Respiratory Medicine 128 (2017) 1–6

Contents lists available at ScienceDirect

Respiratory Medicine

journal homepage: www.elsevier.com/locate/rmed

Anti-*Pseudomonas aeruginosa* IgG antibodies and chronic airway infection in bronchiectasis

Guillermo Suarez-Cuartin [a, b], Alex Smith [c], Hani Abo-Leyah [c], Ana Rodrigo-Troyano [a, b], Lidia Perea [b], Silvia Vidal [b], Vicente Plaza [a, b], Thomas C. Fardon [c], Oriol Sibila [a, b, 1], James D. Chalmers [c, *, 1]

[a] Servei de Pneumologia, Hospital de la Santa Creu i Sant Pau, Universitat Autònoma de Barcelona, Barcelona, Spain
[b] Institut d'Investigació Biomèdica Sant Pau (IIB Sant Pau), Barcelona, Spain
[c] Tayside Respiratory Research Group, University of Dundee, Dundee, UK

ARTICLE INFO

Article history:
Received 17 October 2016
Received in revised form
3 April 2017
Accepted 1 May 2017
Available online 2 May 2017

Keywords:
Bronchiectasis
Pseudomonas aeruginosa
Antibody
Chronic infection

ABSTRACT

Background: Identification of chronic *Pseudomonas aeruginosa* (PA) infection is important in the management of bronchiectasis, but requires repeated sputum sampling. We hypothesized that serum anti-PA IgG antibodies could diagnose chronic PA infection at a single visit.
Methods: Clinically stable bronchiectasis patients were studied prospectively. Chronic PA infection was defined as 2 or more positive sputum samples at least 3 months apart and/or failure to clear PA following eradication treatment. Baseline serum anti-PA IgG was determined by a validated ELISA kit.
Results: A total of 408 patients were included. Sixty of them (14.7%) had chronic PA infection and had higher anti-PA IgG levels (median 6.2 vs. 1.3 units, $p < 0.001$). Antibody levels showed direct significant correlations with exacerbation frequency, the bronchiectasis severity index and sputum inflammatory markers. Fifty-seven patients with chronic PA infection had a positive test, giving 95% sensitivity, 74.4% specificity and AUROC of 0.87. During follow-up, 38 patients had a new PA isolation. Eradication at 12 months was achieved in 89.5% of subjects with a negative antibody test and 15.8% of patients with a positive test.
Conclusions: Anti-PA IgG test is highly accurate to detect chronic PA infection in bronchiectasis patients. In addition, it may be a marker of disease severity and treatment response.

1. Introduction

Bronchiectasis is a chronic lung disease characterized by irreversible dilation of the bronchi, leading to failure of mucociliary clearance and neutrophilic inflammation [1]. This condition predisposes patients to chronic respiratory bacterial infection, which perpetuates airway inflammation [2].

Pseudomonas aeruginosa (PA) is one of the most common organisms isolated in bronchiectasis patients [3,4]. PA infection can be chronic or intermittent, depending on the presence of persistent isolation of this microorganism in respiratory samples or not [5]. Chronically infected patients with PA have worse quality of life, increased exacerbations and poorer prognosis [6]. Therefore, monitoring sputum microbiology to identify PA infection status in bronchiectasis is essential in order to select the best treatment option for each individual [7].

Studies to date have used different definitions of chronic PA infection, with a recent systematic review identifying that 2 positive sputum cultures at least 3 months apart in 1 year is the most widely used definition in bronchiectasis [6]. Methods used in CF are more rigorous, requiring samples every 3 months and at least 50% of them being positive for PA [8,9]. Standard of care for bronchiectasis across Europe currently does not incorporate regular sampling of patients, and in recent audits only 62% of patients in the UK and 27% of bronchiectasis patients from Italy had a sputum sample sent even once per year [10,11]. This presents challenges in clinical practice to identify patients with chronic PA, and in clinical trials to identify a target population with chronic PA.

Serum IgG PA antibodies have been proposed to diagnose

* Corresponding author. Scottish Centre for Respiratory Research, University of Dundee, Dundee, DD1 9SY, UK.
E-mail address: jchalmers@dundee.ac.uk (J.D. Chalmers).
[1] Denotes joint senior authors.

http://dx.doi.org/10.1016/j.rmed.2017.05.001
0954-6111/

Abbreviations list

PA	*Pseudomonas aeruginosa*
CF	Cystic fibrosis
SGRQ	St. George's Respiratory Questionnaire
BSI	Bronchiectasis severity index
MPO	Myeloperoxidase

chronic bronchial infection, being a solution that can potentially identify this status at a single time point without longitudinal sputum sampling. In CF, different serological tests are available and most of these have shown a high sensitivity and specificity to detect chronic PA bronchial infection [12]. However, their routine use remains controversial [13]. Whilst PA infection is considered almost inevitable in CF with a prevalence of over 90% in adults [14], the rate of PA colonisation in bronchiectasis patients is around 21% [6]. Thus, identification of PA infection status in this latter group can be more challenging.

