYWORDS: Mycobacteria. Sample. Diagnostic. Treatment.

INTRODUCTION

Nontuberculous mycobacteria (NTM) are a large and diverse group comprising more than 190 species and subspecies[1]. They colonize different environments such as soil, surfaces, and drinking water[1,2]. Reports of human disease caused by NTM have increased worldwide over the last two decades, with those of the *Mycobacterium abscessus* complex, *M. avium* complex (MAC), and *M. kansasii* frequently reported as being of greatest clinical importance[2,3].

NTM transmission is predominantly through the inhalation of aerosols and dust particles, contaminated water or soil, or aspiration of the gastric contents. Because many NTM are present in the environment, detection in nonsterile biological materials, such as sputum or urine, can be due to contamination or colonization, not necessarily implying disease[4,5].

Despite its low pathogenicity in humans, NTM disease more frequently displays predisposing factors, such as silicosis, bronchiectasis, chronic obstructive pulmonary disease, human immunodeficiency virus (HIV) infection, and immunosuppressive diseases[4,5].

Extrapulmonary NTM infections, particularly the disseminated disease, usually develop in people with congenital or acquired immunodeficiencies (e.g., PLHIV)[6], but can also be associated with medical or cosmetic procedures that expose a wound to sources contaminated with mycobacteria[6].

The difficulty of applying a uniform diagnostic criteria, imprecise characterization

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of pathogens, the lack of clinical reference data, and unknown proportion of people living with HIV (PLHIV) are important factors that hinder the proper evaluation of the epidemiological situation and NTM disease. In addition to these factors, diagnosis and drug sensitivity results delay the expensive treatment. Undesirable adverse drug effects also pose a problem, particularly relevant in low middle income countries[2,3,5].

In Brazil, there are few studies on NTM infection; its prevalence and geographically variable species are still not well known[7].

In 2007, the American Thoracic Society (ATS), in conjunction with the Infectious Diseases Society of America (IDSA), described the treatment criteria for patients with NTM[4]. These criteria were updated in 2020[5]. The treatment generally includes at least three drugs administered for ≥12 months, depending on the species and severity of the disease[5].

Thus, this study aims to evaluate NTM isolates, apply the ATS/IDSA criteria for diagnosis and treatment, and analyze the clinical data and outcomes in patients who received treatment at a specialty hospital.

MATERIALS AND METHODS

This study included analyses of patients with NTM isolates from inpatients and outpatients at a tertiary hospital at the Universidade Federal de Minas Gerais (UFMG). The data were collected between January 2008 and July 2019. All patients, with at least one positive sample from any part of the body with NTM, were eligible. However, only those who met the 2007 ATS/IDSA criteria – in the case of sputum at least two samples were required with identification of the same species[4], ≥ 18 years old with isolates of NTM or *Mycobacterium sp.* – answered a standardized questionnaire survey and had their medical records reviewed, which included clinical, mycobacteriological and radiological data, such as age, sex, symptoms, and comorbidities. For the diagnosis of infection by extrapulmonary NTM, clinical, mycobacteriological and imaging data were analyzed[4,5]. Cure criteria was defined according to van Ingen *et al.*[8].

Identification tests were performed using phenotypic methods[9] or enzyme restriction analysis of the hsp-65 gene (PRA-hsp65)[8] at the Fundacao Ezequiel Dias (FUNED). A drug susceptibility test was not conducted in this study. The nomenclatures of the genera, species, and subspecies were verified in the list of Prokaryotic Names with Standing in Nomenclature[10,11].

The treated patients were administered aminoglycosides for 2–3 months depending on the severity and presence or absence of pulmonary cavity, rifampin, ethambutol, macrolides (clarithromycin or azithromycin), and isoniazid. The latter was not prescribed when MAC was identified. The treatment duration for pulmonary and extrapulmonary diseases ranged from 12 to 18 months[4].

Data collected from the questionnaires were analyzed using SPSS Statistical Software (version 22.0, IBM, Armonk, NY, USA). Descriptive analyses were performed using the Statistical Analysis Software (version 9.4, SAS Institute Inc., Cary, NC, USA).

