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Characterization of Cockroach Allergens as A factor of Dermatitis in Egypt

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ABSTRACT

Cockroach have been recognized as a major cause of asthma in urban environment as the Egyptian randomized areas. Extraction, characterization, and detecting immune response to such allergens were determined. Cockroach allergens proteins were characterized using SDS-PAGE analysis which yield protein fractions of molecular weights ranging from 190-35 KD, while amino acid analysis proved the presence of glycine, cystine, glutamic acid and aspartic acid which sharing with the commercial cockroach extract. HPLC analysis also revealed that there were many peaks closely the same in retention time between commercial and prepared cockroach allergens. Antigen antibody reaction revealed positive reaction to both commercial and local prepared cockroach allergens with human serum samples from persons had highly detectable total IGE-Ab.

INTRODUCTION

Cockroaches have been reported to be associated with asthma in many regions of the world, including U.S.A, Taiwan, Japan, Thailand, Singapore, India and South Africa, Peyton *et al.*, (2001), Fernandez *et al*., (2001), Stelmach *et al*., (2002), and Farrokhi *et al*., (2015).

Several districts of Egypt are highly infected by cockroaches and inhabiting by patients who suffered from sensitivity to such insect, El-Gamal *et al.,* (1995).

In Egypt, two different species of the common native cockroaches are the German cockroach, *Blatella germanica (L.*) and the American one, *Periplaneta americana (L.*) which infest houses, schools, hospitals and other large buildings, Tara (1998).However, sustained removal of cockroach allergen from homes may be difficult to achieve. Cockroach extermination needs to be done in all rooms and should be coupled with efforts to prevent reinfestation to achieve effective control of allergen exposure in the household type, Leaderer *et al*., (2002). Cockroaches are among the most undesirable insect intruders in the home. They are associated with unsanitary conditions, although they occasionally invade the best-kept homes, McConnell *et al.,* (2005).

The insects also produce a secretion that has a repulsive odor and can affect the flavor of food. Cockroaches can cause allergic reactions when sensitive people come into contact with contaminated food or house dust, Hansel *et al.*, (2006).

For nearly a half century, cockroaches have been recognized as a major cause of asthma morbidity in the urban, inner-city environment.

Several cockroach-produced allergens have been identified and characterized, and a few have been produced as recombinant proteins. Gore *et al.*, (2007), reviewed that the current understanding of cockroach allergen biology and the demographics associated with human exposure and sensitization. They also critically evaluate allergen mitigation studies from an entomological perspective, highlighting disparities between successful and failed attempts to lessen the cockroach allergen burden in homes.

This study highlights the characterization and allergenicity of the extracted proteins from cockroach to investigate clinical significance and study of the immune response against cockroach allergens (whole body) of the common native cockroaches which are the German cockroach, *Blatella germanica* (L.) and the American cockroach, *Periplaneta americana* (L.), using hamsters as animal model.

This goal is achieved through seasonal sampling of cockroaches representing different ages were collected from low-cost public housing, extraction of cockroach allergens from the whole body and fragments of the collected cockroaches, determination of total IgE-Ab in sera for patients suffering from sensitivity to cockroach allergen, who live at exposure risk to cockroaches. Identification of the local prepared cockroach allergen extracts, purification, characterization of the major allergen, protein concentration was assayed and detection of protein using PAGE and HPLC techniques.

MATERIALS AND METHODS

Collection of cockroach samples:

The American cockroaches, *Periplaneta americana (L.)* and the German cockroach, *Blatella germanica* (L.)are the most common in Egypt, whole bodies, cast skins, egg shells and feces were considered as dust material during this study. They are most commonly found in restaurants, grocery stores, bakeries, pet shops and other establishments where food is prepared or stored. They can be transported into homes and apartments in boxes from infested establishments. Cockroach index was estimated in many household, the chosen household where the highest cockroach index number which was collected by trapped bags in low socioeconomic levels with highly cockroach infestation. Six households in low-cost public housing studied, all had visible cockroach infestation representing different districts from Egypt.

The collected cockroaches were killed by freezing and then lyophilized to get dry powdered samples for preservation.

Extraction of cockroach allergen:

Crushed cockroach samples were suspended in dulbeco's phosphate-buffered saline (polymethyl sulfonyl fluoride-2 m mol/L, Ethylene diaminete, tetra acetic acid-pH:7- 20m mol/L). Crushed bodies were treated three times with six volume of diethyl ether for defatting, Carsten *et al.* (1990). The resulted mixture was stirred overnight at 4°C and was then centrifuged at 14,000 r.p.m. for 30 minutes at 4°C. Supernatants were filtered through a 0.45 mm Millipore filter, an aliquot of one mg/ml was stored at 4°C and kept for further investigations as prepared crude cockroach allergens extract. A commercial crude extract of American & German cockroach allergen was obtained from (Allergopharma) U.S.A manufactures, it was marked for skin prick testing in solution staining glycerin and phenol, Pollart *et al.*, (1991-A), and used for comparison.

