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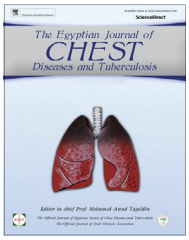

ORIGINAL ARTICLE

Risk factors for atypical mycobacterial disease in patients with smear positive pulmonary TB

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KEYWORDS

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Abstract Non Tuberculous Mycobacteria (NTM) can cause severe infection in selected groups of patients and is very difficult to be differentiated from TB infection clinically or radiologically leading to miss diagnosis and wrong treatment in these cases, the *Aim of the present study is* to study risk factors associated with NTM disease in patients with Acid Fast Bacilli (AFB) smear positive, *Subjects and methods:* 1402 patients with AFB smear positive were included in the study, only 47 patients from the study group proved to have NTM disease (diagnosis was done according to ATS/IDSA criteria). *Results:* the mean age of the NTM patients was 61.8 ± 23.2 years, NTM was more common in older age groups and more common in white race patients, on using logistic regression analysis NTM disease was more commonly associated with old TB infection (42.6%) and with bed ridden patients on tracheostomy (31.9%). The most common organisms isolated were the MAC complex (55.3%) followed by M. Kansasii (34.04%). *Conclusion:* NTM disease should be put into consideration in patients with AFB smear positive and suffering from old TB infection or in bed ridden patients who are on tracheostomy, also if smear is positive for AFB and PCR is negative NTM should be suspected.

Introduction

Pulmonary tuberculosis (TB) remains an important public health problem with an estimated 9.27 million new cases worldwide in 2007 [1]. According to the current treatment guidelines, isolation of *Mycobacterium tuberculosis* from a sputum culture is still recommended to confirm the diagnosis of pulmonary TB [2]. Non-tuberculous mycobacteria (also called atypical mycobacteria, mycobacteria other than tuberculosis, environmental mycobacteria, opportunistic mycobacteria) are ubiquitous pathogens and have been found in soil, domestic tap water, and in animals [3,4]. Although exposure to non-

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Table 1 Clinical and microbiologic criteria for diagnosing nontuberculous mycobacterial (NTM) lung disease.

Clinical (both required)
1. Pulmonary symptoms, nodular or cavitary opacities on chest radiograph, or a high-resolution computed tomography scan that shows multifocal bronchiectasis with multiple small nodules
2. Appropriate exclusion of other diagnoses
And
Microbiologic
1. Positive culture results from at least two separate expectorated sputum samples. If the results from [1] are nondiagnostic, consider repeat sputum AFB smears and cultures
Or
2. Positive culture results from at least one bronchial wash or lavage
Or
3. Transbronchial or other lung biopsy with mycobacterial histopathologic features (granulomatous inflammation or AFB) and positive culture for NTM or biopsy showing mycobacterial histopathologic features (granulomatous inflammation or AFB) and one or more sputum or bronchial washings that are culture positive for NTM
4. Expert consultation should be obtained when NTM are recovered that are either infrequently encountered or that usually represent environmental contamination
5. Patients who are suspected of having NTM lung disease but do not meet the diagnostic criteria should be followed until the diagnosis is firmly established or excluded
6. Making the diagnosis of NTM lung disease does not, per se, necessitate the institution of therapy, which is a decision based on potential risks and benefits of therapy for individual patients

tuberculous mycobacteria often occurs without any clinical manifestations, there are differences in the virulence of these mycobacteria and clinical manifestations may range from no symptoms or signs to destructive or even fatal disease [5]. Also with advances in the recognition of Nontuberculous mycobacterium (NTM), prior studies have addressed the difficulty in distinguishing TB from NTM by using either clinical symptoms or imaging [5,6]. The decision to initiate TB treatment should be based on epidemiology, clinical/radiographic findings, and the results of acid-fast bacilli (AFB)-stained sputum. In endemic areas, it is not uncommon to administer anti-TB treatment empirically pending culture results, given the clinical features suggesting pulmonary TB [3,4]. Although this strategy aims at better disease transmission control, it comes with a price: it is possible to inappropriately treat patients without pulmonary TB with anti-TB drugs, leading to adverse effects and unnecessary costs. In addition the isolation prevalence of NTM has also increased gradually, further complicating the problem [7]. Sputum AFB staining is one of the most readily accessible tools for evaluating patients suspected of having pulmonary TB; however, it is not specific for pulmonary TB [4,8]. Pulmonary non-tuberculous mycobacteria (NTM) infection can cause severe progressive illness which may be preceded by a period of colonization [9,10]. Treatment of pulmonary NTM is complicated, requiring multiple antimycobacterial drugs for >12 months [9,10]. Patients are treated based on sputum smear exams using standard first and second line TB therapy depending on clinical criteria in conjunction with the World Health Organization (WHO) guidelines [11]. As NTM are often resistant to first-line anti-TB medication, presumably many of these cases would be considered treatment failures, and subsequently treated for multi-drug resistant (MDR) disease [12]. Colonization, on the other hand, may be defined by the absence of one or more diagnostic criteria [12]. In the setting of colonization, treatment is often withheld owing to its potential toxicities and the uncertain rate of progression. Ideally, treatment should be initiated before irreversible lung damage occurs or progresses [13].

