Bronchial Wash Culture is Less Valuable in Patients Suspected to have Nontuberculous Mycobacteria Lung Disease for Bilateral Bronchiectasis with Nodules

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Abstract

Backgrounds: Bronchiectasis (BE) with nodules on chest computed tomography (CT) is one of the radiological findings of nontuberculous mycobacteria (NTM) lung disease. However, the NTM lung disease is not diagnosed in some patients who undergo bronchoscopy, though their radiological findings show BE with nodules. We need the indicator to distinguish the patients who would not be given the diagnosis of NTM lung disease with bronchoscopy. Methods: We retrospectively reviewed the records of the patients who underwent bronchoscopy from January 2010 to July 2013 at our hospital because sputum test had not yielded the diagnosis of NTM lung disease or the patients had been unable to produce sputum, though their chest CT had shown BE with nodules. Results: A total of 36 patients were included in this study, and acid-fast bacterium (AFB) culture of bronchial wash was positive in 17 patients. More patients with negative culture in bronchial wash had AFB-negative culture with purulent sputum before bronchoscopy (37% vs. 0%, P = 0.008) and bilateral BE with nodules on chest CT (89% vs. 41%, P = 0.007). Multivariate analysis showed that bilateral BE with nodules on chest CT was the risk factor for the negative culture of AFB with bronchial wash (odds ratio: 0.149; 95% confidence interval: 0.024–0.913, P = 0.040). Conclusion: Patients with bilateral BE with nodules on chest CT have less possibility to have positive AFB culture from bronchial wash when sputum test was not diagnostic.

Keywords: Bronchoscopy, computed tomography, Mycobacterium avium, Mycobacterium intracellulare, nontuberculous mycobacteria

Introduction

Nontuberculous mycobacteria (NTM) lung disease is common and has reported to increase worldwide. 1-6 Bronchiectasis (BE) with nodules on chest computed tomography (CT) is one of the radiological findings of NTM lung disease. 7 However, there exists many differential diagnoses for BE with nodules on chest CT. 8 To make the diagnosis of NTM lung disease, bronchoscopy is needed for the patients who are suspected to have NTM lung disease, when they have negative sputum culture for acid-fast bacterium (AFB) or have no sputum. However, the NTM lung disease is not diagnosed in some patients who undergo bronchoscopy, though their radiological findings are compatible with NTM lung disease. Bronchoscopy is relatively safe, but complications occur to some extent, 9 and we must omit needless examination. Therefore, we need the indicator to distinguish the patients who would not be given the diagnosis of NTM lung disease with bronchoscopy. We retrospectively reviewed the records of the patients who had undergone bronchoscopy because sputum test had not yielded the diagnosis of NTM lung disease, though their CT had showed BE with nodules. Then, we compared the characteristics of the patients whose bronchial washing culture was positive for NTM and those whose bronchial washing culture was negative for NTM.

Methods

This study was approved by the Review Board of our hospital (NO. D1409007). Due to the retrospective nature of this study, written informed consent was waved.

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We retrospectively reviewed the records of the patients who underwent bronchoscopy from January 2010 to July 2013. Inclusion criteria were as follows:
1. The patients who were suspected to have NTM lung disease because of BE with nodules on chest CT
2. No sputum or negative sputum culture for AFB
3. Age older than 20 years old.

Patients who were known to be serologically positive for human immunodeficiency virus were excluded from the study.

Sputum appearance was evaluated according to the classification of Miller. Transbronchial lung biopsy (TBLB) was performed from the segment where heavy small nodular opacities existed. The presence of granuloma was judged by the pathologists at our hospital who were independent of our study. Bronchial washing was performed after TBLB. Twenty milliliters sterile saline was injected into the segmental or subsegmental bronchus which TBLB was performed. Gram stains and the culture for AFB, other bacteria, and fungi were performed with the specimen of bronchial wash. The specimen of bronchial wash was screened with Auramine O for AFB smear testing, and positive for AFB was confirmed with Ziehl–Neelsen staining. Liquid culture medium (Bectec MGIT 960 apparatus; Becton Dickinson, Franklin, New Jersey, USA) was used for AFB culture. NTM was identified by DNA hybridization assay (DDH Mycobacteria assay; Kyokuto Pharmaceuticals Co., Tokyo, Japan).