The UFMG Research Ethics Committee approved the study protocol under Nº 2.843.437 (CAAE 93672218.8.0000.5149).

RESULTS

We obtained 506 clinical samples from 113 patients, 129 of which were positive for NTM or *Mycobacterium sp*., with an annual average of 10 patients. Of these, 59 patients met the disease criteria and 29 received treatment (Figure 1).

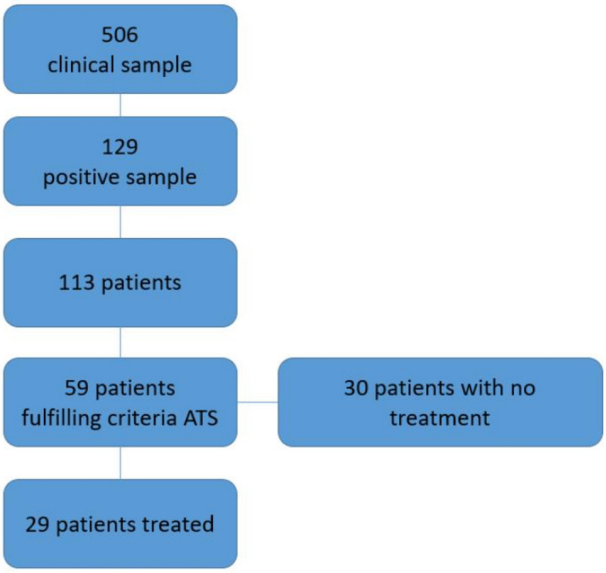

Figure 1 - Study population.

Of the 506 samples, 333 (65.8%) were sputum, 106 (20.9%) urine, 21 (4.2%) bronchoalveolar lavage, 14 (2.8%) tracheal aspirate, 10 (2%) liquor, 9 (1.7%) biopsies, 5 (1%) pleural liquid, 5 (1%) blood and 3 (0.5%) secretions. Of 113 NTM patients, *M. kansasii*, *M. fortuitum*, *M. gordonae*, and *M. intracellulare* were identified in 69 (61.1%) and *Mycobacterium spp.* in 44 (38.9%) patients. The cases of *M. gordonae* occurred in patients with an underlying predisposition (n=2) and transplant recipients (n=1) (Table 1). Although the pleural fluid and blood were not positive for mycobacteria, the samples isolated from other sites were positive.

Table 1 - Species of Nontuberculous Mycobacteria in 113 patients attended at the tertiary hospital of the Universidade Federal de Minas Gerais from 2008 to 2019.

Species of NTM	N	NTM with disease criteria	Treated patients
Mycobacterium sp.	44	19	9*
Mycobacterium kansasii	13	12	7
Mycobacterium fortuitum	8	5	2*
Mycobacterium gordonae	8	2	1
Mycobacterium intracellulare	7	3	2
Mycobacterium abscessus	5	1	0
Mycobacterium peregrinum	5	1	0
Mycobacterium chimaera	4	4	3*
Mycobacterium flavescens	4	2	0
Mycobacterium avium subsp. avium	3	2	2
Mycobacterium immunogenum	2	1	1*
Mycobacterium mucogenicum	2	2	0
Mycobacterium szulgai	2	0	0
Mycobacterium avium	1	0	0
Mycobacterium celatum	1	1	0
Mycobacterium chelonae	1	1	0
Mycobacterium abscessus subsp. bolletii	1	1	1*
Mycobacterium abscessus subsp. massiliense	1	1	1
Mycobacterium duvalii	1	0	0
Total	113	59	29

NTM = Nontuberculous Mycobacteria; *5 patients with extrapulmonary involvement (1 *Mycobacterium sp*., 1 *M. chimaera,* 1 *M. fortuitum*, 1 *M. abscessus subsp. bolletii*, 1 *M. immunogenum*)

Among 113 with NTM isolates, 59 (52.2%) patients met the disease criteria: 29 (49.1%) received treatment for pulmonary or extrapulmonary forms (Table 1). Of 20/29 (68.9%) patients who received treatment, NTM species was identified. Treatment was not instituted for other patients (n=30), as they were lost to follow-up. Among the 29 patients treated for NTM, 24 (82.8%) presented the pulmonary form (Table 2) and clinical characteristics (Table 3).

