

High Seroprevalence of *Bordetella pertussis* in Patients with Chronic Obstructive Pulmonary Disease: A Case-Control Study

Seyyed Hamid Hashemi ^{1,2}, Ebrahim Nadi ³,
Mehrdad Hajilooi ⁴, Mohammad-Ali Seif-
Rabiei ¹, Atefeh Samaei ¹

¹ Brucellosis Research Center, Hamedan University of Medical Sciences, Hamedan, Iran, ² Department of Infectious Diseases, Hamedan University of Medical Sciences, Hamedan, Iran, ³ Department of Internal Medicine, Division of Pulmonary Diseases, Hamedan University of Medical Sciences, Hamedan, Iran, ⁴ Department of Immunology, Hamedan University of Medical Sciences, Hamedan, Iran

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Correspondence to: Hashemi SH

Address: Division of Infectious Diseases, Sina Hospital, Mirzadeh-Eshghi Street, Hamedan 65168, Iran.

Email address: shahashemi@yahoo.com

Background: *Bordetella pertussis* has been suggested to take part in the acute exacerbation of chronic obstructive pulmonary disease (COPD). The aim of this study was to investigate the association between *B. pertussis* and COPD.

Materials and Methods: In this case-control study, 90 patients with COPD and 90 age- and sex- matched control subjects were included. Serum samples were tested for anti-*B. pertussis* IgG and IgA by ELISA. A physician completed a questionnaire including demographic characteristics, habitual history and spirometric findings for each patient.

Results: Of 90 patients with chronic obstructive pulmonary disease, 66 (51%) had mild, 31 (34.4%) had moderate, and 13 (14.4%) had severe disease. There was no significant association between *B. pertussis* IgA seropositivity and COPD. Serum levels of anti- *B. pertussis* IgG were significantly higher in patients with COPD than in the control subjects ($P < 0.001$). No association was observed between *B. pertussis* infection and severity of COPD.

Conclusion: The results suggest that there is an association between *B. pertussis* infection and COPD. Further studies should be planned to investigate the potential pathogenic mechanisms underlying these associations.

Key words: *Bordetella pertussis*; Chronic Obstructive Pulmonary Disease; Whooping Cough

INTRODUCTION

Pertussis or whooping cough is caused by *Bordetella pertussis*. Although its incidence has decreased following worldwide immunization, recent trends show the increasing incidence of pertussis among teenagers and adults (1). Some studies indicate pertussis to be the cause in 20%-30% of patients suffering from chronic cough of more than 2 weeks duration (1, 2).

The clinical features of the disease are different based on the age of onset: classic pertussis is more commonly found in children less than 12 years but adults are more likely to develop the atypical form with mild chronic

coughing episodes sometimes being the only manifestation of the disease (2).

There is a limited body of knowledge on the role of pertussis in development of COPD. Recent studies indicate that beside the known microorganisms causing acute exacerbation of COPD, other pathogens like *Mycoplasma pneumoniae*, *Chlamydia pneumoniae*, and *B. pertussis* are involved in COPD exacerbation (3,4). Although these studies indicate *B. pertussis* as one of the organisms responsible for acute exacerbation of COPD, none of them point to *B. pertussis* as a probable cause of COPD development.

Some other infectious diseases have been indicted as potential risk factors for COPD in adults (5). *Helicobacter pylori* and some latent viral infections such as adenovirus and Epstein-Barr virus have also been indicated as potential predisposing risk factors for the development and progression of COPD in adults (6-8). Based on these findings it seems appropriate to study the relationship between pertussis and COPD.

The aim of this study was to compare the seroprevalence of *B. pertussis* between COPD patients and control subjects, and to evaluate the association between *B. pertussis* seropositivity and severity of COPD.

MATERIALS AND METHODS

Study design, patients and controls

In this case-control study, 90 consecutive patients with COPD referred to the outpatient clinic of respiratory diseases in Shahid Beheshti Hospital were included. Shahid Beheshti Hospital is a referral center for pulmonary diseases located in Hamedan city in the west of Iran.

Diagnosis of COPD was made based on spirometry findings in all patients with a clinical suspicion of COPD. The exclusion criteria for patients with COPD were: a) exacerbation of COPD in the past month, b) history of antibiotic use in the past month and c) full reversibility of spirometric findings. Full reversibility was defined as a 12% increase in FEV₁ after using two puffs of short-acting bronchodilators. The severity of COPD was classified by spirometric data according to the guidelines of the Global Initiative for Chronic Obstructive Lung Disease (GOLD) (9). The control group consisted of 90 age- and sex-matched patients with pulmonary diseases other than COPD who referred to the hospital during the period of the study. Those with known history of COPD or antibiotic use in the past month were excluded.

Ethical considerations

This study was approved by the Ethical Committee of Hamedan University of Medical Sciences, Hamedan, Iran and written informed consents were obtained of all participants.

Data Collection and measurements

After obtaining informed consent, a questionnaire including demographic characteristics, history of cigarette smoking, and pulmonary function test results was filled out for each subject, and a serum sample was obtained and stored at -20°C until use.

Antibody titers against pertussis toxin (anti-PT) were measured by ELISA using specific kits for anti-PT IgG and IgA (IBL, Homburg, Germany). Anti-PT IgG titers more than 24 U/mL and anti-PT IgA titers more than 12 U/mL were considered to be positive.

Statistical analysis

Data were analysed using SPSS statistical package version 15, and $P < 0.05$ was considered statistically significant. T-test was used for analysis of quantitative variables and chi-square test for qualitative variables.