Protein concentration and profile:-

Protein concentration was assayed by total protein colorimetric method [Diamond Diagnostics] using Specord 200 photometer to read the absorbance of the local prepared crude cockroach allergens extract and the commercial crude cockroach allergen extracts, standardized against reagent blank, Henry *et al.*, (1964). Sodium dodecyl sulfate-polyacrylamide gel electrophoresis was carried, using cockroach allergens as samples, according to the method described by Laemmli *et al.*, (1970).

Amino Acids Analysis:-

Amino acid standard mixture was evaluated by Epp Biotronik, LC.3000, amino acid analyzer (Beckman Instrument) , using LC 3000 standard H1, ready-made buffers HI (4-buffer system), column type H 125x4 mm, pre-column type H 60x4 mm were used for identification of the prepared cockroach allergen extract compared with the commercial crude cockroach extracts.

High Performance Liquid Chromatography (HPLC):-

HPLC apparatus Beckman system Gold, dual pump, module 125 was used. The purified allergen samples (the prepared cockroach allergen extract and the commercial crude cockroach extract) were passed through the apparatus to fractionate the crude cockroach allergens, according to Susan *et al.*, (1991).

Determination of antigenic property of cockroach allergen extracts with human serum antibodies:-

Antigen-antibody reaction test was carried out for confirmation antigenic property of the local prepared crude cockroach allergen extracts compared with the commercial allergen extracts using ELISA technique. The sera were used as known antibody samples from persons had highly detectable IgE-Ab compared with normal persons as control samples had normal ranges of total IgE-Ab analysis. Microtiter strip ELIZA plate 96 sterile non coated wells was used in this test.

Statistical analysis:-

The data were analyzed on a personal computer for parametric IgE-Ab using student "t" test.

RESULTS

Characterization of cockroach allergens:

In this study, one thousand, two hundred and forty six cockroaches were trapped from different households for the preparation of the crude allergens, Fig. (1). In each household, cockroach samples were collected from houses, occupied by patient with asthma. The patients have detectable serum total IgE-Ab.

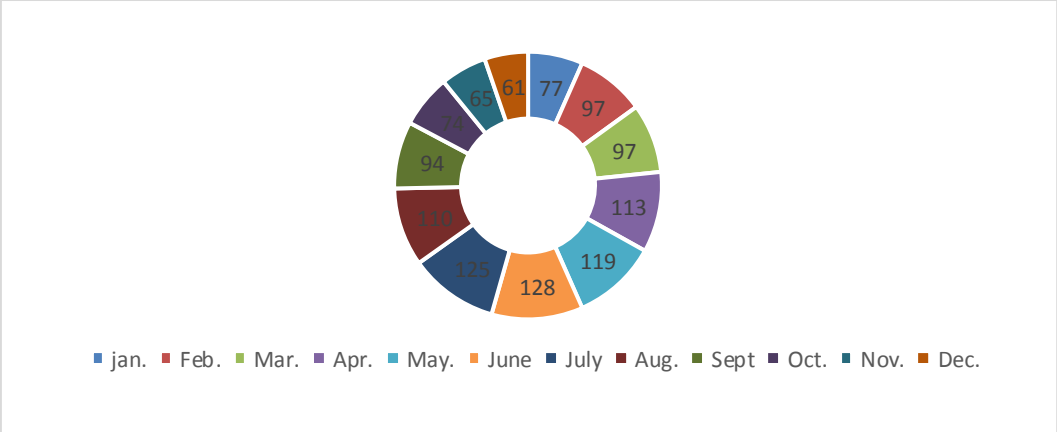


Fig. 1: Total number of the trapped cockroaches / month of the year which were collected from different households.

Determination of antigenic property of cockroach allergen with human serum antibodies:

The human sera were used as known antibody samples. Antigen-Antibody reaction revealed positive reaction to both mixture of equal crude whole body extracts from American & German cockroaches (CRa-M) local prepared and commercial allergen as antigen with the serum samples which were taken from persons had highly detectable total IgE-Ab (ranged from 980-2100 IU/ml), which in case of serum samples were taken from normal persons as a control had normal T.IgE level (ranged from 17-100 IU/ml) the normal is less than 150 IU/ml. The result showed in Table (1). There is a high significant difference between human

serum antibodies compared with human serum total IgE-Ab, as shown in Table (2).

Table 1: Antigenic property of cockroach allergen extracts with human serum antibodies compared with human serum total IgE-Ab.

Total	Ag*/Ab Reaction (IU/ml)	Ag/Ab Reaction (IU/ml)	Total IgE-Ab (IU/ml)
Blank	0.0	0.0	0.0
Serum from Five persons had highly detectable total IgE-Ab	25.0	26.0	1200
	19.0	20.0	980
	30.0	32.0	2100
	27.0	28.0	1500
	21.0	22.0	1000
Mean value	24.4	25.6	1356
Serum from five Persons had Normal T.IgE-Ab level as a Control	0.30	0.33	40
	0.40	0.41	65
	0.20	0.22	17
	0.20	0.20	24
	0.40	0.41	100
Mean value	0.3	0.32	49.2

Ag*/Ab: commercial crude cockroach extract (CRa-M) as antigen / human serum antibodies. Ag /Ab: the total crude cockroach extract (CRa-M) as antigen / human serum antibodies.

