ORIGINAL ARTICLE

SPECIFIC SERUM IMMUNOGLOBULIN G TO CHLAMYDIA PNEUMONIAE IN HEALTHY CHILDREN AND ADULTS (SOUTH-EAST OF IRAN)


Department of Immunology, School of Medicine, Kerman University of Medical Sciences, Kerman, *Department of Internal Medicine, Ali-ebn-Abilaleb Hospital, Rafsanjan University of Medical Sciences, Rafsanjan, **Department of Microbiology and Immunology, School of Medicine, Rafsanjan University of Medical Sciences, Rafsanjan-Iran

Background: Chlamydia pneumoniae (C. pneumonia) is an obligate intracellular bacterium and recognized as a risk factor for several diseases such as asthma, atherosclerosis and arthritis. The aim of this study was to determine the sero-prevalence of C. pneumonia in healthy subjects in different age groups. Methods: The serum levels of anti C. pneumonia IgG were measured by using of ELISA. Results: Totally, 630 subjects (164 children and 466 adults) were included into study. The sero-prevalence and the mean titer of anti C. pneumonia antibody were 11.3% and 14.48±2.18 RU/mL; at age ≤10 years, 15% and 17.47±2.40 RU/mL at age 11–20 years, 21% and 25.15±4.56 RU/mL at age 21–30 years group, 40% and 53.77±6.40 RU/mL at age 31–40 years, 94% and 146.41±8.95 RU/mL at age 41–50 years, 98% and 153.59±10.38 RU/mL at age 51–60 years, 96% and 138.80±12.78 RU/mL at age 61–70 years, respectively. The differences of the sero-prevalence and the mean titer of anti C. pneumonia antibody between age groups were significant (p<0.0001). The sero-prevalence and the mean titer of anti C. pneumonia antibody were 11.6% and 14.33±1.49 RU/mL in children and 65.5% and 97.40±4.46 RU/mL in adults. The sero-prevalence and the mean titer of anti C. pneumonia antibody were significantly higher in adults in comparison with those in children (p<0.0001). Conclusion: These findings showed that the sero-prevalence and titer of anti C. pneumonia IgG were increased with advanced ages and were higher in adults as compared to children.

Keywords: Chlamydia pneumonia; IgG; Sero-prevalence; Age; Adults; Children; Iran

INTRODUCTION

Chlamydia pneumoniae (C. pneumonia) is an intracellular pathogen that causes acute respiratory diseases. Almost all humans can expect to be infected with C. pneumoniae at least once throughout their life. Reinfections are also common and infections may become chronic. Following the primary infection, it can persist in the host, and acts as a potential risk factor for chronic respiratory disease or atherosclerosis. Moreover, epidemiologic studies have clearly indicated an important etiologic role of C. pneumoniae for several diseases including lung cancer, chronic obstructive pulmonary disease, asthma, migraine, Alzheimer’s disease and osteoporosis. Furthermore, it has been reported that C. pneumoniae easily spreads from the respiratory organs via the circulatory system and goes to tissues including brain, heart, vasculature, and joints. In our previous study, a higher sero-prevalence of anti C. pneumoniae and anti-Chlamydial heat shock protein-60 IgG was observed in patients with ischemic heart disease as compared with healthy subjects.

Respiratory infection with C. pneumoniae occurs throughout the world and in all age groups. It has also been reported that asymptomatic infections are frequent and the bacteria associated with 6–22% of lower respiratory tract infections in children and adults that change with geographic conditions, age and diagnostic methods in the investigated populations.

The epidemiological parameters influence the prevalence of C. pneumoniae IgG and IgA antibodies. However, serological testing remains the most frequently utilized method for diagnose C. pneumoniae infection.

There is no study regarding the sero-prevalence of the C. pneumoniae across age groups in the Iranian population. This study conducted for the first time to evaluate the serologic immunoglobulin G (IgG) response to C. pneumoniae, across age groups and also in children and adults in an Iranian population.

