In vitro analysis of allergens among patients with Asthma - A clinical study

Girish Dandagi^{1,*}, Vilol Joshi², Siddappa Dhaduti³

¹Professor, ^{2,3}Resident, Dept. of Pulmonary Medicine, Belagavi Institute of Medical Science, Belgaum, Karnataka, India

*Corresponding Author:

Email: girishdandagi.2009@rediffmail.com

Abstract

Background: Allergy is one of most basic immunological response which happens after exposure to substances which are safe to dominant part of individuals. On the planet 20-30% of the tenants are known to experience the ill effects of different hypersensitive issue like bronchial asthma, atopic dermatitis, unfavorably susceptible rhinitis and urticaria. From the previous 10-15 years there is a sudden radical increment in the quantity of hypersensitivities in both developed and developing nations. Urbanization, changing ways of life and expanding contamination in Indian sub-mainland has expanded the predominance of asthma. Different sorts of allergens in patients with unfavorably susceptible turmoil is a fundamental factor as a result of the clinical highlights which relies upon the nature and qualities of allergens in charge of refinement. As and when patients are getting presented to these allergens there is high limit of exacerbating the asthmatic condition. Consequently we plan to recognize such different types of allergens which we saw in a group of patients giving history of asthma.

Materials and Methods: Eighty patients giving history of asthma or recently analyzed instances of asthma were incorporated into our study group. Age ranged between 17 to 40 years. All patients with asthma were analyzed utilizing GINA 2018 rules. Patients with other foundational conditions prompting indications like asthma or windedness, history of smoking, patients on medicines were avoided from our present examination.

Before the beginning of the study ethical clearance was obtained from Institutional review board. Every one of the patients was educated about the investigation and consent was obtained. The 5ml of patient's serum was sent for in vitro examination of allergen by Immuno-EIA technique. The aggregate IgE esteem for serum more than 100KU/L is viewed as positive for sensitivity later serum IgE levels for particular allergens determined.

Results: Total of 80 patient were included in study 50 males and 30 females. Age ranged between 17 to 40years with meanage of 28.5years. Highest number of allergens noted was Housedust mite (70%) with least being chalk powder, cement dust, soap (1%) respectively.

Conclusions: Immuno-EIA remains as a one of the most rapid, easily performed, most trusted invitro technique to screen the patients with different kinds of allergens. This can help us further to manage the patient accordingly.

Keywords: Allergen; Asthma; Immuno EIA.

Introduction

Among whole world total population around 20-30% of them are known to experience the ill effects of unfavorably susceptible allergens, for example, bronchial asthma, hypersensitive rhinnitis, atopic dermatitis, urticaria, etc[1]. Presently asthma being a one of the most widely recognized chronic disease grown-ups which being on rise[2,3]. The allergic Rhinitis and its effect on Asthma (ARIA) 2008 announced that allergic rhinitis as a major fundamental allergic sickness alongside asthma which causes significant ailment and disability worldwide[4]. Another couple of investigations which affirmed that asthma and related allergic disorders are exceptionally common in westernized or urbanized social disorders than in rural developing countries[5-7]. A multinational study which was led in 2012, found that larger part of nations did not have precise information identified with allergy[8, 9].

So many different investigations already have additionally said about that the predominance of asthma and allergic susceptible conditions have persistently increased in different regions of the world[5,10,11]. Overall, there is sudden increase in the number of allergies ubiquitously including developing nations that were once immune from these allergens are likewise experiencing this condition.

In the course of recent couple of decades circumstance in India has changed radically. As indicated by an investigation report the predominance of asthma is around 14% individuals[12]. Another investigation recommends that over 25% of the populace experiencing major allergic problems out of which respiratory allergy constitute 73.4%[13,14]. Various different examinations from India have revealed hypersensitivity pervasiveness going from 3.5% up to 29.5%[2,3,15-18].