Aim of the work

The aim of the present study is to study the risk factors associated with Non-Tuberculous (NTM) mycobacterial disease in patients with smear positive pulmonary TB.

Subjects

The study was done in the pulmonary rehabilitation center, ministry of health, state of Kuwait. Pulmonary rehabilitation center is the main center in the state of Kuwait that is specified to diagnose and treat Tuberculosis and all cases of TB whether pulmonary or extra-pulmonary are referred to the center for evaluation and putting the plane of treatment, also pulmonary rehabilitation center is the supervisor of the central TB laboratories specified for microbiological diagnosis of TB. 1402 patients with AFB smear positive sputum or BAL admitted to the center were included in the present study, cases were admitted for isolation and more assessment to confirm the diagnosis of pulmonary TB and to start specific treatment, the study was done in the period from 1/1/2010 to 30/6/2013, from these patients 47 patients were confirmed to have NTM disease (according to ATS criteria Table1).

Methods

All patients were subjected to the following:

1- History taking.
2- Physical examination.
3- Three successive samples of Sputum for AFB were collected in the early morning for 3 successive days. If the patient was not able to give sputum spontaneously induction of sputum using a hypertonic saline nebulizer in the early morning preceded by salbutamol inhalation or bronchoscopy and broncho-alveolar lavage was used to get the sample for bacteriological analysis, all sputum

or BAL samples were sent for direct smear examination using Ziehl–Neelsen stain and culture for mycobacterial tuberculosis obtained using solid and liquid media and samples were incubated for 8 weeks before the final results were declared.
4- In bed-ridden cases on tracheostomy tracheal aspirate were sent for culture and sensitivity for acid-fast bacilli.
5- TB polymerase chain reaction (PCR) performed with in-house IS6110-based PCR assays [14] was done in sputum or BAL samples positive for AFB and suspected to be atypical mycobacteria.
6- Chest X ray.
7- Computed tomography (CT chest) with contrast was done in all suspected cases of being atypical mycobacteria infection.
8- In cases identified to be Non-Tuberculous Mycobacteria (NTM) isolates were identified by DNA probes (AccuProbe; Gen-Probe Incorporated and GenoType line-probe assays; Hain Life Science.
9- NTM disease was diagnosed according to ATS/IDSA criteria (Table 1) [15].

Statistical analysis

All statistical analyses were conducted using the software package SPSS 20.0 for Windows® (SPSS Inc., Chicago, IL, USA). Data are presented as frequencies for categorical variables, and by mean ± standard deviation for numerical variables. Categorical variables were compared using a chi-square test, and continuous variables were compared using an independent unpaired *t*-test. Multivariate analysis was conducted using a logistic regression model to determine the independent predictive factors for atypical mycobacteria disease among cases of smear positive pulmonary TB. *p*-values less than 0.05 were considered significant.

Results

From 1-1-2010 to 30-6-2013 out of the 1402 patients with sputum or BAL smear positive for AFB; 47 patients were confirmed to have NTM disease (according to ATS criteria Table 1).

From Table 2: NTM disease was found mainly in older age groups (mean age was 61.8 years) although there was a wide range for the age distribution, NTM was more common in males than in females and was more common in the white race. The most common co-morbidity associated with NTM disease was old TB infection which was present in 20 patients (42.6%) followed by bed ridden patients who were on tracheostomy which was found in 15 patients (31.9%), this was followed by COPD which was found in 7 patients (14.9%). Table 3 demonstrates the clinical characters of the NTM patients.

