

ORIGINAL ARTICLE

ANIMAL ALLERGY AMONG HUSBANDRY PERSONNEL IN RIYADH

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Background: The importance of specific animal allergy in immuno-pathology of asthma and atopic diseases remained to be defined. **Methods:** We measured total and specific IgE. Western blotting of some allergens was also characterised. **Result:** There was a significant elevation of IgE in 3 persons among 15 samples collected. Western blotting showed common husbandry allergens from 3 sources that have common allergenicity. **Conclusion:** Exposure to animal products exacerbates allergic asthma in adults, suggesting that preventive measures should be taken to reduce their sensitivity.

Keywords: Animal Allergy, IgE, RAST, Asthma

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INTRODUCTION

Animal allergy (AA) is an important occupational disease among personal working with animals such as mice, rats, pigs, and rabbits. The common manifestations of animal allergy are rhinitis, conjunctivitis and asthma.¹⁻³ Symptom severity can vary from mild to severe upper respiratory tract symptoms. Any person exposed to the urine of these animals are at risk of developing animals allergy.

The incidence of occupational lung disease due to common occupational agents is being under studied by many Saudi scientists. The prevalence of animal allergy in KSA is between 15 and 45%, which is similar to USA (11-20%) and Japan (23%). The prevalence of specific IgE is about 15-30%. Symptoms typically appear between one month and several years after initial exposure. Studies showed these symptoms to include rhinitis, conjunctivitis, urticaria, and Asthma. In these studies, hair, fur dander, urine and serum have been the most frequently used allergens for the most commonly studied species.⁴⁻⁷

The debate between total and specific IgE:

Taking an allergic history is critical, not only in selecting the appropriate allergens for testing, but also for testing the allergy test results in order to diagnose food allergy, allergic asthma or allergic rhinitis. An IgE-related mechanism must be demonstrated since many exogenous substances may cause otherwise clinically indistinguishable syndromes (including bronchospasm and urticaria) by mechanisms not related to IgE sensitization of mast cells and basophils. The absence of an allergen specific IgE-induced response argues against an allergic mechanism as a cause of the symptoms. To establish the immunologic mechanism, it is necessary to demonstrate the presence of allergen-specific IgE

antibodies at a level sufficient to induce an immunologic response following appropriate antigen challenge *in vivo* or by measuring the quantity of allergen specific IgE *in vitro*.

Test results for the presence of allergen specific IgE, whether *in vitro* (RAST) or *in vivo* (skin test) may be considered clinically relevant only if there is a history compatible with symptoms induced by exposure to the allergen. Additionally, test results indicating merely the presence of allergen-specific IgE in the serum do not necessarily indicate that the patient has clinical disease related to exposure to this allergen. All test results must be interpreted in the context of the patient's allergy history, and laboratory tests should not be used as the sole criteria for establishment of the diagnosis of allergy.

Unfortunately, one single measurement or observation cannot determine clinical allergy. The specific IgE only constitute one part of the allergic cascade including inflammatory cells, mediator releasability and end organ sensitivity. This makes impossible the use of serum specific IgE as the sole criteria to determine atopy. Any utilization of a serum specific IgE level must be in the clinical context of the likelihood of the presence of an allergic disease.

MATERIAL AND METHODS

Blood was collected by venipuncture from 10 patients suspected to be with allergy. Blood was allowed to clot, serum collected after centrifuged at 1,500 rpm for 5 min. Serum was stored in closed ampules at -20 °C. All sera used were taken from individuals suspected of having allergic symptoms associated with direct or indirect exposure to AA. The skin prick test results and clinical records were not available for this study.

The study was conducted between June and September 2013 in Salman Bin Abdul Aziz

University Hospital, Al Kharj, KSA. All participants provided informed consent before enrolment in the study. The study protocol conformed to the ethical guidelines of the 1975 Declaration of Helsinki and was approved by the Ethics Committee of Salman Bin Abdul Aziz University Hospital. Determination of eligibility was based on medical history and physical examination. Additionally, 5 healthy controls ranging in age from 29 to 55 years (Mean age 40.56±9.13) were run in parallel.