As allergy is a most common amongst the hypersensitivity disorders causing asthma in human system. These responses happen when a man's invulnerable framework responds unusually to regularly safe substances, present in nature which is called an allergen [14]. Among at least four types of hypersensitivity it is one of them. Which is likewise called as type I (or immediate) hypersensitivity response. The uprising burden of allergic diseases in India has expanded the prevalence and in addition seriousness of the condition [19, 20]. A multi-centric population based investigation, by Indian study on epidemiology of asthma, Respiratory symptoms and

chronic bronchitis (INSEARCH) 21 has been directed, which secured 12 centers including both country and urban regions spread over various parts of India. Collectively for all the centers which was observed to be 2.05% (territory, 0.4% - 4.8%)[21].

A proper test to comprehend the allergen and its avoidance assume an imperative part in controlling asthma. Expanded presentation to allergen in defenseless people, can lead to allergic sensitization. Sensitization play an essential role in development in severity in development of asthma. In a patient with asthma, Identification of allergens and subsequent severity can improve symptoms. In many cases, a patient will have different allergies and the avoidance plan should focus on every positive allergen[22].

There are not many literatures which notices in regards to in vitro examination of allergens among patients with asthma. Thus, we go for this in vitro concentrate to break down various kinds of allergens in asthmatic patients.

Materials and Methods

Eighty patients giving history of asthma or recently analyzed instances of asthma were incorporated into our investigation. Age group ranges between 17 to 40 years. All patients with asthma were analyzed utilizing GINA 2018 guidelines[23]. Patients with other similar conditions prompting indications like asthma or shortness of breath, history of smoking, patients on medicines were prohibited from our present study.

Before the beginning of the investigation ethical clearance was obtained from Institutional review board. Every patient was educated about the study and consent was acquired. The 5ml of patient's serum was sent for in vitro investigation of allergen by Immuno-EIA technique which was performed with the assistance of Doekes G. standard rules with modification[24]. This machine assesses around 200 allergens which are faced in the day to day life.

Principle of the Procedure

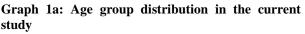
The EIA Allergen-Particular IgE assay utilizes a little plastic device known as a test chamber to expose the patient serum at the same time to various allergens or allergen blends. The Test chamber contains discrete portions of cellulose string, each with an allergen or allergen blend covalently bound to it. Each test chamber likewise contains one negative blanking control and one positive procedural control.

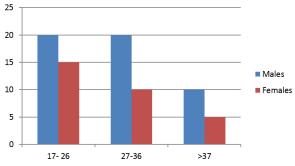
The EIA Allergen-Particular IgE assay is controlled by filling a test chamber with patient serum. IgE in the serum ties to the allergen-covered cellulose strings during incubation. The test chamber is then washed with buffer to expel unbound serum components. Enzyme named against IgE is then added to the chamber and couples with the serum IgE bound to the strings. Following a second wash, the test chamber is loaded up with a photo reagent blend that responds with the photo reagent mixture that reacts with antibody which was added to create chemiluminescence. The measure of light discharged by each string is specifically relative to the measure of allergen-particular IgE in the patient serum.

The aggregate IgE estimated for serum assessed more than 100KU/L is viewed as positive for sensitivity later serum IgE levels for particular allergens were resolved. The qualities more than 0.35U/L were thought to be sure for those specific allergens.

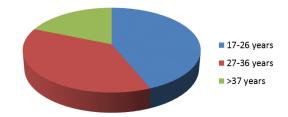
Results

On the basis of in vitro tests, total of 80 patients were included in the study. There were 50 males and 30 females. Age ranged between 17 to 40years with mean age of 28.5 years.