From Table 4: the most common radiographic feature detected in these patients was the nodular or reticulonodular infiltrate which was present in 36 patients (76.6%) followed by consolidation which was present in 17 patients (36.2%), disease was bilateral in 19 patients (40.4%) and was unilateral in 28 patients (59.6%).

From Table 5: 141 samples were studied (through sputum, BAL or tracheal aspirate), from these samples 120 (85.1%) samples were positive for AFB and 126 (89.4%) samples showed positive culture for NTM with a minimum of 2 cultures for each patient , PCR test for mycobacteria TB was negative in all samples whether positive or negative for AFB. On using Gene probe identification of the NTM species the most isolated organism was MAC complex (mycobacteria avium–intercellulare complex) which was present in 26 patients (55.3%) followed by mycobacteria Kansasii which was present in 16 patients (34.04%), *Mycobacteria Abscessus* was present in 4 patients (8.53%) and *Mycobacteria gordoni* was present in 1 (2.13%) patient. On using multivariate logistic regression analysis to identify risks associated with NTM disease among AFB smear positive cases the only 2 variables that showed significant correlation were old TB infection and bed ridden patients on tracheostomy ($p < 0.05$).

Discussion

The aim of the present study was to study the possible risk factors associated with non tuberculous mycobacterial disease in smear positive pulmonary TB patients, at the center in which the study was done is the main center in the state of Kuwait dealing with Tuberculosis; from every patient admitted to the center 3 samples of sputum, induced sputum, tracheal aspirate or BAL were sent for AFB direct examination and at the same time for AFB culture and sensitivity to be sure

Table 2 Epidemiologic criteria and co-morbidities associated with NTM Disease.

Age in years	61.8 ± 23.2
Age range	36–83
Male:female	29 (61.7%)–18 (38.3%)
Ethinisty	White:non white = 30/17
Co-morbidities associated with NTM disease	
Old TB history	20 (42.6%)
Bed ridden on tracheostomy	15 (31.9%)
COPD	7 (14.9%)
Malignancy	3 (6.3%)
Bronchiectasis	2 (4.3%)

Table 3 Clinical characteristics of NTM patients.

Chronic cough	32 (68%)
Sputum production	30 (63.8%)
Fever	22 (46.8%)
Hemoptysis	5 (10.6%)
Chest pain	7 (14.9%)

Table 4 Radiographic characters (by CXR and CT chest) of the NTM patients.

Unilateral vs Bilateral	19 (40.4%):28 (59.6%)
Consolidation	17 (36.2%)
Cavitation	7 (14.9%)
Nodular, reticulonodular	36 (76.6%)
Honeycombing	4 (8.5%)
Pleural effusion	2 (4.3%)

Table 5 Microbiology characters of NTM patients.

No. of NTM patients	47
No. of AFB smear positive samples	120 (85.1%) of 141 samples
PCR for mycobacteria TB	−ve in all positive and negative samples (100%)
Culture +ve for NTM	126 (89.4%)
Gene probe identification for NTM (*Done in 47 patients*)	
MAC	26 (55.3%)
Kansasii	16 (34.04%)
Abscessus	4 (8.53%)
Gordoni	1 (2.13%)

