Journal of Immunology and Allergy
ISSN: 2582-6549
Armentias-Medina A, et al, 2021- J Immuno Allerg
Research Article

Urticaria, Anaphylaxia and Asthma from Contact with Work Air in Farmers and Agronomists Due to Bruchus Pisorum

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Accepted Date: 06-14-2021
Published Date: 07-14-2021

Abstract

Background: There are few reports of contact urticaria from the inhalation of allergens from legume pests.

Objective: To study the origin of an outbreak of contact urticaria, asthma and anaphylaxis in farmers and agronomists who work handling dried peas.

Method: Allergenic extracts composed of Bruchus lentis and B. pisorum, healthy peas, peas treated with aluminum phosphide and parasitized peas were used for in vivo tests (prick-test, oral challenge and bronchoprovocation) in affected patients and in five controls. with a history of atopy from other legumes. In addition, patch testing with live Bruchus pisorum, patch testing with more common insecticides, molecular component analysis, and Ig-E immunodetection were carried out.

Results: Positive responses were found for the prick-test and the bronchoprovocation test to extracts of parasitized peas and B. pisorum, but the oral challenge was negative. A common 25 kDa band for infested peas and a 30 kDa band for infested pea and B. pisorum body was detected in all patients. The response for pea allergens was negative for all patients, unlike controls with a history of allergy to lentils and peanuts.

Conclusion: It was determined that B. pisorum is a cause of symptoms of immediate hypersensitivity mediated by Ig E by inhalation of the allergen or by puncture of spicules or mushrooms of B. pisorum.

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DOI: https://doi.org/10.37191/Mapsci-2582-6549-2(1)-020

Keywords: Bruchus pisorum; Pea; Contact urticaria; Legume allergy.

Introduction

In recent decades, the prevalence of allergic diseases has increased, where legumes play an important role in this global increase, since they are a food consumed throughout the world [1]. In the Mediterranean basin, cases of allergic reactions have increased. in farmers and agronomists who work in the cultivation of legumes such as lentils and green peas. It is not clear whether the pathogenesis is due to the inhalation of legume proteins or parasitic proteins, since there are no commercial extracts of these parasites.

Legumes are frequently infested by parasites of the Bruchidae family, this infestation, especially in the spring months, has been related to the destruction of seeds that cause large losses in the crop, assuming a negative agricultural impact [2]. In addition, within this family, there are numerous species that have been related to hypersensitivity reactions through contact or inhalation of the cooking steam of their allergens when they infest legumes. Hypersensitivity to the parasite itself has rarely been previously described [1,3].

Of the parasites that infest legumes, the species of the Bruchidae (Bruchus) family are the ones that most frequently infest legume crops, being a common parasite of legumes. Adult's measure 4-8 mm and are oval in shape, dark brown in color and macroscopically visible. They have elytra and hind wings where the mites are housed (Figure 1); when attacked they can defend themselves by emitting spicules similar to the pine processionary. The main species in our ecosystem are Bruchus lentis on lentils, Bruchus pisorum on peas, and Bruchus rufimanus on beans [4].

Green pea is the most common processed legume consumed in the United Kingdom and the United States, hence the importance of its allergenic study [5]. The allergens that have been characterized in green pea are: Pis s1 (44 kDa globulin) similar to vicilin that may be responsible for peanut sensitization by Ig E-mediated cross-reactivity bound to vicilin, Ara h 1 (peanut protein) [5,6], Pis s2 (63 kDa covicillin) [5] which together with Pis s1 make up the main pea allergens capable of cross-reacting with the main lentil allergen Lenc1 [7], Pis s IFR (isoflavone reductase) similar in 56% to the IFR of pollen from birch [8] and Pis s8 (profilin), allergen with cross-reactivity to grass pollen mediated by Ig E [9]. It has been determined that the albumin fraction is the cause of the major allergic symptoms, and that the globulin and glutelin fractions assist the allergenic power of the green pea [8]. A common peculiarity in most legume allergens is their ability to resist thermal, chemical and proteolytic denaturation, however, the albumin fraction is capable of maintaining its allergenic power even when it reaches temperatures of 60°C for 30 minutes or 100°C for 5 minutes [10].