RESULTS

Ninety patients with COPD (65 males and 25 females, mean age of 67.7 ± 1.2 years) and 90 age- and sex- matched control subjects (53 males and 37 females, mean age of 61.6 ± 1.2 years) were included in the study. According to the GOLD classification, 66 (51%) patients with COPD had mild, 31 (34.4%) had moderate, and 13 (14.4%) had severe disease. No patient was found to have very severe COPD.

Table 1 shows the seroprevalence of *B. pertussis* infection in patients with COPD and control subjects. There was no significant difference in anti-PT IgA seropositivity between the study and control groups ($P=0.448$). The frequency of anti-PT IgG seropositivity was significantly higher in patients with COPD than in controls ($P<0.001$).

Table 1. Seroprevalence of *B. pertussis* infection in COPD patients and controls

	COPD patients (N=90)	Controls (N=90)	P-value
Anti-PT IgA			
Positive: n (%)	56 (62.2)	51 (56.7)	$P = 0.448$
Anti-PT IgG positive: n (%)	83 (92.2)	46 (51.1)	$P < 0.001$

COPD: Chronic obstructive pulmonary disease, PT: Pertussis toxin

The frequency of anti-PT IgA and anti-PT IgG seropositivity in different stages of COPD is shown in Table 2. No significant association was found between the severity of COPD and the frequency of anti-PT IgA and anti-PT IgG seropositivity.

Table 2. Seroprevalence of *B. pertussis* infection according to the severity of COPD

Severity	Anti-PT IgA positive n (%)	Anti-PT IgG positive n (%)
Mild	40 (60.9)	33 (50)
Moderate	21 (67.7)	17 (54.8)
Severe	8 (62)	6 (46.2)

COPD: Chronic obstructive pulmonary disease, PT: Pertussis toxin

DISCUSSION

In this study, we did not find any significant relationship between *B. pertussis* IgA seropositivity and COPD, but there was a statistically significant association between *B. pertussis* IgG seropositivity and COPD.

In general, specific IgA and IgG levels increase during acute pertussis. IgG increases and decreases gradually but IgA has a short half-life (5-6 days) and rapidly disappears (10). The median time for disappearance of antibodies is approximately six months for anti-PT IgG and three months for anti-PT IgA (11). However, a proportion of adults have significant titers of anti-PT antibodies. These are thought to develop repeated subclinical infections of *B. pertussis* (12).

The presence of IgA indicates recent infection while the presence of IgG alone indicates a history of infection (10). Based on these assumptions, our findings indicate no difference between patients with COPD and control group regarding active pertussis. We also did not find any significant correlation between the seropositivity of specific antibodies and the severity of COPD. These results indicate that pertussis infection does not influence the severity of COPD. However, the higher level of IgG as an indicator of previous pertussis and/or repeated subclinical infections in COPD patients, point to the pertussis infection as a potential risk factor for developing COPD later in life.

It is well known that inflammation of the respiratory tract plays an important role in pathogenesis of COPD (13). Smoking is known as the most important cause of COPD but considering the fact that only a small portion of smokers develop COPD and it can be found in nonsmokers as well, other probable causes of COPD like the influence of recurrent or chronic infectious respiratory diseases have also been investigated (14). According to animal models, which have been used to investigate the pathogenesis of COPD, the role of microbial agents alone or in combination with smoking in causing a chronic inflammatory response has been proposed. Lipopolysaccharide (LPS), a component of the cell wall of Gram-negative bacteria, is a strong proinflammatory compound (15). In animal models, chronic LPS inhalation causes pathological changes similar to human emphysema (16).

It is well known that LPS induces tumor necrosis factor (TNF)-alpha. TNF-alpha overexpression in mice has been reported to cause some diverse effects, including pulmonary emphysema and pulmonary fibrosis (17,18). Other researchers have reported that TNF-alpha plays a critical role in smoking-related emphysema. The high levels of TNF-alpha are found in the serum of smokers (19).

Repeated releases of TNF-alpha may be related to the high concentrations of bacterial endotoxin in tobacco and bioactive LPS in cigarette smoke (20,21). Considering these findings, TNF-alpha is believed to play a major role in development of COPD.

Although the role of *B. pertussis* in exacerbation of acute COPD has been demonstrated, there is no investigation on its probable effect on COPD pathogenesis. Considering the unique microbiological and clinical manifestations of pertussis infection, it seems likely to play a role in pathogenesis of COPD and our findings reinforce this assumption.

Pertussis may have a mild but chronic presentation and may not produce typical symptoms of severe cough. High incidence of seropositivity to *B. pertussis* has been reported in patients with chronic laryngotracheitis (22). The effect of

pertussis infection on respiratory mucosa might be related to its chronic nature (23). The conventional understanding indicates *B. pertussis* to be an extra cellular pathogen colonizing the respiratory epithelium but recent studies show that the bacteria like *chlamydia* and other intracellular microorganisms are capable of proliferation within the cells and can cause chronic infections (24). Different mechanisms have been found to facilitate intracellular survival of *B. pertussis* like adhesion, secretion of different toxins effective on various cells and antigenic changes.

Although *B. pertussis* is rarely present in the saliva three weeks after the onset of coughing, its presence inside the alveolar macrophages might play a role in its pathogenesis and prolong the course of disease. *B. pertussis* secretes different virulence factors like filamentous hemagglutinin, pertussis toxin, adenylate cyclase and tracheal cytotoxin. These toxins cause the inhibition of macrophage function and survival of the bacteria (25,26). These characteristics of pertussis infection and its higher prevalence among adults in the recent years indicate a possibility for its involvement in the occurrence of COPD, which necessitates further research to evaluate its role in chronic respiratory diseases.

In conclusion, our findings indicate a correlation between pertussis and COPD. Considering the fact that our results are based on a serological case-control study, further investigations are necessary to calculate the relative risk of COPD in patients with a history of pertussis.

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