Table 2: Comparison between antigenic properties of the allergens with normal human Serum antibodies compared with patient serum total IgE-Ab.

Groups	Means value			S.E			P. values
	Ag*/Ab	**Ag/Ab**	**IgE-Ab.**	**Ag*/Ab**	**Ag/Ab**	**IgE-Ab**	
Control	0.30	0.32	49.2	0.045	0.046	22.1	

Measuring protein concentration and SDS PAGE analysis:-

Protein concentration was assayed by total protein colorimetric method [Diamond Diagnostics] using Specord 200 photometer absorbance; the readings were between 2.8-3.0 mg/dl.

Protein analysis:-

SDS-PAGE analysis (using Image Master analyzer) of crude cockroach, whole body, cast skin and egg shells extracts revealed that the molecular weights estimated to be (190, 139, 132, 106, 90, 75 and 35 KD) and separated protein bands of the crude cockroaches together (lane 3،4) or separated (lane A، G) and the commercial one are measured and represented in Fig. (2).

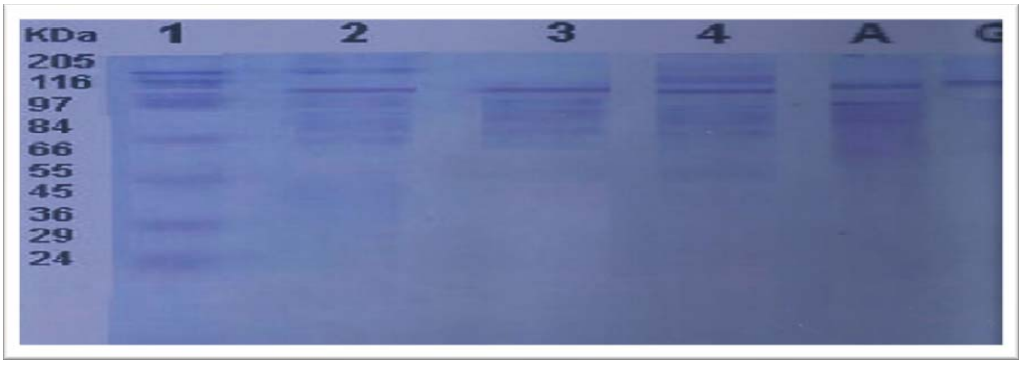
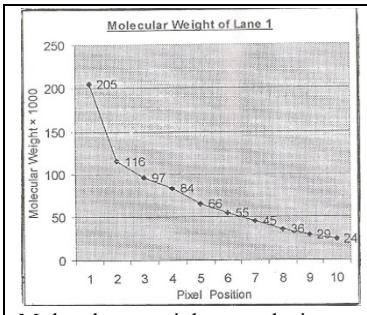


Fig. 2: A photograph of SDS-PAGE of separated components of crude cockroach extracts.
Lane 1:- The high molecular-weight protein marker.
Lane 2:- The commercial crude cockroach extract.
Lane 3:- The local crude cockroaches extract contained 0.4 mg/ml of protein.
Lane 4:- The local prepared crude cockroach extract contained 0.5 mg/ml of protein.
Lane A:- The local prepared crude American cockroach extract,Whole body, cast skin and egg shells contained 0.5 mg/ml of protein.
Lane G: - The local prepared crude German cockroach extract,Whole body, cast skin and egg shells contained 0.5 mg/ml of protein.

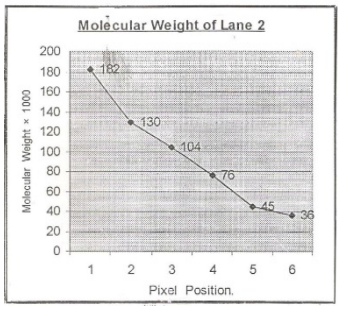
Many bands with MWt ranging from (35 to 190 KD) were separated from the local prepared CRa-C extracts (lane 3&4) and commercial CRa-M extract (lane 2) compared with wide range molecular-weight gel marker (lane 1) proved the purity in preparation of the local prepared CRa-M extracts. At least six antigenic bands with MWt ranging from (35 to 190 KD) of the crude cockroach extracts were visible after staining.

The American crude cockroach allergen extracts was almost sharing in many bands (lane A) with the German CRa extracts (lane G) with total protein concentration ranging between 0.4-0.5 mg/ml.No significant difference of SDS-PAGE patterns was detected between the local prepared extract and the commercial crude cockroach allergen extracts that were manufactured in U.S.A.

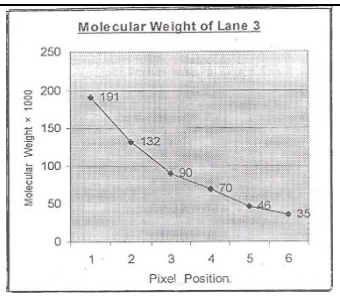
Amino Acids Analysis:- The amino acid analysis of the purified allergens [commercial and prepared crude cockroach extracts allergen] is illustrated in Figs. (3&4) and Tables (3&4). The two crude cockroach allergen extracts were very closely related in the presence of glycine amino acid at the same time (21 minutes). The most notable differences were found in other amino acids, this is due to the commercial CRa-M mixed with phenol and glycerol that made the analysis impossible, on the other hand the difference types of amino acids of the prepared CRa-M of cockroach allergens were found with 49%concentration as ammonium sulphate followed by Cystine 9%, Alanine 6.2%, Histidine 5.2%, Leucine 5%, Glycine & Glutamic Acid4.6%, Aspartic Acid 3.1%. Other types of amino acid were found in low percent as Threonine, Serine, Valine, Methionine, Tyrosine, Phenylalanine and Lysine.