Green peas have been implicated in allergic reactions (itching, abdominal pain and urticaria) in atopic patients when eaten cooked, tolerating when handled and eaten raw, showing a positive result for boiled peas and negative when fresh which could suppose

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a new hypothesis that the pea, when exposed to high temperatures, could increase its allergenicity through a change in the protein structure such as the formation of neoantigens or stabilization of bonds [11]. Cases of bronchial asthma have also been described after being exposed to cooking vapors, the skin test being positive for the extract of legumes and notably positive in the specific IgE test for legumes, subsequently the presence of cross-reactivity between legumes was demonstrated [12]. Occupational asthma has also been reported after exposure to pea flour [13,14].

On the other hand, the lentil pest, Bruchus lentis (bruchidae family), has been implicated in the presence of IgE-mediated rhinoconjunctivitis and occupational asthma in farmers and agronomists who came into contact with inhaled and ingested particles of lentils infested with B. lentis [4,15]. In these cases, the prick test, bronchial and oral provocation test were found to be positive for infested lentils and B. lentis extracts but negative for non-infested raw or boiled lentils [4]. In addition, allergic symptoms caused by other legume pests, such as the bean plague, Apion Godmani [16], and a relationship between sensitization to Eurygaster and Ephestia with episodes of occupational asthma have been described in bakers who work with cereals, demonstrating that parasitized cereals showed a significant increase in their allergenic potential compared to pure wheat flour [17].

Figure 1: Bruchids parasitize legumes and in turn they are parasitized by mites. Bruchus pisorum mite, physiogastric female.

Objective

In the study hypothesis, it is considered whether the cause of allergic symptoms in farmers and agronomists who work with peas is caused by the pea proteins themselves or by contact with the parasite B. pisorum, which is a plague of peas in our country. ecosystem. Patients with immediate hypersensitivity symptoms (contact urticaria, asthma, and anaphylaxis) related to inhalation of B.

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pisorum-infested pea dust are studied by in vivo and in vitro studies.

The objective is to determine the cause of airborne contact urticaria, asthma and anaphylaxis in farmers and agronomists studying the disinfection of dried peas.

Material And Methods

An analytical observational study of cases and retrospective control of patients who came to the Hospital has been carried out.

Patients

Symptoms of immediate hypersensitivity (contact urticaria, asthma and anaphylaxis) were observed in six patients (3 farmers and 3 agronomists) after inhalation of pea powder infested by Bruchus pisorum. The patients had dedicated themselves to cultivating and disinsecting the infested pea. and not infested for years with no problem. They related their symptoms to a treatment of seed peas with aluminum phosphide, with rodenticidal and insecticidal activity, the symptoms began. Both agronomists and farmers had been working while handling the peas without protection, highlighting in them skin lesions similar to those caused by the pine processionary (Figure 3): immediate and transitory contact urticaria with pinkish lesions and an edematous base [18]. These lesions appeared after 30 minutes, were located on the fingertips and disappeared after a few hours. All patients consumed legumes and peas in their diet without presenting the symptoms described above.

As controls, 5 patients with immediate allergic symptoms due to sensitization with other legumes, specifically lentils and peanuts, were chosen.

Through a detailed anamnesis and through a more targeted survey, the most relevant demographic data of the patients were obtained.

It was decided to carry out an in-depth allergological study of all the possible allergenic sources involved to assess the origin of the symptoms observed in these patients.

Plague

The parasite was identified as B. pisorum belonging to the Bruchidae family of the Coleoptera order, a spice native to Asia and found mainly in Europe, making up the main pest of peas. Examples were identified using the dichotomous key of Yus-Ramos et al. (Figure 2), capable of taxonomically characterizing the seed beetles found in Europe based on the external morphological characters of the adult [19]. The microscopic examination of the samples was carried out by means of scanning electron microscopy.

Extract for diagnosis

Diagnostic extracts were prepared from infested and non-infested dried and boiled peas, non-infested peas treated with aluminum phosphide and infested peas treated with aluminum phosphide, B. pisorum, B. lentis and lentils infested by B. lentis, used for the skin prick test (SPT), oral and bronchial provocation test, and for in vitro determinations. On the one hand, infested and non-infested peas treated with

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aluminum phosphide with live insects were ground for 30 minutes to obtain fine powder.