of the diagnosis and to detect drug sensitivity, as NTM disease is very difficult to be differentiated from pulmonary TB on clinical or radiological features mycobacterial culture results can be the corner stone to solve the problem and avoid the un-necessary isolation of these patients and in the same time it helps in prescribing the correct drug regime for these patients [15]. In the present study the main risk factor associated with atypical mycobacterial disease was the presence of old TB infection (42.6% of NTM cases had old TB), old TB causes parenchymal destruction in the lung with fibrosis or cavitary changes leading to reduction in the physiologic protective mechanisms of the lung against invasion by bacteria, and as NTM are non-virulent organisms the diseased lung constitutes good media for invasion by these organisms, on the other hand the patients with old TB infection may be at increased risk for poor nutrition, poor hygienic measures in the surrounding environment leading to increased risk for NTM disease [16–19]. Da costa et al. in 2013 studied the occurrence of NTM infection in an endemic area of tuberculosis, the NTM infection constituted 13.5% of positive mycobacterial cultures over 2-years period, a major risk factor for NTM pulmonary disease was previous tuberculosis (76%) [20]. The second risk factor associated with NTM in the present study was bed ridden patients who are on tracheostomy for long periods (31.9% of cases), these patients have deficiency in the natural protective mechanisms of the upper airways and they are in continuous contact with the external environment together with the impaired cough reflex in most of these patients leading to easily invasion of the lungs by opportunistic organisms like NTM, adding to this is the nutritional deficiency and other co-morbidities associated with bed ridden condition leading to decrease in the immune status of these patients. In a study done by Derac et al. in 2012 they used a comparative, population-based design to examine aerosol-generating behaviors in the home and garden and several pulmonary and non-pulmonary conditions as potential risk factors for *Mycobacterium avium* complex lung disease in an HIV-negative, general adult population and they concluded that aerosol-generating activities seem not to be risk factors for *Mycobacterium avium* complex lung disease in HIV-negative adults, but prior lung disease and immune-suppressing drugs seem to be associated with susceptibility [21]. On raising the concept of colonization of NTM in these patients all patients included in the present study had at least 2 positive cultures of NTM associated with newly developed clinical manifestations or radiologic manifestations, the concept of airway colonization by NTM has never been tested rigorously. There is not enough known about the pathophysiology of NTM lung disease to be sure that colonization is not, in fact, an indolent or slowly progressive infection. No pathologic studies have been done to demonstrate the absence of tissue invasion, and more recent studies with HRCT have shown that these patients often have a combination of multifocal bronchiectasis and nodular parenchymal disease now believed to be due to mycobacterial disease [15,22,23]. As regards the gene probe identification the most common organism encountered in the present study was *Mycobacterium avium* complex (MAC) which was found in 55.3% of the cases followed by M. Kansasii which was found in 34.04% of the cases, the NTM distribution is different from one region of the world to the other, lack of large studies and reporting data adds to difficulty in interpreting data. In the last 2 reports published in the USA about the prevalence of Non-Tuberculous Mycobacteria MAC was the most common to be isolated followed by M. fortuitum then M. Kansasii [24,25] while in the study done by Derac et al. in 2012 which was done in Brazil the distribution was different as the rapidly growing *Mycobacterium massiliense* and *Mycobacterium simiae* complex were most dominant [21].

Conclusion

NTM disease should be put into consideration in patients with AFB smear positive and suffering from old TB infection or in bed ridden patients who are on tracheostomy, also if smear is positive for AFB and PCR is negative NTM should be suspected.

Conflict of interest

We have no conflict of interest to declare.

References

[1] P.R. Donald, P.D. van Helden, The global burden of tuberculosis – combating drug resistance in difficult times, N. Engl. J. Med. 360 (23) (2009) 2393–2395.
[2] H.M. Blumberg, W.J. Burman, R.E. Chaisson, et al, American thoracic society. Centers for disease control and prevention and the infectious diseases society American thoracic society/centers for disease control and prevention/infectious diseases society of America: treatment of tuberculosis, Am. J. Respir. Crit. Care Med. 167 (4) (2003) 603–662.
[3] J. Banks, Environmental Mycobacteria, in: PDO Davies (Ed.), Clinical tuberculosis, Chapman and Hall, London, 1994, pp. 265–275.
[4] E. Wolinsky, Non-tuberculosis mycobacteria and associated disease – state of the art, Am. Rev. Respir. Dis. 119 (1979) 107–159.