The NTM species of the patients treated with or without comorbidity are described in Table 4. The outcomes of the 29 patients who underwent treatment were as follows: 22 (75.8%) were cured, four (13.7%) died from NTM infection (*M. intracellulare*; n=2, *M. avium subsp. avium*; n=1, and *M. chimera*; n=1), and three (10.3%) were lost to follow-up. Serious adverse reactions were not observed. One patient with corneal lesion was previously described in another article[11].

DISCUSSION

Based on these results, in addition to identifying the high proportion of cured patients among the treated ones, it is important to identify patients with NTM disease as per the ATS/IDSA criteria.

The largest number of NTM are identified in the Southeast and South regions, which may be due to the increase in laboratory coverage and greater investment in laboratory techniques for diagnosis[6]. Our study identified the majority of the species (61.1%).

Efforts have been made in recent years by researchers at FUNED-MG to increase and improve the identification of NTM. Some studies report that the drug susceptibility testing have great value in guiding therapy, particularly when tested for aminoglycosides, rifampicin, and ethambutol, in addition to genotypic tests to detect mutations responsible for drug resistance[5,11,12]. Microbiological diagnosis, even if insufficient, is essential for the diagnosis of an infection. To achieve this, it is essential to correctly identify NTM's species[4,5].

Diseases caused by NTM are emerging worldwide, a fact that has been related to the improvement of diagnostic methods, population aging, and the presence of associated immunosuppressive diseases such as HIV infection, pulmonary sequelae, and smoking. Moreover, iatrogenic and nosocomial infections caused by NTM are

Table 2 - Sociodemographic and clinical characteristics of treated patients with Nontuberculous Mycobacteria (n=29).

Variables	N (%)
Age	
51-86	16 (55.1)
21-50	13 (44.9)
Gender	
Male	19 (65.5)
Female	10 (34.5)
Comorbidities	
Silicosis	2 (6.9)
HIV/Aids	5 (17.2)
COPD	1 (3.5)
Asthma	3 (10.3)
Organ transplant	4 (13.8)
Non-cystic fibrosis bronchiectasis	1 (3.5)
Chronic kidney disease	1 (3.5)
Cystic fibrosis	3 (10.3)
Sequelae to TB	3 (10.3)
Malignant neoplasm	2 (6.9)
No comorbidities	4 (13.8)
Tobacco use	
Never	10 (34.5)
Current	13 (44.8)
Ever	5 (17.2)
Unknown	1 (3.5)
Total	29

HIV/Aids = Human Immunodeficiency Virus; COPD = Chronic Obstructive Pulmonary Disease; TB = tuberculosis.

becoming more frequent, as are breast implant surgeries and ophthalmologic surgical procedures to correct refraction problems[1,13]. Importantly, the awareness of medical professionals regarding these diseases has also increased[6].

In our study, the pulmonary form was the most frequent, as described in other studies[6,13], associated with main preconditions like HIV/AIDS, organ transplantation, cystic fibrosis, TB sequelae, and being a current or former smoker, which corresponded to the profile of patients attended to in a tertiary hospital.

The signs and symptoms of the pulmonary form (most frequent) are similar to those of pulmonary TB. Patients with such symptoms may be inadequately treated for TB, since it is often only considered when there is treatment failure for TB and the patient is in a more advanced stage of the disease. Thus, the diagnosis requires a high index of suspicion and combined efforts of microbiologists and physicians[14].

Table 3 - Clinical characteristics of treated patients with pulmonary or extrapulmonary Nontuberculous Mycobacteria (n=29).