On the other hand, whole body extracts of Bruchus, infested and non-infested peas are prepared in 5% phosphate buffered saline. These preparations were centrifuged at 17,700g for 30 minutes, they were dialyzed until a molecular weight of 3.5 kDa was obtained as a cut-off point and finally they were sterilized by means of a filter with pores of 0.22 µm in diameter with the objective of avoid the passage of endotoxins. The concentration of allergenic proteins was determined using the Lowry method [20] shown in Table 1. For the prick-test, part of the extracts was 50% glycerinated. For the bronchial challenge test, another part of the extract was adjusted to 1mg / ml in 9% saline solution. For the oral challenge, healthy and infested peas were boiled for 30 minutes. The remaining extracts were used for in vitro studies.

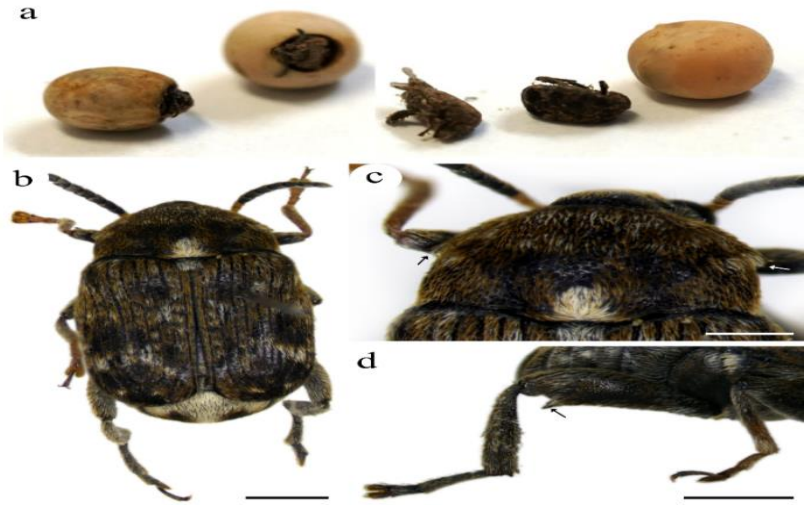

Figure 2: Bruchus pisorum, the pea weevil. A) In and out of the pea. (B-D) Body of Bruchus pisorum. (B) Whole body. (C) Detail of pronotum, showing teeth. (D) Detail of the posterior femur with the characteristic large pointed setae (arrow).

Allergenic extract	mg protein/ml extract
Healthy boiled pea	5.19
Healthy boiled lentil	6.22
Pea infested by B. pisorum not treated with aluminum phosphide	2.9
Pea infested by B. pisorum treated with aluminum phosphide	3.2
Bruchus pisorum	1
Bruchus lentis	0.47
Lentils infested by B. lentis	3.33

Table 1: Protein concentration of the allergenic extract used for diagnosis.

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Tests performed

Skin puncture test (Prick test) The prick tests were carried out with a standard battery of extracts from AlK-Abelló (Madrid, Spain; composed of pollen, mites, animal dander, food and mold), infested, non-infested and healthy peas, and extracts from the entire Pisorum body following a protein concentration of approximately 1mg/ml. Together, they include extracts from storage mites, B.Lentis body, and lentils infested with B.Lentis extracts as previously described [15]. A wheal area of 7mm2 or a diameter greater than 5mm at 15 minutes after puncture is considered positive. A histamine phosphate compound (10mg / ml) was used as a positive control and 0.9% sterile saline solution was used as a negative control.

Patch test

This test was carried out with the most common insecticides used in legume cultivation: propiconazole, metaldehyde, formaldehyde, mercaptobenzothiazole, cypermethrin and isoproturon. The patch test could not be performed with the insecticide currently being used in the crop, aluminum phosphide, as it can be fatal on contact with the skin (H310), ingestion (H300) or inhalation (H330). According to the committee's precautionary advice, the insecticides were tested at 0.01% allergen. and vehicle (petrolatum). The test was read on day 2, 3 and 4.