MATERIAL AND METHODS

From August to December 2013, a cross-sectional sero-prevalence study was carried out among healthy subjects in the Rafsanjan (a city located in Kerman province, in South-East of Iran). A total of 630
subjects (485 men and 145 women), including 164 children (aged 5–17 years) and 466 adults (aged 20–60 years) were enrolled in the study. The subjects were interviewed with regard to symptoms of upper or lower respiratory tract infection and none of them had any history of respiratory or any other relevant diseases. All subjects were basically healthy, with no acute or chronic illnesses. Indeed each individual with disease (such as history of recurrent infections, asthma, allergy and atopic diseases, any suspected immunological disorders, cigarette smoking and use of any drugs) were all excluded from the study. The other exclusion criteria were malignancy, surgery and using antibiotics within 4 weeks prior to blood collection.

Adults were recruited among blood donors of Rafsanjan Blood Transfusion Centre. They were randomly selected according to registration number. Children were recruited from randomly selected health centres. Informed consents were obtained from parents of all the children before blood samplings. Moreover, this study was evaluated and approved by the Ethical Committee of Rafsanjan University of Medical Sciences.

Two to three mL of peripheral blood was collected from each participant at the time of interview. The blood samples were centrifuged and the sera were separated and frozen at –20°C until analysis.

Serum levels of anti C. pneumoniae immunoglobulin G were measured by using the commercial enzyme-linked immune-sorbet assay (Euroimmun, Germany). Briefly, the control samples and serum samples were pipetted into the microtitre wells coated with C. pneumoniae specific immune-dominant antigens and then the plates were incubated. In case of positive samples, C. pneumoniae specific IgG antibodies would bind to the antigens. The wells were washed with washing solution to remove non-band antibodies, enzyme-labelled anti-human IgG was pipetted into each well and the plates were incubated again. In this stage the conjugate binds to the antibody-antigen complexes. Thereafter, the wells were washed with washing solution to remove excess conjugate. A fresh substrate solution, tetramethylbenzidine, was added, and the plates were incubated. If specific antibody to the antigen is present in the patient's serum, a blue colour develops which is proportional to the concentration of anti C. pneumoniae antibodies in the serum. The enzyme reaction was stopped with H2SO4 solution. The absorbance of each well was determined at 450 nm with a spectrophotometer ELISA microplate reader. According to manufacturer guidelines the results were expressed as relative units (RU/mL) and the values of >20 RU/ml were considered as positive.

Differences in variables were analysed using Kruskal-Wallis, Mann-Whitney U-test, Chi-square and Fisher exact tests as appropriate and P-values of less than 0.05 were considered significant. All the available data were analysed by SPSS.

RESULTS

The sero-prevalence of C. pneumoniae were 11.3% at age ≤10 years, 15% at age 11–20 years, 21% at age 21–30 years group, 40% at age 31–40 years, 94% at age 41–50 years, 98% at age 51–60 years and 96% at age 61–70 years, respectively (Figure-1). The statistical analysis showed that differences of the sero-prevalence C. pneumonia between age groups are significant (p<0.0001). The sero-prevalence of C. pneumonia was 11.6% in children and 65.5% in adults. The statistical analysis also showed that difference of the sero-prevalence of C. pneumonia was significantly higher in adults in comparison with those in children (p<0.0001). Totally, 51.4% of participants were found to be seropositive for anti C. pneumonia antibody.

In children, the sero-prevalence of C. pneumonia was 12.3% in male and 9.5% in female. In adults, the sero-prevalence of C. pneumonia was 66.4% in male and 62.1% in female. No significant difference was observed between male and female regarding the sero-prevalence of between male and female in both children and adults subjects. Moreover, No significant difference were observed between male and female regarding the sero-prevalence of C. pneumonia between male and female in all age groups.