Graph 1b: Age group distribution in the current study

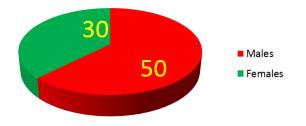


Graph 1a and 1b shows, the age group ranged from 17-40 years, which were subdivided into 3 groups

- 1. 17---26
- 2. 27---36
- 3. >37 years

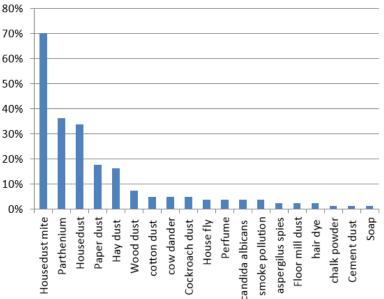
The age group between 17- 26 included 20 (40%) male and 15 (50%) female. The age group between 27-36 included 20 (40%) male and 10 (33.33%) female. The age group >37 included 10 (20%) male and 5 (16.66%) female. The highest numbers of males (40%) and female (50%) are group of 17–26 years.

Graph 2: Gender distribution in the current study



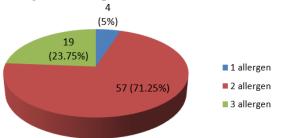
Graph 2 shows, among total 80 patients 50(62.5%) were males and 30 (37.5%) females

Graph 3: Different types of allergens noted in the current study



Graph 3 shows, highest number of allergens noted was house dust mite (70%) with least being Chalk powder, cement dust, soap (1%) respectively.

Graph 4: Distribution of number of allergens affecting individual patients



Graph 4 shows, 57 (71.25%) of them had allergy to two different allergens, 19 (23.75%) patients had allergy to three different allergens followed by 4 (5%) had allergy to one allergen.

Discussion

Total of 80 patients were included in the study. There were 50 males and 30 females. Age group ranged between 17 to 40years with mean age of 28.5years. (Graph 1a) Highest number of allergens noted was house dust mite (70%) with least being chalk powder, cement dust, soap (1%) respectively.

The age group ranged from 17-40 years, which were subdivided into 3 groups. (Graph 1b) 1)17-26 years, 2) 27-36 years 3) >37 years. The age between 17-26 years included 20(40%) male and 15(50%) years' female. The age between 27-36 years included 20(40%) male and 10(33.33%) female. The age group >37 years had included 10(20%) male and 5(16.66%) female. The highest numbers of males 40% and female 50% are group of 17 – 26 years. Among total 80 patients 50(62.5%) were males and 30 (37.5%) females. (Graph 2) Highest number of allergens noted was house dust mite (70%) with least being chalk powder, cement dust, soap (1%) respectively. (Graph 3)Among 80 patients 57(71.25%) of them had allergy to two different allergens, 19(23.75%) patients had allergy to three

different allergens followed by 4 (5%) had allergy to one allergen. (Graph 4)

According to World health organization report there are 300 million asthmatic patients and it is probably going to increase to 400 million by 2025. Patients with asthma and allergic diseases have a susceptible illnesses have a lessened personal life. According to the World Health Organization asthma causes 2.5lakhs deaths annually[25]. Hypersensitivity is accepted to be interceded by Immunoglobulin E (IgE), which is one of the significant mediators of quick hypersensitivity response. Allergic diseases caused by specific IgE immune response to surface receptors on cells like most pole cells present and eosinophils[26]. The allergen might be noticeable when an individual inhales, chemicals that one interacts with allergens[27]. 80% of asthmatics are sensitive to indoor allergens[28-30]. Dust mite are the most pervasive source overrunning most homes in Australia, New Zealand, UK and Western Europe, broad locales of Asia and South America and mild and subtropical areas of the US[31].

In our investigation house dust mites noted in 70% cases which was as per noted by Afaf et al.[32] and utilization of the IgE level is a promising demonstrative instrument in the analysis of house dust mite. While house dust mite and house dust was observed to be of lesser than the consequences different investigations[33,34]. Allergenicity to Parathenium extracts was recorded in 12% bronchial asthma patients from Bangalore [35] and in north India Punjab[36], the rate of parathenium as an allergen was lesser than our present study which could be because of variation in test sample. In Northern India (Punjab)[36] likewise demonstrated that a huge extent of bronchial asthma patients is sensitized to Parthenium as per our study.