Variables	N (%)
Cough	
Yes	25 (86.2)
No	4 (13.8)
Expectoration	
Yes	21 (72.4)
No	8 (27.6)
Hemoptysis	
Yes	8 (27.6)
No	21 (72.4)
Night sweating	11 (37.9)
Yes	
No	17 (56.6)
Unknown	1 (5.5)
Fever	
Yes	19 (65.5)
No	10 (34.5)
Loss of appetite	
Yes	21 (72.4)
No	7 (24.1)
Unknown1 (3.5)	
Chest pain	
Yes	12 (41.3)
No	13 (44.8)
Unknown	4 (13.9)
Dyspnea	
Yes	26 (89.6)
No	3 (10.4)

Some studies have reported that MAC and *M. kansasii* are the most common pathogens that cause lung diseases[1,6]. However, *M. kansasii*, *M. chimera*, *M. fortuitum*, and *M. intracellulare* are the main species with respect to disease criteria and indications for treatment. These differences in results are probably due to geographic differences[15].

M. gordonae is commonly found in the environment and clinical laboratories and is almost always considered non-pathogenic. However, *M. gordonae* causes infections, particularly in patients with an underlying predisposition or immunosuppression, such as Acquired Immunodeficiency Syndrome (AIDS), undergoing peritoneal dialysis in transplant recipients[5]. Our study-enrolled patients had an underlying predisposition and were immunosuppressed.

Few studies have described the prevalence of NTM in the extrapulmonary form; however, these differences in

Table 4 - Description of the species of treated patients with Nontuberculous Mycobacteria and their comorbidities (n=29).

Comorbidities	Species of NTM
Silicosis	1 *Mycobacterium sp.* 1 *Mycobacterium avium subsp. avium*
HIV/Aids	2 *Mycobacterium kansasii* 1 *Mycobacterium avium subsp. avium* 1 *Mycobacterium sp.* 1 *Mycobacterium chimaera*
COPD	1 *Mycobacterium fortuitum*
Asthma	2 *Mycobacterium sp.* 1 *Mycobacterium intracellulare*
Organ transplant	3 *Mycobacterium sp.* 1 *Mycobacterium gordonae*
Non-cystic fibrosis bronchiectasis	1 *Mycobacterium sp.*
Chronic kidney disease	1 *Mycobacterium kansasii*
Cystic fibrosis	2 *Mycobacterium kansasii* 1 *Mycobacterium abscessus subsp. massiliense*
Sequelae to TB	1 *Mycobacterium sp.* 1 *Mycobacterium kansasii* 1 *Mycobacterium chimaera*
Malignant neoplasm	1 *Mycobacterium kansasii* 1 *Mycobacterium intracellulare*
No comorbidities	1 *Mycobacterium chimaera* 1 *Mycobacterium fortuitum* 1 *Mycobacterium abscessus subsp. bolletii* 1 *Mycobacterium immunogenum*

HIV/Aids = Human Immunodeficiency Virus; COPD = Chronic Obstructive Pulmonary Disease; TB = tuberculosis.

involvement may be due to environmental influences and the dynamics of the disease[12,16].

In some NTM cases that are treated as TB, the patient may positively respond to treatment, since drugs are effective in some NTM species, as for *M. kansasii*, which responds effectively to treatment even in the absence of macrolides[4,5,6].

In a study conducted in Brazil, the authors described that the cure rate of patients with MAC was 60.7%, whereas that of those infected with *M. kansasii* was 73.3%[17]. Regardless of species, the cure rate was higher (75.8%) in patients treated during this study.

The application of the "Recommendations for the diagnosis and treatment of diseases caused by nontuberculous mycobacteria" in Brazilian reference health units may contribute to greater success in the treatment of NTM.

This study had limitations in terms of the follow-up and outcomes of patients who did not undergo treatment.

CONCLUSION

We conclude that the major species identified in this study was *M. kansasii*. The most frequent symptoms of the patients who underwent treatment were dyspnea, coughing, sputum, and loss of appetite, and the proportion of cured patients was high among the treated patients.

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AUTHORS' CONTRIBUTIONS

GCSB idealized the work, collected the data, developed the experiments, built the database and interpreted the data; INA participated in the database verification and wrote the article; LBS participated in the database collection and wrote the article; CJA and EAL wrote and revised the article; CMP carried out the statistical analysis and adequacy of the database; ALAC adapted the database and revised the article; AFL, WSC, SSM and LJAF conceived and designed the study, analyzed the data and corrected the article; LJAF participated in data collection and analysis and wrote the article. The authors approve the final version of the manuscript and are responsible for all its aspects.

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