Total IgE measurement provided a useful insight into an individuals allergy drive. All specific IgE testing was performed on Phadia immune CAP 250 and immune CAP 1000 system. Reference range for total IgE was 0.35–200 U/L, and for specific IgE 0.1–0.4 U/L.

Allergens from Duck feather, Dog dander, Pigeon droppings, Pigeon feather, Turkey feathers, and Cat hair were collected. Samples were collected from the animals using metabolic cage, for a maximum of 24 hours. Samples of interest were extracted, degraded, washed, and filtered through a 35 mM filter for later use in electrophoresis.

Protein samples were denatured with SDS. Electrophoresis was performed in a solid support (acrylamid). The distance of protein migration towards the anode was inversely proportional to protein size and MW of the sample DTT stain was added to gel, so the bands could be visualised under X-ray.

RESULTS

Serum IgE antibodies against (Table-1) were found in 4 of 10 patients (40%). We found a significant relationship between elevated IgE and the 8%, Basophils ($p=0.002$). Serum from 5 subjects with highest IgE to hair, feather and dander were used in the immunoblotting study of allergens. Hair is a more complex allergen source than droppings with MW allergens, some of which were less clearly differentiated than droppings.

Table-1: Patient details, basophilia and IgE level

Patient	Age	Nationality	Allergy	Basophils (%)	Total IgE	Specific IgE
1	45	Saudi	Asthma	7	1135	108
2	33	Bangladeshi	No	10	73	0.2
3	48	Bangladeshi	allergy	11	874	81
4	33	Saudi	allergy	0	6	0.1
5	38	Bangladeshi	No	0	48	0.1
6	33	Saudi	Asthma – allergy to penicillin	1	201	0.6
7	36	Bangladeshi	Asthma – allergy	1	442	290
8	35	Bangladeshi	Allergy to rabbits	12	1600	49
9	26	Bangladeshi	No	1	0.7	0.3
10	35	Bangladeshi	No	1	91	0.1