Open epicutaneous test

For the open application test, the B. pisorum parasite was applied directly on the affected skin or on healthy skin measuring 3x3 mm both in the upper part of the back and in the flexor area of the arm. The reaction is characterized by an urticarial lesion in the form of a wheal or welt that was measured at 20, 40 and 60 minutes after the application, thus avoiding the loss of the positive reaction since the reaction normally appears after 15-20 minutes but is possible delay up to 45 or 60 minutes. The skin was then observed at 7, 48, 72 and 96 hours for possible late skin reactions (Figure 3).

Broncoprovocation test

This test (BCT) was recreated following previous reports to all patients including study cases and controls [15]. An extract consisting of infested peas and whole bodies of B. pisorum at a concentration of 0.01mg/ml diluted in 0.9% saline solution was used as preparation, used as dilution buffer.

Oral provocation test

The oral challenge test was performed in a double-blind, placebo-controlled study, using infested peas not treated with EPA, since ingestion can be fatal. All patients were informed and asked to sign the informed consent. They were recommended to avoid suspicious foods and medications for at least a week before the test, and steps were taken to control reactions that could cause death. The infested peas were ground to obtain a fine powder that was introduced into empty opaque capsules keeping double blind, the initial dose for each patient depended on their previous allergic response, for example, the farmer who had previous symptoms of anaphylaxis when working with peas. An

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initial dose of 5 mg was administered, increasing to 500 mg in a capsule when no symptoms occurred. An opaque sucrose-based capsule, randomly administered to patients during this test, was also included as a negative control. There were sample losses for the test since an agronomist with asthma symptoms, and a control with asthma symptoms and lentil anaphylaxis, did not give their consent. All symptoms and signs were recorded before each challenge.

Determination of specific IgE/Component diagnosis

For the determination of legume-specific Immunoglobulin E, the CAP system (Thermo Fisher Scientific, Uppsala, Sweden) was used, based on a fluoroenzyme-immunoassay (FEIA) capable of detecting very low levels of serum specific Ig E [21]. The ADVIA-Centaur platform from Bayer Diagnostics (Tarrytown, New York) was used. I don't know if I have to reflect these references: yes, the commercial names and city of the supplier are mandatory) determining the Ig E for the pea protein: Pis s1, Pis s6 and Pis s8. Pis s1 or vicilin was purified and labeled with biotin using as indicated in other studies [22]. For the diagnosis by allergenic components or molecules, the ISAC 112 panel (thermo Fisher Scientific) was used, capable of carrying out a simultaneous and grouped study through a biochip with 112 components of 51 allergens that are coupled in a microchip [21].

Immunodetection Ig E

The immunoblotting or electrophoresis test for the study of allergenic proteins was performed on polyacrylamide gel with 15% sodium dodecyl sulfate using reducing conditions. Each lane was loaded with 0.5 µg extract from peas, lentils, infested peas, infested lentils, B. pisorum, and B. lentis. Once each lane has been loaded with the allergenic extract and thanks to SDS-PAGE (polyacrylamide gel electrophoresis with sodium dodecyl sulfate), the proteins are denatured, losing their three-dimensional structure and separating easily, and can be electrotransferred to a nitrocellulose membrane (Hybond-ECL, Amersham biosciences, Little Chalfont, UK), blocking with 5% bovine serum albumin in phosphate buffered saline (PBS) for one hour at room temperature. The membranes were incubated following a 1/5 dilution in patient sera and then with a 1/1000 dilution of murine monoclonal anti-human IGE antibody (Mouse Anti-Human IgE Fc-HRP, Southern Biotech, Birmingham, Alabama). Finally, Ig E-bound proteins were detected by enhanced chemiluminescence (Perkin Elmer Life Sciences, Boston, Massachusetts). The transferred allergenic protein extracts were incubated only with 5% bovine serum albumin to be used as negative controls.