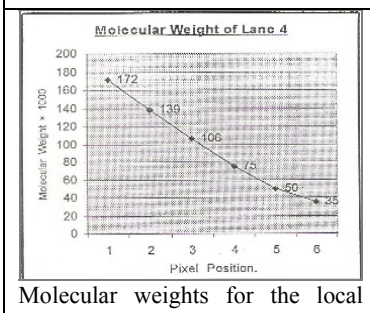
Molecular weight analysis as protein marker



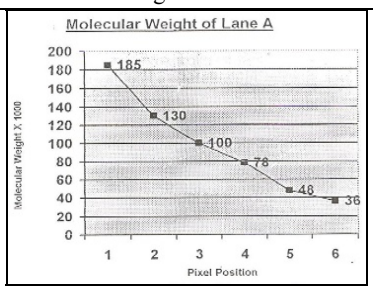
Molecular weights for commercial crude cockroach allergen



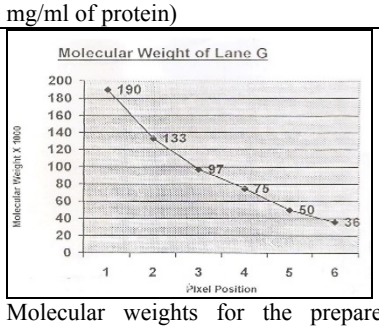
Molecular weights for the local prepared crude cockroach allergen (0.4 mg/ml of protein)



Molecular weights for the local prepared crude cockroach allergen (0.5 mg/ml of protein)



Molecular weights for the prepared crude American cockroach allergen (0.5 mg/ml of protein)



Molecular weights for the prepared crude German cockroach allergen (0.5 mg/ml of protein)

Fig. 3: Molecular weight analysis of cockroach allergent proteins.

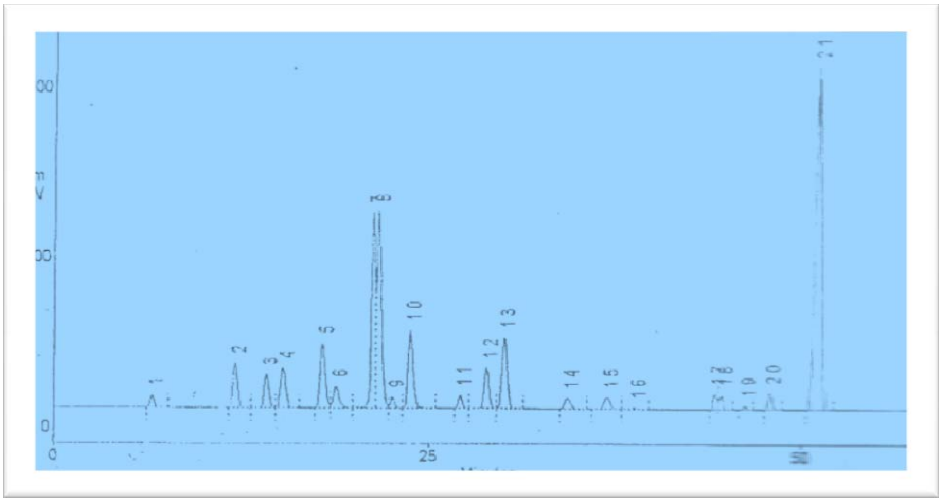


Fig. 4: Amino acids analysis of the local prepared crude cockroach allergen extracts.

Table 3: Amino acids analysis of the commercial crude cockroach allergen extracts

Peak No.	Peak Name	Time	Concentration % Ug/ml	Concentration%
1	Glycine.	21.21	0.50	100.00
Total		----	0.50	100.00

Table 4: Amino acids type related to different retention times and concentrations of the local prepared crude cockroach allergen extracts.

Peak No.	Peak Name	Retention time	Concentration (ug/ml)	Concentration %
2	Aspartic acid.	12.16	6.11	3.14
3	Threonine.	14.26	4.58	2.36
4	Serine.	15.38	4.61	2.37
5	Glutamic acid.	18.02	9.12	4.69
7	Glycine.	21.37	8.78	4.52
8	Alanine.	21.73	12.18	6.27
10	Cystine.	23.79	17.22	8.87
11	Valine.	27.08	1.28	0.66
12	Methionine.	28.82	5.53	2.84
13	Leucine.	30.01	9.71	5.00
14	Tyrosine.	34.26	2.73	1.41
15	Phenylalanine.	36.93	3.15	1.62
17	Histidine.	44.16	10.13	5.21
20	Lysine.	47.88	4.05	2.08
21	Ammonium sulphate.	51.08	95.08	48.95
Total			194.26	100.00

High Performance Liquid Chromatography, (HPLC):

The purified allergen samples were analyzed to detect fractionation of the crude cockroach allergens CRa-M. The purified allergens of the commercial &the local prepared crude cockroach extracts were illustrated in Figs. (5,6 & 7) and Table (5).