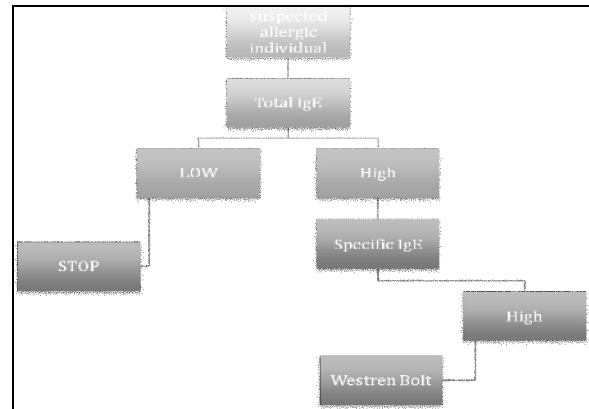


Figure-1: The protocol we used in this study

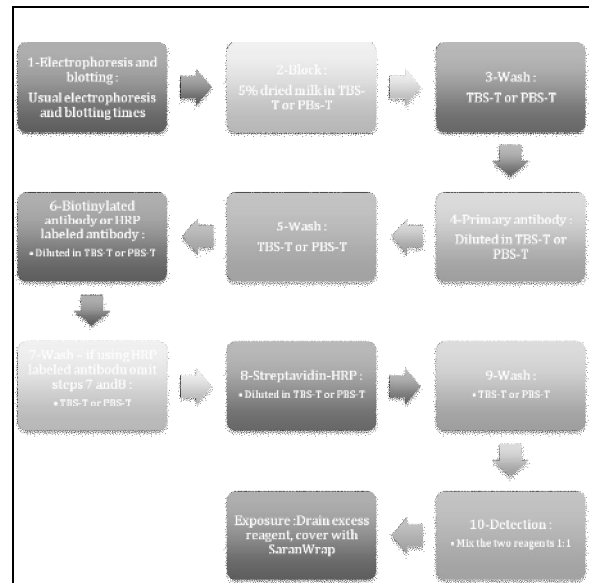


Figure-2: The protocol of the Western blotting

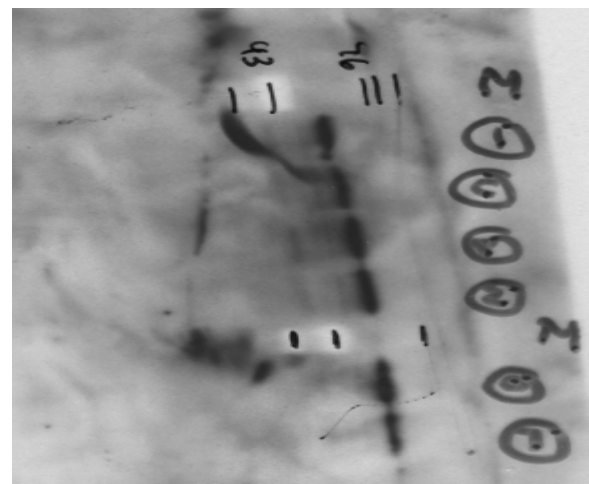


Figure-3: Western blotting result after electrophoresis in 15% gel

It shows also the different allergies. At (60) KDa extracted from: 1- duck feather, 2-Dog hair, 3-Pigeon droppings, 4-pigeon feather, 5-Turkey feather, 6-Cat hair.

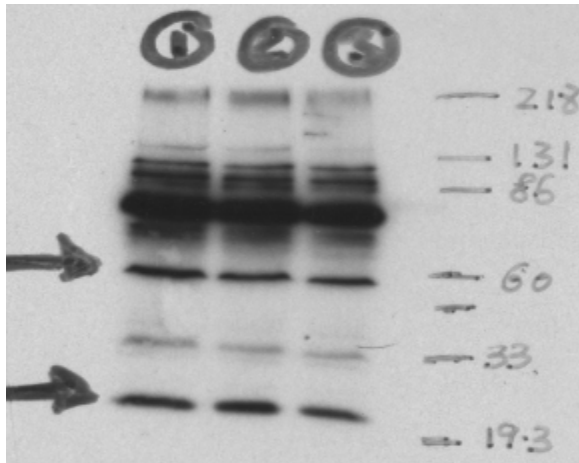


Figure-4: Western blotting results of 3 extracts including: 1-Duck feather, 2-Pigeon Feather, 3-Turkey feather.

There were 2 common bands at (60) kDa and (20) kDa.

DISCUSSION

The use of Tween 20 as a blocking agent has been recently criticised. Although it is thought to help in the detection of low IgE binding components, it has been shown to dissociate protein from nitrocellular membrane at low concentration, and comes as non-specific antibodies binding.^{8,9}

The analysis of allergens, like hair, droppings, feather and dander was also affected by the variable resolution of close bands. In total, 8 allergens were identified. Specific IgE for hair component at 60 and 20 kDa were detected in more than 60% of the sera tested.

Ninety percent (90%) of the sera tested with IgE to 60 kDa allergens. A 20 kDa protein is however a minor component of male feather. A negative result could be influenced by the relative amount of allergens in each extract source and, in this test, would lower the level of significance. This shows there is statistically significant agreement in these subjects which are negative to most of allergens studied. Therefore, further evidence is needed to show that there is immunological cross-reactivity between these extracts.¹⁰⁻¹²

CONCLUSION

Although blood samples are used conventionally in the measurement of IgE and other parameters' levels internationally, and for greater convenience and

validity; this requires repeated sampling, to maintain the sensitivity and privacy of information.

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