Results

We studied 6 patients, three agronomists and three farmers, who had been cultivating and collecting peas for more than 10 years, two of them were women agronomists and the rest were men, with the mean age of the patients being 49 years. The demographic and clinical data of the patients together with the results of the in vivo tests (prick-test, BCT, OAT and OC) and in vitro (specific IgE, component diagnosis and Western-Blot) are shown in

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Table 2. The most common symptom among our patients was contact urticaria associated with asthma in four patients (1,3,5,6), 5 in the symptomatic context of anaphylaxis. Five of them also presented contact urticaria. Regarding contact urticaria, the lesions frequently appeared in the abdominal area and on the fingertips attributed to the manipulation of the peas, causing stabbing pain and itching of the skin after 15 to 30 minutes of contact. (Figure 3). Morphologically, they appeared as a wheal-shaped urticarial lesion with an edematous and erythematous base, very similar to that caused by the pine processionary [18], suggesting contact urticaria by air. Usually, our patients were in constant contact with large quantities of dry peas, which were handled without gloves or any other type of protection, suggesting the importance of contact with potentially allergenic proteins from both the pea and its parasite. They used to work seated at sorting tables where they sorted the peas and stated that they frequently found live insects when emptying the bags of peas into the planter.

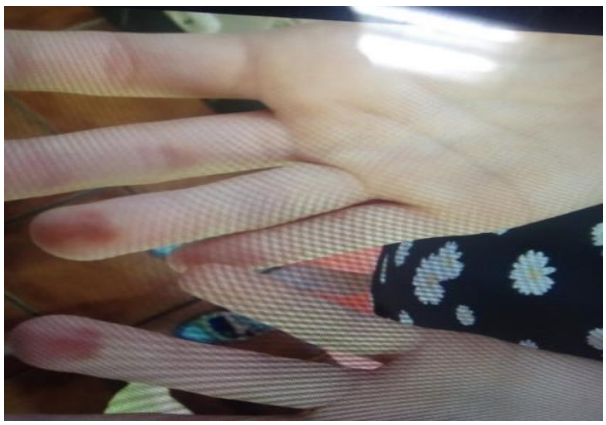

Figure 3: Fingertip lesions in an agronomist handling pea infested by live B. pisorum.

The patients presented these lesions 15 minutes after the separation of the peas parasitized by live B. Pisorum. They are erythematous lesions with an edematous base in the shape of a wheal or welt that suggests contact urticaria by air, similar to the lesions caused by the pine processionary. The results of the in vivo and in vitro tests, collected in Table 2, were analyzed.

In vivo test results

The skin prick test (SPT) was positive for dried and boiled peas parasitized and treated with EPA in all patients and negative in controls, for dry and boiled peas not infested were negative in all patients and positive for two controls that presented asthma due to lentils, which may suggest the cross-reaction between the vicilin of the pea Pis s1 and the lentil allergen lenc 1 [7], for prick with bruchus pisorum they were positive in all patients and negative in controls.

- All patients presented a skin reaction similar to that described above (figure 3) in the upper back and flexor arms 20 minutes after contact,

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disappearing after 24 hours in the open patch test (OAT)

- In the oral provocation test, one patient and one control did not give their consent to carry out the test and the rest of the sample was negative for the test with infested peas, not showing any symptoms when ingesting infested peas in the form of fine powder placed in an opaque capsule.
- For the bronchoprovocation test, infested pea extract and live Bruchus pisorum extract were used, experiencing symptoms of bronchospasm with the first extract in five of six patients and with the whole-body extract of bruchus in all cases. Only two controls who presented asthma and lentil urticaria were positive with the infested pea extract, the control with lentil urticaria symptoms remaining positive in the bruchus test.
- The patch test with insecticides was negative for the entire sample.
- The open skin test with live bruchus was positive in all 6 patients.