The HPLC analysis revealed many distinct peaks which corresponding to the molecular weights calculated from of SDS-PAGE analysis. In comparing between the purified allergens of the commercial & the local prepared crude cockroach extracts, the results revealed that there were many peaks closely the same in retention time as shown in Fig. (7) and Table (5).

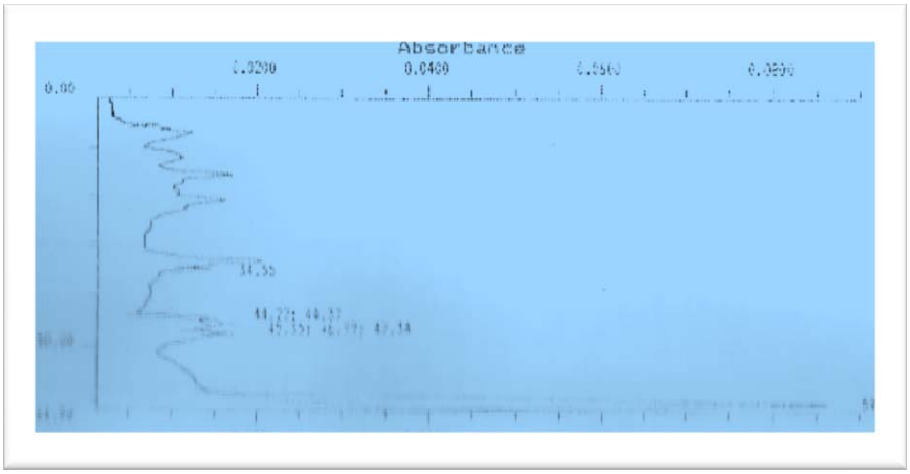


Fig. 5: Purification of the commercial crude cockroach allergen extracts by HPLC gel filtration and fractions were analyzed for protein content.

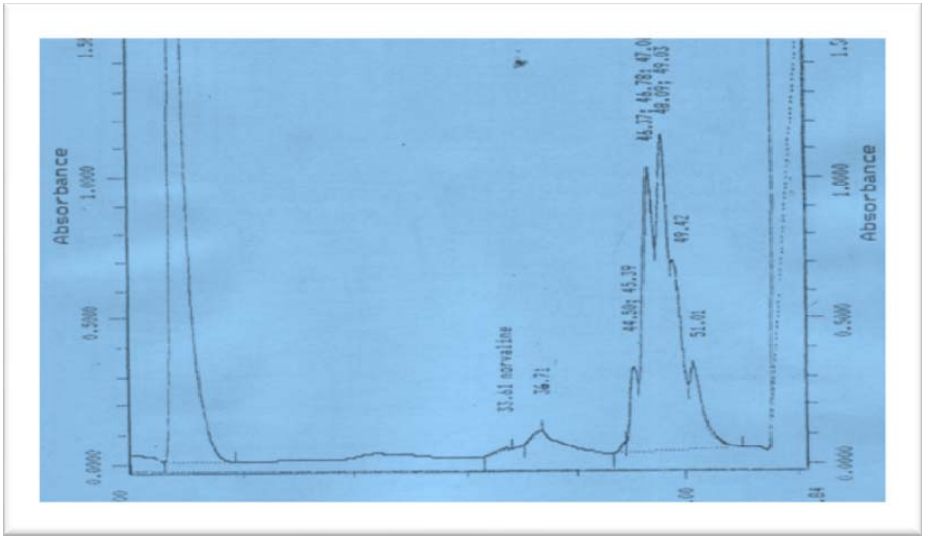


Fig. 6: Purification of the local prepared crude cockroach allergen extracts by HPLC gel filtration and fractions were analyzed for protein content