Wood dust exposure is a typical presentation in the work environment. Numerous items we utilize are produced using wood and many laborers are presented to wood dust amid their fabricate; occupations with presentation to wood dust incorporate craftsmen, furniture creators, bureau producers, instrument creators, and saw process specialists. there have been various epidemiologic examinations that have demonstrated expanded respiratory adversely health effects in furniture producers and sawmill workers[37,38]. In a Danish investigation of furniture laborers, just seven of 131 (5.3%) workers with asthma had IgE to pine, the transcendent wood being utilized; showing that sensitization to wood dust is just a single component for asthma noted in the carpentry industries[39].

Among outdoor air allergens, it was observed to be less predominant, which was in accordance to past studies[40-43]. Moreover, prevalence of allergy to cockroach was resolved to be 5% which was lower than consequences of different investigations[33,44-47] while the sensitization to cockroach allergens is the most widely recognized indoor allergens in a few locales with comparable climates to our region[48-50].

Similar to our study cockroach allergens were found in numerous poor inner city regions have been related with asthma onset or worsening in numerous countries[51-53], 40– 60% of patients with asthma in urban and inner city zones has Immunoglobulin E (IgE) antibodies to cockroach allergens[54]. Two instances of atopic asthma caused by candida albicans announced that had large amounts of serum IgE antibodies. Whereas, in our present investigation we noted 4% candida albicans[55] allergen positive. Utilization of unclean cooking fuels (Smoke pollution) has been related with increased chances of asthma[56].

Asthma were related with hoisted levels of house dust from the kitchen smoke and living room floors where as in our study comparative outcomes acquired in our present investigation with 34% of asthmatics. Dust weight and Asthma Predominance in the National survey of Lead and Allergens in Housing[57]. In India, there is a high prevalence of Aspergillus sensitizations and ABPA complicating asthma[58,59]. Wheat grain prompting hypersensitivity in flour millworkers noted in four instances of bronchial asthma. Where as in our present examination we noted 3% flour mill allergen positive[60].

Very few literature which mentions regarding allergen paper dust (18%), cotton dust (5%), cow dander (5%), Housefly (4%), perfume (4%) hair dye (3%), cement, chalk powder, and soap (1%) separately. Subsequently every allergen must be noted and recorded and distributed area astute which can assist the practitioners with analyzing it appropriately by prompting the right testing strategy and which additionally encourages them giving a legitimate treatment mind.

Conclusions

In conclusion, various types of environmental allergens can induce asthma. Immuno-EIA remains as a one of the most rapid, easily performed, most trusted in vitro technique to screen the patients with these kinds of exposures. This can further be helpful for effective treatment of these patients by immunotherapy. Major steps need to be taken at both personal and government levels to curb the rising pattern of allergies.

Conflicts of interest: Nil

Acknowledgements

I would like thank the director BIMS, Belagavi and Endocrine lab Gujarat for helping to conduct this research work.

References

- 1. Schafer T, Ring J. Epidemiology of allergic diseases. *Allergy* 1997;52:1-14.
- 2. Pearce N, Aït-Khaled N, Beasley R, Mallol J. Worldwide trends in the prevalence of asthma symptoms: phase III of

the International Study of Asthma and Allergies in Childhood (ISAAC). *Thorax* 2007;62:758-66.