Patient / sex	**Age**	**Stage symptoms UC**	**Prick raw/boiled healthy pea**	**Prick parasitized raw/boiled pea treated with EPA**	**Prick B. pisorum**	**OAT with Bruchus**	**OC infested pea**	**BCT with infested pea**	**BCT with Bruchus extract**	**Patch test**
1. ABC/H/agronomis	43	Asthma	-/-	6x6/3x3	4x4	+	NR	+	+	-
2. AIG/M/agronomis	47	UC etapa 3 VK-M	-/-	5x5/3x3	5x7	+	-	+	+	-
3. IAC/M/ agronomis	42	Asthma /UC stage 2 VK-M	-/-	5x5/3x3	5x5	+	-	-	+	-
4. GHT/H/ farmer	54	UC, stage 2 VK-M	-/-	3x3/3x3	3x3	+	-	+	+	-
5. JCC/H/ farmer	57	Anaphylaxis/UC; Etapa 3 VK-M	-/-	3x3/3x3	2x2	+	-	+	+	-
6. IDG/M/ farmer	56	Asthma/UC, stage 1 VK-M	-/-	5x5/3x3	5x5	+	-	+	+	-

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7. Control	32	Urticaria/asthma for lentils	+/+	-/-	-	-	-	-	-	-
8. Control	37	Urticaria-peanut esophagitis	-/-	-/-	-	-	-	-	-	-
9. Control	18	Asthma/ Anaphylaxis for lentils	+/+	-/-	-	-	NR	+	-	-
10. Control	42	Peanut Urticaria	-/-	-/-	-	-	-	-	-	-
11. Control	30	Lentil urticaria	-/-	-/-	-	-	-	+	+	-

Table 2: Demographic and clinical data of patients and controls. BCT, bronchoprovocation test; UC, contact urticaria; NR, not done; OAT, open epicutaneous test; OC, oral provocation test; VK-M, von Krogh G; H, man; M, woman

In vitro test results (Figure 4)

- Ig E polyacrylamide gel electrophoresis for Bruchus and infested peas revealed serum Ig E reactive protein bands in all patients and controls 7, 9 and 11. In contrast, in the specific IG E immunoblot test for Len c1 (lentil allergen) no pea-specific Ig E protein band was revealed.
- Ig E-reactive protein bands were detected in controls 7 and 9 for infested lentils.
- In all patients it was detected: a 25 kDa band present in infested peas and lentils but not for Bruchus pisorum and Bruchus lentis; 18 kDa bands (found in previous studies of B.lentis asthma) (15) related to asthma and urticaria in all patients and in control 9 (contact urticaria /lentil asthma) and 11 (asthma /lentil anaphylaxis), which had a positive bronchoprovocation test to infested peas; band of around 30 kDa for infested peas and B.pisorum in all patients and in control 9, who started the symptoms when he started working in a legume warehouse.
- It seems that there is a partial recognition of proteins, which may lead to a positive response due to a cross-reaction by allergens of lentil (Lenc 1) and pea (Pis s1).

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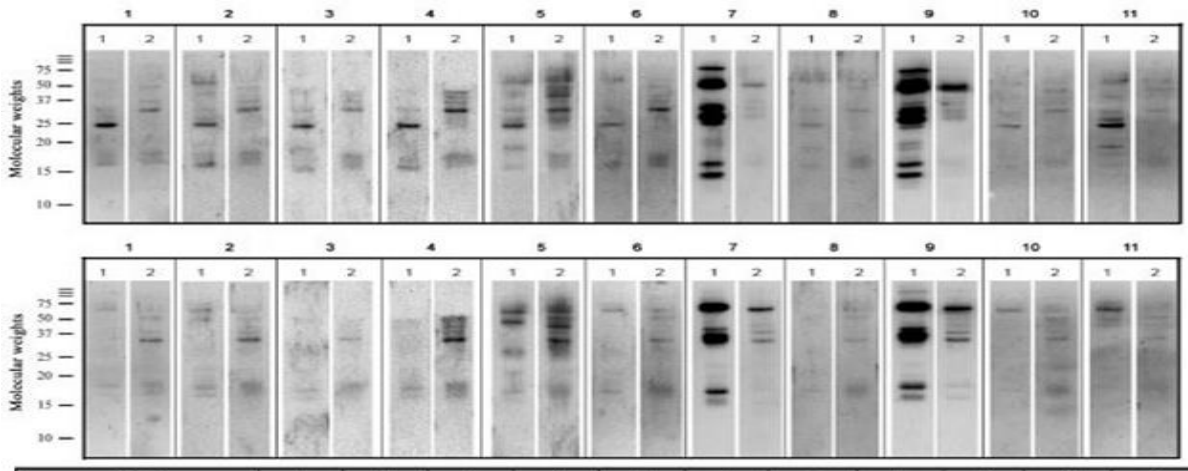