We hypothesized that specific anti-PA IgG antibody determination may be useful for identifying chronic PA infection in bronchiectasis patients.

2. Methods

2.1. Study design and ethics

This is a prospective study that included 408 clinically stable bronchiectasis patients. The study protocol was approved by the East of Scotland Research Ethics Committee (12/ES/0059) and all participants gave written informed consent to participate.

2.2. Participants

Patients were consecutively recruited from a specialist clinic at Ninewells Hospital in Dundee, United Kingdom 2012–2015 and were followed-up for 12 months. The diagnosis of bronchiectasis was confirmed in all cases with compatible clinical history of cough with sputum production and/or recurrent respiratory infections and presence of bronchial dilatation on high-resolution chest computed tomography scan. Patients with age less than 18 years; unable to give informed consent; patients with CF; active allergic bronchopulmonary aspergillosis (ABPA); active non-tuberculous mycobacterial disease; a primary diagnosis of pulmonary fibrosis with traction bronchiectasis and patients with immunodeficiency or receiving immunoglobulin replacement therapy were excluded.

2.3. Clinical assessment

All patients were clinically stable as defined by the absence of an exacerbation that required antibiotic or steroid treatment within 4 weeks prior to inclusion. Quality of life was assessed using the St. George's Respiratory Questionnaire (SGRQ) as this study was initiated prior to the availability of the disease specific Quality of Life Bronchiectasis Questionnaire (QOL-B). The aetiology of bronchiectasis was assessed as recommended by the British Thoracic Society guidelines [1]. Bronchiectasis severity index (BSI) and FACED score were determined as previously described [15,16].

2.4. Bacteriology

Spontaneous sputum samples were obtained for bacteriology and inflammatory marker measurement. Qualitative and quantitative bacteriology determination was performed in all samples as described previously [7]. Quality of sputum was evaluated using the Murray-Washington criteria [17].

Patients were classified into two groups according to previous chronic isolation of PA in sputum. Chronic PA infection was defined as 2 or more positive sputum samples at least 3 months apart and/or failure to clear PA following eradication treatment [18]. Standard of care at the study centre is to send sputum at all clinical encounters with a target for a minimum of 3 sputum cultures per year in expectorating patients. In a sensitivity analysis we evaluated a definition described by Lee et al. [19], referred to as the Leeds criteria. This required patients to have at least 3 sputum samples in the previous 12 months and at least 50% of samples to be positive for PA.

2.5. Specific anti-PA IgG measurement

Blood samples were obtained from all patients and processed for later antibody analysis by a validated commercially available ELISA kit (Pseudomonas-CF-IgG ELISA Kit. Statens Serum Institut, Denmark) following the manufacturer instructions [12,20,21]. The cut-off value for a positive ELISA Unit/10 result was 2.96 as determined by the manufacturer.

2.6. Airway biomarkers

Sputum samples were centrifuged at 50,000*g* for 90 min to obtain the soluble fraction. Neutrophil elastase activity and myeloperoxidase (MPO) activity in sputum supernatants were measured by chromogenic assay as previously described [7].

2.7. Statistical analysis

Results are presented as mean and standard deviation (SD) for continuous parametric data, and median and interquartile range for continuous non-parametric data. Categorical data is presented as frequencies and percentages. Continuous variables were analysed using t and ANOVA tests, whereas categorical variables were analysed using X^2 tests. Biomarkers and Anti-PA IgG levels were correlated by linear regression. A p value of less than 0.05 was considered significant. Statistical analysis was performed using the SPSS 22 software for Windows (SPSS, Chicago, Illinois, USA) and GraphPad Prism Version 6 (GraphPad Software Inc., San Diego, California, USA).

3. Results

3.1. Patient description

Four hundred and eight patients with clinically stable bronchiectasis were included. Of them, 247 (60.5%) were female, and mean age was 65.4 ± 12.7 years. The most frequent bronchiectasis aetiologies were idiopathic (43.9%) and post-infective (19.6%). Mean FEV_1 was 70.7 ± 24.4% of predicted value, and mean BSI score was 7.6 ± 4.7 points.

Sixty (14.7%) patients met the criteria for chronic PA infection at baseline. Table 1 shows the characteristics of the subjects, grouped by whether they met the criteria for chronic PA airway infection or not. Patients with chronic PA infection had significant more severe bronchiectasis (BSI score median 14.5 vs. 6 points, $p < 0.001$; and FACED score median 5 vs. 1 point, $p < 0.001$), more prior exacerbations (median 4 vs. 1, $p < 0.001$), and worse MRC dyspnoea score (median 3 vs. 2 points, $p < 0.001$). Patients with chronic PA infection had lower FEV_1% of predicted (median 72.5 vs. 55.3, $p < 0.001$)