- Pal R, Dahal S, Pal S. Prevalence of bronchial asthma in Indian children. *Indian J Community Med* 2009;34 (4):310-316.
- 4. Bousquet J, Khaltaev N, Cruz AA. Allergic rhinitis and its impact on asthma (ARIA) *Allergy* 2008;63:2-160.
- Strachan D, Sibbald B, Weiland S, Aït-Khaled N, et al. Worldwide variations in prevalence of symptoms of allergic rhinoconjunctivitis in children: the International Study of Asthma and Allergies in Childhood (ISAAC) *Pediatr Allergy Immunol* 1997; 8: 161-76.
- Worldwide variation in prevalence of symptoms of asthma, allergic rhinoconjunctivitis, and atopic eczema: ISAAC. The International Study of Asthma and Allergies in Childhood (ISAAC) Steering Committee. *Lancet* 1998;351:1225-32.
- 7. Worldwide variations in the prevalence of asthma symptoms: the International Study of Asthma and Allergy. www.mathewsopenaccess.com 1998.
- Prescott SL, Pawankar R, Allen KJ, Campbell DE, et al. A global survey of changing patterns of food allergy burden in children. *World Allergy Organ J* 2013;6(1):21.
- 9. Holgolt ST. The epidemic of allergy and asthma. *Nature* 1999;402:B2-4.
- 10. Von Mutius E. The rising trends in asthma and allergic disease. *Clin Exp Allergy* 1998;28(5):45-9.
- Chinn S, Jarvis D, Burney P, Luczynska C, et al. Increase in diagnosed asthma but not in symptoms in the European Community Respiratory Health Survey. *Thorax* 2004;59(8):646-51.
- 12. Surge in allergic diseases in India: WAO. Zee News. 2011;16:20-7.
- Shaikh WA. Allergies in India: An analysis of 1,619 patients attending an allergy clinic in Bombay. *Int Rev Allergy Clin Immunol* 1997;3:101-10.
- Vishwanathan R. Definition, incidence, etiology and natural history of Asthma. *Ind J Chest Dis* 1964;6:108-24.
- 15. Paramesh H. Epidemiology of asthma in India. *Indian J Pediatr* 2002;69:309-12.
- Shah JR, Amdekar YK and Mathur RS. Nation wide variation in prevalence of bronchial asthma- (part of the international study of asthma and allergies in childhood-ISAAC). *Indian J Med Sci* 2000;54:213-20.
- Sharma S, Gupta RC, Dixit R, Sharma S, et al. Prevalence of asthma in school children with allergic condition in rural areas of Ajmer, India. *Chest* 2008;134:5298-301.
- Chhabra SK, Gupta CK, Chhabra P and Rajpal S. Risk factors for development of bronchial asthma in children in Delhi. *Ann Allergy Asthma Immunol* 1999;83(5):385-90.
- Anonymous. All India Coordinated Project on Aeroallergens and Human Health Report. Ministry of Environment and Forests, New Delhi; 2000.
- Chhabra SK, Gupta CK, Chhabra P, Rajpal S. Prevalence of bronchial asthma in schoolchildren in Delhi. J Asthma 1998;35:291-6.
- Jindal SK, Aggarwal AN, Gupta D, Agarwal R, Kumar R,Kaur T, *et al.* Indian study on epidemiology of asthma, respiratory symptoms and chronic bronchitis in adults (INSEARCH). *Int J Tuberc Lung Dis* 2012;16:1270-77.
- Baxi SN, Phipatanakul W. The role of allergen exposure and avoidance in asthma. *Adolesc Med State Art Rev* 2010;21:57–71,viii–ix.