The diagnosis of NTM lung disease was made with the diagnostic criteria of NTM lung disease proposed by ATS.

**Statistical analysis**

Data were presented as mean ± standard division, if not otherwise specified. SPSS statistics version 19.0 (IBM, Armonk, New York, USA) was used for statistical analyses. The comparison was made with the t-test or Mann–Whitney U-test for continuous variable. The categorical variable was compared with Pearson’s Chi-square test or Fisher’s exact test. Multivariate analysis was performed by logistic regression analysis for the factors which showed P < 0.1 by univariate analysis. Statistical significance was set at P < 0.05 and all tests were two tailed.

**RESULTS**

Thirty-six patients were included in this study. Table 1 shows the characteristics of the patients. Twenty-seven patients were female and mean age was 67.5 ± 7.0 years old. Twenty-four patients had bilateral BE with nodules on chest CT. Twenty-four patients had a negative culture for AFB with sputum and 12 were unable to produce sputum. Of 24 patients whose sputum culture was negative for AFB, seven patients had purulent sputum. About the underlying disease, six patients (17%) had collagen vascular diseases, five patients (14%) had active malignancy, four patients (11%) had diabetes, and two patients (6%) had chronic respiratory diseases. Three patients (8%) received immunosuppressant. Table 2 shows the results of TBLB and bronchial wash culture. Thirty-two patients underwent TBLB and five patients (16%) had granuloma with TBLB. All patients underwent bronchial wash. AFB smear with bronchial wash was positive in seven patients (19%), and AFB culture was positive in 17 patients (47%). Of those with positive bronchial wash culture for AFB, 15 patients (88%), two patients (12%), and one patient (6%) had positive bronchial wash culture for Mycobacterium avium, Mycobacterium intracellulare, and Mycobacterium kansasii, respectively. One patient had positive results for both M. avium and M. intracellulare. All the patients with positive bronchial wash culture for AFB met the criteria for NTM lung disease proposed by ATS.

**Table 1: Characteristics of the patients (n=36)**

| Characteristic                              | Number (%) |
|--------------------------------------------|------------|
| Female:male                                | 27:9       |
| Age (years old)                            | 67.5±7.0   |
| Symptoms of the initial visit, number of the patients (%) |           |
| Cough                                      | 14 (39)    |
| Fever                                      | 4 (11)     |
| Sputum                                     | 13 (36)    |
| BMI (kg/m²)                                | 20.5 (19.0‐21.9) |
| Smoking history (current/ex/never)          | 3/11/22    |
| Laboratory data                            |            |
| WBC (/μl) (n=36)                           | 5740±1700  |
| CRP (mg/dl) (n=36)                         | 0.152 (0.050-0.354) |
| ESR (mm/h) (n=27)                          | 16 (6-34)  |
| BE with nodules on CT                      | 24:12      |
| Nature of sputum for culture (n=24)        |            |
| Purulent:mucinous                          | 7:17       |
| Underlying conditions, number of the patients (%) |               |
| Collagen vascular disease                  | 6 (17)     |
| Active malignancy                          | 5 (14)     |
| Diabetes                                   | 4 (11)     |
| Chronic respiratory diseases               | 2 (6)      |
| Use of immunosuppressant                   | 3 (8)      |

Data are presented as mean±SD, median (25 percentile–75 percentile) or number of the patients (%), if not otherwise specified. BMI: Body mass index, WBC: White blood cell, CRP: C-reactive protein, ESR: Erythrocyte sedimentation rate, BE: Bronchiectasis, CT: Computed tomography, SD: Standard deviation

**Table 2: Results of transbronchial lung biopsy and bronchial wash**

| Criteria                                      | Number (%) |
|-----------------------------------------------|------------|
| Granuloma with TBLB (n=32) (%)               | 5 (16)     |
| Bronchial wash (n=36) (%)                    |            |
| AFB smear positive                           | 7 (19)     |
| AFB culture positive                         | 17 (47)    |
| Mycobacterium avium                         | 15 (88)    |
| Mycobacterium intracellulare                | 2 (12)     |
| Mycobacterium kansasii                      | 1 (6)      |