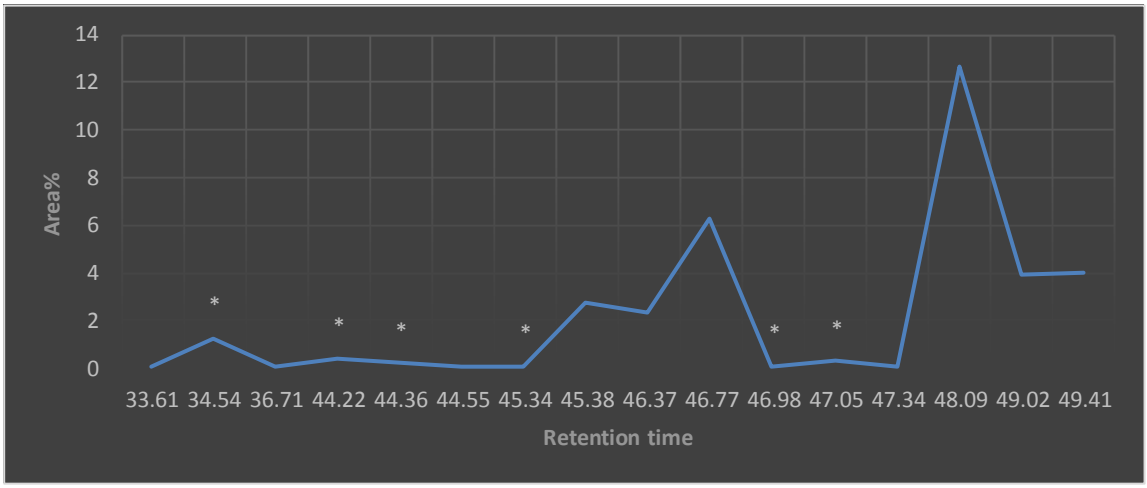


Fig. 7: Comparison between Retention time of local prepared and commercial crude (*) cockroach extracts with the percentage of peak area by HPLC.

Table 5: Purification of the local prepared crude cockroach allergen extracts and the commercial (*) HPLC gel filtration and fractions were analyzed for the protein content.

Peak Number	Retention Time	Peak Area	Peak Height	Area %	Height %
1	33.61	11.89	0.0092	0.122	0.019
1*	34.54	1.255	0.0031	1.273	2.819
2	36.71	8.90	0.0015	0.091	0.003
2*	44.22	0.417	0.0010	0.423	0.919
3*	44.36	0.205	0.0010	0.208	0.897
3	44.55	17.55	0.0346	0.180	0.071
4*	45.34	0.112	0.0005	0.114	0.475
4	45.38	266.41	0.2913	2.733	0.599
5	46.37	227.67	0.6808	2.335	1.398
6	46.77	612.98	0.9938	6.288	2.041
5*	46.98	0.118	0.0002	0.120	0.230
7	47.05	291.78	0.8803	2.993	1.807
6*	47.34	0.025	0.0003	0.025	0.241
8	48.09	1234.54	1.1033	12.66	2.266
9	49.02	381.10	0.6634	3.909	1.362
10	49.41	387.43	0.5994	3.974	1.231
11	51.08	475.73	0.3007	4.880	0.618

*: results for commercial cockroach allergen.

DISCUSSION

In some patient clinic of allergy & Immunology Centers, in Egypt, it was observed that many patients suffering from allergic diseases (atopic dermatitis) and with relatively high titers of serum total IgE-Ab showed positive cockroach skin prick tests, Merdan *et al.*, (2015).

This observation attracted our attention that cockroaches might be the source of allerginicity and also it is an increasingly important public health source of allergens. This assumption may be supported by Bennet *et al*., (1993), Williams *et al*., (1999), and Yeh (2006).

Since the cockroach is a popular insect in houses, we set a plane to prove that cockroach allergens induce atopy.

To achieve this goal, the collected cockroach (whole body) both sexes were killed by freezing and then lyophilized to get dry powder samples for the preparation of the local crude cockroach allergens. This procedure was followed according toZwick *et al.*, (1990) and Zwick *et al.*, (1990).

During the present work, the purified cockroach allergens and the commercial cockroach allergens were examined for the determination of total protein profile by photometric assay. The results revealed similarity between both allergens (2.8 – 3.0 mg/dl). This result may assure that the local prepared allergen is similar to the already manufactured commercial one. It also proved that our preparation is on the right track. Our results ran parallel with those reported by Gore *et al.*, (2007).

Several concentrations of protein extracts for both types of allergens were tested until we reached the suitable electrophoretic analysis by SDS-PAGE of the purified allergens. Total protein were carried out using dilution (0.5 mg/dl) for both types of allergens which gave the best results with SDS-PAGE and showed several protein bands ranging from 190 – 35 KD. These results are in accordance with Wu *et al.*, (2005), who identified the characteristics of major cockroach allergens with molecular weights ranging from 6 to 120 KD.

No significant difference in SDS-PAGE pattern between the local prepared CRa extracts and the commercial one. This result indicates that the responsible protein fractions for induction of allergy are identical in both tested WBE of locally prepared and commercial one. Our result ran parallel with those reported by Gore *et al.*, (2007) and WU *et al.*, (1998).

The protein concentration (0.5 mg/dl) of the CRa extracts identified at least six different bands on SDS-PAGE. The most

dense stained band ranged between 75-35 KD. Similar protein fractions particularly at 35 and 75 KD were reported by Pollart *et al.*, (1991-A).

This finding is slightly different from those determined by Stankus *et al*., (1990).

This concentration in specifying may be due to cockroach species strains and method of preparation of the crude extracts. The similarities and differences between the SDS patterns in this study and those reported by other authors may be related to some extent to the differentiation of SDS analysis of each individual cockroach species. It was found that the protein extracted from whole body of *Blatella germanica* alone was different from M.W of those produced from *Periplaneta americana* WBE.

These differences were detected by Helm *et al*., (1996), and WU *et al.,* (1998), who noticed several allergens to which there was a strong band at 75 KD, while the first author restricted the band at 36 KD.

For subsequent analysis two mixtures were prepared; one is a locally prepared mixture of American & German cockroaches and the other mixture is a commercial allergen from both species.