- Global Initiative for Asthma. Global Strategy for Asthma Management and Prevention, 2018. Available from: www. ginasthma. org. Last accessed 2018 on June 12.
- Doekes G. Enzyme immunoassays for total and allergen specific IgE in population studies. *Occup Environ Med* 1996;53:63–70.
- Pawankar R, Canonica GW, ST Holgate ST, Lockey RF, Blaiss M: The WAO White Book on Allergy (Update. 2013).
- 26. Corry DB, Kheradmand F. Induction and regulation of the IgE responses. *Nature* 1999:102:B18-23.
- Boyce JA, Assa'ad A, Burks AW, Jones SM, et al. Guidelines for the diagnosis and management of food allergy in the United States: report of the NIAID-sponsored expert panel. *J Allergy Clin Immunol* 2010;126:S1-58.
- Simpson A, Soderstrom L, Ahlstedt S, Murray CS, Woodcock A, Custovic A. IgE antibody quantification and the probability of wheeze in preschool children. J Allergy Clin Immunol 2005;116:744-9.
- Holt PG, Rowe J, Kusel M, Parsons F, Hollams EM, Bosco A. Toward improved prediction of risk for atopy and asthma among preschoolers: a prospective cohort study. J Allergy Clin Immunol. 2010;125:653-9.
- Lötvall J, Akdis CA, Bacharier LB, Bjermer L, Casale TB, Custovic A, et al. Asthma endotypes: a new approach to classification of disease entities within the asthma syndrome. J Allergy Clin Immunol 2011;127:355-60.
- Liam CK, Loo KL, Wong CM, Lim KH, Lee TC: Skin prick test reactivity to common aeroallergens in asthmatic patients with and without rhinitis. *Respirology* 2002;7:345–50.
- 32. Afaf Abd El-Raouf Taha, Samia E. Etewa, Sara A. Abdel-Rahman, Asmaa M. Farouk Al Ghandour, Amal H. Atta, Asmaa M. Darwish. House dust mites among allergic patients at the Allergy and Immunology Unit, Zagazig University: an immunologic and serologic study. 2018;42(3):405–15.
- 33. Khazaei HA, Hashemi SR, Aghamohammadi A, Farhoudi F, Rezaei N: The study of type 1 allergy prevalence among people of South-East of iran by skin prick test using common allergens. *Iran J Allergy Asthma Immunol* 2003;2:165–68.
- 34. Thomas WR. Geography of house dust mite allergens. *Asian Pac J Allergy Immunol* 2010;28:211-24.
- 35. Rao M, Prakash O, Subba Rao PV. Reaginic allergy to parthenium pollen evaluation by skin test and RAST. *Clin Allergy* 1985;15:449-54.
- Suresh PV, Gupta D, Behera D, Jindal SK. Bronchial provocation with parthenium pollen extract in bronchial asthma. *Indian J Chest Dis Allied Sci* 1994;36:104.
- Wiggans RE, Evans G, et al. Asthma in furniture and wood pro cessing workers: a systematic review. Occupational Med 2016;66:193-201.
- Pérez-Rios M, Ruano-Ravina A, et al. A meta-analysis on wood dust exposure and risk of asthma. *Allergy* 2010;65:467-73.
- 39. Skovsted TA, Schlünssen V, et al. Only few workers exposed to wood dust are detected with specific IgE against pine wood. *Allergy* 2003;58:772-79.
- 40. Khazaei HA, Hashemi SR, Aghamohammadi A, Farhoudi F, Rezaei N: The study of type 1 allergy prevalence among people of South-East of iran by skin prick test using common allergens. *Iran J Allergy Asthma Immunol* 2003;2:165–68.
- 41. Ezeamuzie CI, Thomson MS, Al-Ali S, Dowaisan A, Khan M, Hijazi Z: Asthma in the desert: spectrum of the sensitizing aeroallergens. *Allergy* 2000;55:157–62.