The mean titre of anti C. pneumonia antibody were 14.48±2.18 RU/mL at age ≤10 years, 17.47±2.40 RU/mL at age 11–20 years, 25.15±4.56 RU/mL at age 21–30 years group, 53.77±6.40 RU/mL at age 31–40 years, 146.41±8.95 RU/mL at age 41–50 years, 153.59±10.38 RU/mL at age 51–60 years, 138.80±12.78 RU/mL at age 61–70 years, respectively (Figure-2). The statistical analysis showed that differences of the mean titre of anti C. pneumonia antibody between age groups are significant (p<0.0001). The mean titre of anti C. pneumonia antibody was 14.33±1.49 RU/mL in children and 97.40±4.46 RU/mL in adults. The statistical analysis showed that differences of the mean titre of anti C. pneumonia antibody were significantly higher in adults in comparison with those in children (p<0.0001).

In children, the mean titre of anti C. pneumonia antibody was 14.84±1.77 RU/mL in male and 12.84±2.78 RU/mL in female. In adults, the mean titer of anti C. pneumonia antibody were
98.61±5.07 RU/mL in male and 93.14±9.39 RU/mL in female. No significant difference were observed between male and female regarding the mean titre of anti C. pneumonia antibody between male and female in both children and adults subjects. Moreover, No significant difference were observed between male and female regarding the mean titre of anti C. pneumonia antibody in all age groups.

Figure 1: Sero-prevalence of C. Pneumonia in healthy subjects in different age groups

Figure 2: The mean titer of anti C. pneumonia antibody in healthy subjects in different age groups

DISCUSSION

The enzyme immunoassay has been introduced one of the most reliable test for diagnosis of C. pneumonia infection. The results of the present study showed that the overall prevalence of C. pneumonia was 51.4%. The sero-prevalence of infection was relatively low in children and increased with advanced ages to reach a plateau of at around 41–50 years of age, which remained approximately stable in 41–70 years of age. The seropositivity was higher in adults than in children and teenagers. The seropositivity in males was also slightly higher than females. Sero-epidemiologic studies have indicated C. pneumoniae infection is worldwide and that sero-prevalence in different parts of the world was estimated to be 40–60% in adults and 10–20% in children. The sero-prevalence of 65.5% in adults of this study was one of the highest reported worldwide. Accordingly, this high sero-prevalence of C. pneumoniae in the asymptomatic subjects represents that infection with this bacteria is common in Iran.

The IgG antibody prevalence of C. pneumoniae in healthy adult populations was shown to be 43.4% in Japan, 12 40–52% in US13 66.7% in Jordan, 11 87.5% in China,15 65.5–70.0% in Singapore,16 72.3% in Taiwan,17 54–72% in Finland18 and 61.6% in South-Korea. In a study from Turkey the frequency of C. pneumonia was 3.5% in children between the ages of 5 and 15 years. The accounts for these geographical differences in the prevalence of C. pneumoniae have not been clearly understood. However, this discrepancy may be largely attributed to differences including race and ethnic background of participants, population density, socioeconomic status and hygienic conditions.

The results of the present study also showed that the antibody prevalence increased rapidly with age. The advanced increasing in antibody prevalence with age is similar to that described by others. The most important prominent increase was observed in the 31–40 and 41–50 years. Thus, it is likely that most individuals acquired infection during the third and fourth decades of life. In the present study, the prevalence of C. pneumoniae infection reached to 98.0% at 51–60 years of age, suggesting the highest antibody prevalence reached maximum at sixth decade of life and remained high thereafter. These results represent that the primary infection is acquired during the first five decades of life, and in older ages high antibody levels are likely maintained by reinfection or persistent infection. In consideration of the short duration of C. pneumoniae-specific antibody, the higher sero-prevalence in the older ages indicating that the persistent infection or reinfection with C. pneumoniae during adulthood is frequent. The overall gender difference was not significant in this study which is similar to other studies.

In conclusion, this sero-prevalence study showed that infection with C. pneumoniae was prevalent in an Iranian population. Both the sero-prevalence and mean titre of anti C. pneumonia IgG were increased with advanced ages and were higher in adults as compared to children.
REFERENCES


Address for Correspondence:
Abdollah Jafarzadeh, Department of Microbiology and Immunology, Medical School, Rafsanjan University of Medical Sciences, Rafsanjan-Iran
Tel: + 98 391 523 4003
Email: jafarzadeh14@yahoo.com