Antigenic property of cockroach allergen extracts with human serum antibodies was tested by ELISA technique. The two mixtures (allergens) were used as antigens against the human serum samples which were used as antibodies. Antigen-Antibody reaction revealed positive reaction to both locally prepared mixture and the commercial one, in sera sample taken from patients who had a highly detectable total serum IgE-Ab. While negative reactions in sera sample taken from people with normal total serum IgE-Ab. This result may clarify that our prepared allergen is active antigens. Information about the IGE binding epitope of cockroach allergens may help in designing diagnostic and therapeutic approach to cockroach allergy. Lee *et al*., (2015).

The results of amino acids analysis for the local cockroach allergens and commercial allergens revealed that the two crude cockroach allergen extracts (local and commercial) were similar in presence of glycine amino acid at the same time (21 minutes). The most notable differences found in other amino acids were due to the phenol and glycerol mixed with the commercial CRa which made the analysis impossible. The different types of amino acids of the local prepared cockroach allergens were found with 49 % concentration as ammonium sulphate followed by cystine 9%, alanine 6.2%, histidine 5.2%, leucine 5%, glycine & glutamic acid4.6%, aspartic acid 3.1%. Other types of amino acid were found in low percent as threonine, serine, valine, methionine, tyrosine, phenylalanine and lysine. Our findings were almost identical to results of amino acids analysis made by Carsten *et al*., (1990), while Jeon *et al*., (2014), found that using Elisa technique and amino acid analysis investigate allergenicity of German cockroach. They also found that the amino acid sequencing of German cockroach's chemotrypsin allergen showed 32.7 to43.1%identity to dust mite chemotrypsin. On applying another analysis, High Performance Liquid Chromatography (HPLC) analyzer used to determine the fractionation of the purified locally prepared allergen, compared with the commercial formulated one, many peaks were found. The purified allergens demonstrated two overlapping peaks, eluting at 26 to 35 minutes with the sharpest peak eluting at 30 to 34 minutes. The use of this fraction for chromate-focusing studies on whole body extracts of cockroach allergens confirmed the presence of significant antigens. Similar results were reported by Stankus *et al*., (1990), Carsten *et al*., (1990), and Gore *et al.*, (2007).

These results highlight the antigenic properties of cockroaches and more investigations are going on to correlate such antigens with the children allergy and may improve their immunotherapy forms.

REFERENCES

B. A. Merdan, Magda H. Radi, W. A. M. Kamal and E. E. Fouda (2015).

Immunological and Histological Response to Cockroach Allergens in Egypt. European Journal of Biological Sciences. 7 (2): 91-98.

Bennet G. W. and Owens J. M. (1993). Cockroaches as carriers of bacteria. Lancet., 20., 341(8847): 732.

Carsten Schou, Peter Lind, Enrique Fernandez-Caldas, Richard F. Lockey, and Henning Lowenstein, (1990). Identification and purification of an important cross-reactive allergen from American (*Periplaneta Americana*) and German (*Blattella germanica*) cockroaches. J. Allergy Clin. Immunol., 86:935-946.

El-Gamal Y., Awad A., Hosny E., El-basiony S., Galal E., (1995). Cockroach sensitivity in asthmatic Egyptian children. Pediatr. Allergy Immunol. 6 (4):220-222.

Farrokhi S., Gheybi M. K., Movahed A., Tahmasebi R., Iranpour D., Fatemi A., Etemadan R.,Gooya M., Zandi S., Ashourinejad H., Alavizadeh S., and Khoddami S., (2015). Common aeroallergens in patients with asthma and allergic rhinitis living in southwestern part of Iran: based on skin prick test reactivity.

Fernandez-Caldas E.; Codina R.; Ledford D. K.; Trudeau W. L., and Lokey R. F. (2001). House dust mite, cat and cockroach allergen concentrations in day care centers in Tampa Florida. Ann. Allergy Asthma Immunol., 87(3): 196-200.

Gore J. C., and Schal C., (2007). Cockroach allergen biology and mitigation in the indoor environment. Annu. Rev. Entomol., 52: 439-463.

Hansel N. N., Eggleston P. A., Krishnan J. A., Curtin-Brosnan J., Rand C. S., Patino C. M., and Diette G.B., (2006). Asthma-related health status determinants of environmental control practices for inner-city preschool children. Ann. Allergy Asthma Immunol. 97(3): 409-417.

Helm R., Cockrell G., Stanely J. S., Brenner R. J., Burks W. and Bannon G. A. (1996). Isolation and characterization of a clone encoding a major allergen (Bla g Bd 90 KDa) involved in IgE-mediated cockroach hypersensitivity. J. Allergy Clin. Immunol. 98 (1): 172-180.

Henry R. J., (1964). Clinical chemistry. Harper and Row Publishers, New York. P. 181.

Jeon G.K. Y., Son M., Lee J. H., Hong C. S., and Park J. W. (2014). Allergenic Characterization of a Novel Allergen, Homologous to chymotrypsin, from German cockroach. Allergy Asthma Immunol Res. 2014 Dec.18 (Epub ahead of print).

Laemmli U. K. (1970). Cleavage of structural proteins during the assembly of the head bacteriophage T4. Nature 227: 680-685.