- 42. Ezeamuzie CI, al-Mousawi M, Dashti H, al-Bashir A, al-Hage M, al-Ali S:Prevalence of allergic sensitization to inhalant allergens among blooddonors in Kuwait--a desert country. *Allergy* 1997;52:1194–200.
- 43. Arbes SJ Jr, Gergen PJ, Elliott L, Zeldin DC: Prevalences of positive skin test responses to 10 common allergens in the US population: results from the third National Health and Nutrition Examination Survey. J Allergy Clin Immunol 2005;116:377–83.
- Fereidouni M, Farid Hossini R, Assarehzadegan MA, Jabberi Azad F, Varasteh A. Skin prick test reactivity to common aeroallergens among allergic rhinitis patients in Mashhad, *Iran allergo et Immunopathol* 2009;37:73–9.
- Almogren A. Airway allergy and skin reactivity to aeroallergens in Riyadh. Saudi Med J 2009;30:392-96.
- Farhoudi A, Pourpak Z, Mesdaghi M, Kazemnejad A, Chavoshzadeh Z. The study of Cockroach allergy in Iranian Children with asthma. *Acta Medica Iranica* 2003;41:150–55.
- Ghaffari J, Khademloo M, Saffar MJ, Rafiei A, Masiha F. Hypersensitivity to house dust mite and cockroach is the most common allergy in north of Iran. *Iran J Immunol* 2010;7:234–39.
- Ezeamuzie CI, Thomson MS, Al-Ali S, Dowaisan A, Khan M, Hijazi Z. Asthma in the desert: spectrum of the sensitizing aeroallergens. *Allergy* 2000;55:157–62.
- 49. Sattar HA, Mobayed H, al-Mohammed AA, Ibrahim AS, Jufairi AA, Balamurugan P, Mary VP, Bener A. The pattern of indoor and outdoor respiratory allergens in asthmatic adult patients in a humid and desert newly developed country. *Eur Ann Allergy Clin Immunol* 2003;35:300–05.
- Bener A, Safa W, Abdulhalik S, Lestringant GG: An analysis of skin prick test reactions in asthmatics in a hot climate and desert environment. *Allerg Immunol* (Paris). 2002;34:281–86.
- 51. Davey G, et al. Brittonw Wheeze, allergic sensitization and geohelminth infection in Butajira, *Ethiopia Clin Exp Allergy* 2005;23:301-07.
- 52. Silva JM, et al. A prospective study of wheezing in young children: the independent effects of cockroach exposure,

breast-feeding and allergic sensitization. Pediatr. *Allergy Immunol* 2005;16:393-40.

- 53. Chen YC, Tsai CH, Lee YL. Early-life indoor environmental exposures increase the risk of childhood asthma *Int J Hyg Environ Health* 2011;212:19-25.
- 54. Sohn MH, Kim KE. The cockroach and allergic diseases. *Allergy Asthma Immunol Res* 2012;4:264-69.
- Akiyama K, Shida T, H. Yasueda, H. Mita, T. Yamamoto, H. Yamaguchi. Atopic asthma caused by Candida albicans acid protease: case reports. *Eur Academy Allergy Clin Immunol* 1994;49(9):778-81.
- 56. Kumar R, Kumari Dolly, Srivastava Prakriti, Khare V, Arora N, Gaur SN, *et al.* Identification of IgE mediated food allergy and allergens in older children and adults with asthma and allergic rhinitis. *Indian J Chest Dis Allied Sci* 2010;52:217-24.
- 57. Leslie E, Samuel J. Arbes, Jr., Eric S. Harvey, Robert C. Lee, Päivi M. Salo, Richard D. Cohn, Stephanie J. London, and Darryl C. Zeldin. Dust weight and asthma prevalence in the National survey of lead and allergens in housing. *Environ Health Perspect* 2007;115(2):215–20.
- Agarwal R, Aggarwal AN, Gupta D, Jindal SK. Aspergillus hypersensitivity and allergic bronchopulmonary aspergillosis in patients with bronchial asthma: systematic review and meta-analysis. *Int J Tuberc Lung Dis* 2009;13:936–44.
- Agarwal R, Khan A, Gupta D, Aggarwal AN, Saxena AK, et al. An alternate method of classifying allergic bronchopulmonary aspergillosis based on highattenuation mucus. *PLoS One* 2010;5:e15346.
- Millworker's Asthma: Allergic Responses to the Grain Weevil. J. A. LUNN. Br J Industr Med 1966;23:149.

How to cite this article: Dandagi G, Joshi V, Dhaduti S. In vitro analysis of allergens among patients with Asthma - A clinical study. Indian J Immunol Respir Med. 2018;3(4):177-182.