Data are presented as the number of the patients (%). One patient had positive results for both Mycobacterium avium and Mycobacterium intracellulare. TBLB: Transbronchial lung biopsy, AFB: Acid-fast bacillus
Table 3 shows the comparison with the patients having AFB-positive culture and those having AFB-negative culture in bronchial wash. Body mass index (BMI), white blood cell, C-reactive protein, erythrocyte sedimentation rate, and underlying conditions did not differ between two groups. However, more patients with negative culture in bronchial wash had AFB-negative culture with purulent sputum before bronchoscopy (37% vs. 0%, P = 0.008) and bilateral BE with nodules on chest CT (89% vs. 41%, P = 0.007). Logistic regression was performed for bilateral BE with nodules on chest CT, AFB-negative culture with purulent sputum before bronchoscopy, and C reactive protein. Bilateral BE with nodules on chest CT was the risk factor for the negative culture of AFB with bronchial wash (odds ratio [OR]: 0.149; 95% confidence interval [CI]: 0.024–0.913, P = 0.040) [Table 4]. All the patients who had AFB-negative culture in bronchial wash did not meet the diagnostic criteria of NTM lung disease proposed by ATS11 during the follow-up period (median follow-up period was 18.9 months).

**DISCUSSION**

Almost 34% to 53% of the patients whose CT showed BE with nodules had the positive culture for *M. avium-intracellulare* complex (MAC).[8,12,13] However, these studies did not limit the patients whose sputum test had not yielded the diagnosis of NTM lung disease. In this study, 47% of the patients with BE with nodules had bronchial wash culture positive for AFB though their sputum test had not yielded the diagnosis of NTM lung disease. These data showed the usefulness of bronchoscopy for the patients who were suspected to have NTM lung disease because of BE and nodules on chest CT and whose sputum test had not yielded the diagnosis. However, in 53% patients, bronchoscopy was not diagnostic. If we can predict the patients who will have negative AFB culture with bronchial wash, fewer patients would receive needless examination. In this study, bilateral BE with nodules was the risk factor for negative AFB culture with bronchial wash. When the area of opacity is wide, the burden of NTM is heavy, and it was speculated that sputum culture is more likely to be positive in such cases. Thus, when the patients with bilateral BE with nodules have negative sputum culture of AFB, it might be controversial for NTM lung disease. They might have less possibility to have NTM lung disease, and other diagnoses should be considered for these patients. Moreover, purulent sputum is suitable for detecting the pathogen. The patients might have less possibility to have NTM lung disease, when they have negative AFB culture with purulent sputum, although OR of negative AFB culture with purulent sputum for predicting negative AFB culture with bronchial wash could not be calculated due to a small number of patients in this study. Recently, the usefulness of serological test for MAC has been reported.[14–19] The necessity of bronchoscopy to diagnose the MAC lung disease might decrease. However, the sensitivity is not adequate in some studies with 60% to 77%.[15,17,18] Especially, sensitivity of 43% is reported in rheumatoid arthritis patients.[19] It might be difficult to deny MAC lung disease with negative MAC antibody. In addition, to gain cultivated strain is important for two reasons other than diagnostic value. First, Clarithromycin (CAM) is the key drug of chemotherapy for *M. avium* and *M. intracellulare* and the choice of the regimen is depended on the susceptibility to CAM according to ATS guideline.[21] Second, *M. avium* lung disease and *M. intracellulare* lung disease are different in prognosis and response to the multiagent chemotherapy.[20] It is important to identify which the patients have *M. avium* lung disease or *M. intracellulare* lung disease to predict the prognosis and the treatment response. Besides, it is difficult to distinguish MAC lung disease from other NTM lung diseases completely from their CT findings.[21] Therefore, when serological test for MAC...
CONCLUSION

In summary, patients with bilateral BE with nodules on CT are less likely to have the positive AFB culture from bronchial wash when sputum test was not diagnostic. A large, prospective study is needed to confirm our findings.

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Conflicts of interest
There are no conflicts of interest.

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