LeadererB. P., Belanger K., Triche E., Holford T., Gold D. R. Kim Y., Jankun T., Ren P., McSharry Je., Platts-Mills T. A., Chapman M.D., and Braken M. B., (2002). Dust mite cockroach cat and dog allergen concentrations in homes of asthmatic children in the northeastern United states: impact of socioeconomic factors and population density. Environ. Health Perspect. 110 (4): 419-425.

Lee M. F., Chang C. W., Song P. P., Hwang G. Y., Lin S. J., and Chen Y. H. (2015). IgE-Binding Epitope Mapping and Tissue Localization of the Major American Cockroach Allergen Per a 2. Allergy Asthma Immunol Res. 2015 Mar.5 (Epub ahead of print).

Lee M. F., Chang C.W. Wang N. M., Lin S. J. and Chen Y. H. (2014). Serine protease inhibitor gabexatemesilate attenuates American cockroach - induced bronchial damage and inflammatory cytokine release. J. Investig Allergol Clin. Immunol., 24(5):338-45.

McConnell R., Milam J., Richardson J., Galvan J., Kwong K., and Platts-Berhane K., (2005). Educational intervention to control allergen exposure in the homes of Hispanic childrenin Los Angeles. Clin. Exp. Allergy: 35(4):426-433.

Peyton A., Eggleston P.A., and Arruda L. K., (2001). Ecology and elimination of

cockroaches and allergens in the home. J. Allergy Clin. Immunol, 107(S): 422-429

Pollart S., Smith F., Morris E., Gelber L., Palatts-Mills T., and Chapman M. (1991-A). Environmental exposure to cockroach allergens: analysis with monoclonal antibody-based enzyme immunoassays. J. Allergy Clin. Immunol., 87: 505-510.

Roul S., Leaute-Labreze C., Perromat M., Ducombs G. and Taieb A. (2001). Sensitization to cockroach allergens evaluated by skin tests in children with atopic dermatitis. Ann. Dermatol. Venereol., 128(2): 115-117.

Stankus R.P., Horner W. E. and Lehrer S. B. (1990). Identification and characterization of important cockroach allergens. J. Allergy Clin. Immunol., 86:781-786.

Stelmach I., Jerzynska J., Stelmach W., Majak P., Chew G., Bobrowska M., and Kuna P., (2002). Cockroach allergen Bla g 2 levels in houses in lodz. Pol. Merkuriusz. Lek. 12(72):462.

Susan M., Donald E., Lisa D., Mary L., Thomaa A. E., willliam M. and MartinD., (1991). Identification, quantitation, and purification of cockroach allergens using monoclonal antibodies. J. Allergy Clin. Immunol. 87:511-521.

Tara W. B., (1998). Cubing cockroach and their allergens. Agricult. Rese., 352:374-385.

Williams L. W., Reinfried P. E., and Brenner R. J. (1999). Cockroach extermination does not rapidly reduce allergen in settled dust. J. Allergy Clin. Immunol., 104: 702-703.

WU C. H., and LAN J. (1998). Cockroach hypersensitivity: isolation and partial characterization of major allergens. J. Allergy Clin. Immunol., 101:832-840.

Wu C.H., and Lee M.F., (2005). Molecular characteristics of cockroach allergens. Cell. Mol. Immunol., 2(3):177-180.

Yeh K. W., (2006). Allergens and allergic diseases in children. Acta Paediatr. Taiwan, 47(4):169-174.

Zwick H., Wolfgang P., Kaspar S., Helmuth R. and Theodor W. (1990). Allergenic structures in cockroach hypersensitivity. J. Allergy Clin. Immunol., 87:626-630.

ARABIC SUMMERY

توصيف مواد الصرصور المسببة للحساسية كعامل من التهاب الجلد في مصر

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يعتبر الصرصار احد اسباب الازمات التنفسية و حالات الربو فى الاماكن الشعبية و العشوائية حيث ينتشر تواجده فى المدارس و المستشفيات و المنازلز و قد استهدف البحث مقارنة نسب تواجد الحشرة فى الشهور المختلفة من السنة عند التجميع من المنازل ، كما استهدف عزل و تعريف المواد المسببة للحساسية من هذه الحشرة. و لعزل البروتينات المسببة للازمات التنفسية، تم استخدام Cntsra *et al*. (1990) ، كما تم تنميط بروتينات الحساسية باستخدام تقنية البولياكريمالايد و حساب الاوزان الجزيئية ، و استخدام جهاز تحليل الاحماض الامينية لمقارنة تركيب بروتينات الحساسية فى الحشرات المجمعة من القاهرة و تلك المحضرة للسوق التجارى كعينة حاكمة. و اوضحت النتائج نسبة عالية جدا من التماثل بين المواد المحضرة للحساسية فىالعينتين.وفى دراسة للتعريف بخواص المستحضر تم حقن الفئران من نوع الهامستربروتينات الصرصار و قد استخدمت تقنية قياس الممتز المناعى (اليزا) لقياس قيم الاجسام المضادة المناعية المتكونة (T-EgI-bA) عند استخدام عينات الدم المسحوبة من الاشخاص ذوى الحساسية لمستحثات الصراصير و كذلك الاشخاص الاصحاء للمقارنة.