[5] N. AlJarad, P. Demertzis, D.J. Meecham Jones, et al, Comparison of characteristics of patients and treatment outcome for pulmonary nontuberculous mycobacterial infection and pulmonary tuberculosis, Thorax 51 (1996) 137–139.
[6] M. Maiga, S. Siddiqui, S. Diallo, et al, Failure to recognize nontuberculosis mycobacteria leads to misdiagnosis of chronic pulmonary tuberculosis, PLoS One 7 (5) (2012) e36902.
[7] T.K. Marras, P. Chedore, A.M. Ying, F. Jamieson, Isolation prevalence of pulmonary non-tuberculosis mycobacteria in Ontario, 1997–2003, Thorax 62 (8) (2007) 661–666.
[8] Diagnostic standards and classification of tuberculosis in adults and children. This official statement of the American thoracic society and the centers for disease control and prevention was adopted by the ATS Board of directors, Jul 1999. This statement was endorsed by the council of the infectious disease society of America, September 1999, Am. J. Respir. Crit. Care Med. 161 (4 Pt 1) (2000) 1376–1395.
[9] American thoracic society diagnosis and treatment of disease caused by nontuberculous mycobacteria, Am. J. Respir. Crit. Care Med. (1997) (156S1–19.19).
[10] British thoracic society management of opportunist mycobacterial infections: joint tuberculosis committee guidelines 1999, Thorax (2000) (55210–218.218).
[11] World Health Organisation website, fourth ed. of treatment of tuberculosis/guidelines (2009), Available: <http://www.who.int/tb/publications/2010>.
[12] Theodore K. Marras, Pamela Chedore, Alicia M. Ying, Frances Jamieson, Isolation prevalence of pulmonary non-tuberculosis mycobacteria in Ontario, 1997–2003, Thorax 62 (8) (2007 August) 661–666.
[13] L.O. Larsson, M.W. Bentzon, A. Lind, et al, Sensitivity to sensitins and tuberculin in Swedish children. Part 5: a study of school children in an inland rural area, Tuber. Lung Dis. 74 (1993) 371–376.
[14] J.R. Sun, S.Y. Lee, C.L. Perng, J.J. Lu, Detecting *Mycobacterium tuberculosis* in Bactec MGIT 960 cultures by inhouse IS6110-based PCR assay in routine clinical practice, J. Formos. Med. Assoc. 108 (2) (2009) 119–125.
[15] American Thoracic Society/Infectious Disease Society of America, An official ATS/IDSA statement: diagnosis, treatment, and prevention of nontuberculous mycobacterial diseases, Am. J. Respir. Crit. Care Med. 175 (2007) 367–416.
[16] E. Wolinsky, Nontuberculosis mycobacteria and associated diseases, Am. Rev. Dis. 119 (1) (1979) 107–159.
[17] T.K. Marras, C.L. Daley, Epidemiology of human pulmonary infection with nontuberculosis mycobacteria, Clin. Chest Med. 23 (3) (2002) 553–567.
[18] K.B. Gupta, R. Gupta, A. Atreja, M. Verma, S. Vishvkarma, Tuberculosis and nutrition, Lung India 26 (1) (2009) 9–16.
[19] H. Gruft, J.O. Falkinham 3rd, B.C. Parker, Recent experience in the epidemiology of disease caused by atypical mycobacteria, Rev. Infect. Dis. 3 (5) (1981) 990–996.
[20] A.R.F. da Costa, J.O. Falkinham III, M.L. Lopes, A.R. Barretto, et al, Occurrence of nontuberculous mycobacterial pulmonary infection in an endemic area of tuberculosis, PLoS Negl. Trop. Dis. 7 (7) (2013) e2340, http://dx.doi.org/10.1371/journal.pntd.0002340.
[21] M.A. Dirac, K.L. Horan, D.R. Doody, J.S. Meschke, D.R. Park, et al, Environment or host?: a case-control study of risk factors for *Mycobacterium avium* complex lung disease, Am. J. Respir. Crit. Care Med. 186 (7) (2012) 684–691.
[22] E.F. Patz, S.J. Swensen, J. Erasmus, Pulmonary manifestation of nontuberculosis mycobacteria, Radiol Clin. North Am. 33 (1995) 719–729.
[23] Y.J. Jeong, K.S. Lee, W.J. Koh, J. Han, T.S. Kim, O.J. Kwon, Nontuberculous mycobacterial pulmonary infection in immunocompetent patients: comparison of thin-section CT and histopathologic findings, Radiology 231 (2004) 880–886.
[24] S. Ostroff, L. Hutwagner, S. Collin, Mycobacterial species and drug resistance patterns reported by state laboratories, the 93rd American Society for Microbiology General Meeting, May 16, 1993, Atlanta, GA. 1992, Abstract U-9, p. 170.
[25] Centers for Disease Control and Prevention, Nontuberculous mycobacteria reported to the public health laboratory information system by state public health laboratories, United States, 1993–1996, cdc.gov.