Patient	1		2		3		4		5		6		7		8		9		10		11	
Peas (A) / *B.pisorum* (B)	(A)	(B)	(A)	(B)	(A)	(B)	(A)	(B)	(A)	(B)	(A)	(B)	(A)	(B)	(A)	(B)	(A)	(B)	(A)	(B)	(A)	(B)
75 kDa										x			x				x					
55 kDa										x							x					
48 kDa		x	x						x	x	x		x	x			x	x				x
42 kDa			x	x		x		x		x		x					x	x		x	x	
37 kDa		x		x		x		x		x		x					x	x		x	x	x
35 kDa	x	x	x	x	x	x	x	x	x	x		x	x	x	x	x	x	x	x	x	x	x
28 kDa													x				x				x	
25 kDa	x		x		x		x		x		x		x		x		x		x		x	
22 kDa													x				x					
21 kDa													x				x				x	
20 kDa									x				x		x		x				x	
19 kDa	x	x	x	x	x	x	x	x														
17 kDa	x	x	x	x	x	x	x	x	x	x	x	x	x		x	x	x		x		x	
15 kDa													x				x					
14 kDa																	x					
13 kDa																	x					

Patient	1		2		3		4		5		6		7		8		9		10		11	
Lentils (C) / *B. lentis* (D)	(C)	(D)	(C)	(D)	(C)	(D)	(C)	(D)	(C)	(D)	(C)	(D)	(C)	(D)	(C)	(D)	(C)	(D)	(C)	(D)	(C)	(D)
100 kDa																	x	x				
60 kDa	x		x						x	x	x	x	x	x		x	x	x	x	x	x	x
48 kDa		x		x		x	x	x			x	x		x	x		x		x		x	x
42 kDa								x		x		x	x	x			x	x		x	x	
37 kDa		x		x				x		x		x	x	x			x	x		x		x
35 kDa		x		x		x		x		x		x	x	x		x	x	x		x		x
18 kDa	x	x	x	x	x	x	x	x	x	x	x	x	x		x	x	x	x	x	x		
17 kDa		x	x	x	x		x	x	x	x			x				x	x				

Figure 4: Ig-E Western Blot.

Discussion

Bruchus pisorum is an exotic species native to Asia that in ancient times was dispersed throughout Europe where it has managed to acclimatize, being considered one of the most intractable pests of peas in this area [23,24]. We studied an outbreak of contact uritis, asthma and anaphylaxis in workers who handled peas infested by B. pisorum.

Based on the clinical history of our patients, all of them presented symptoms of atopy after having handled peas for long periods of time. As described in previous studies, B. lentis proteins could be a cause of rhinoconjunctivitis and IgE-mediated asthma in patients who inhale allergens from lentils infested by this parasite, as well as a cause of occupational asthma in parasitized lentil crop workers [4,15].

Legumes are a rich source of proteins, lipids and vitamins, making it a food consumed throughout the world. Cases of allergic

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adverse reactions after ingestion have been reported in IgE-mediated sensitized patients, but cases of allergic reactions after inhalation of its vapor have been rarely reported, however, in the updated literature, symptoms of immediate hypersensitivity (asthma, rhinitis, angioedema, oral pruritus) after exposure to legume vapors in both adult and pediatric age [1]. Cases of occupational asthma due to exposure to pea flour have also been reported in the scientific literature in flour mill workers that processed peas, starch and protein [14], and cases of occupational asthma in workers in the parquet industry who used pea flour for industrial parquet processing [13].

Therefore, knowing that the pea is a legume that is consumed all over the world and therefore its cultivation is very widespread, we reviewed the scientific literature where cases of allergy to pea flour are reported after inhalation [13,14], recently cases studies that indicate that there are allergic symptoms in lentil workers due to its parasite [4,15], other reported cases of asthma due to legume pests [17] and knowing that B. pisorum is a cosmopolitan pest of pea in Europe capable of transmitting its allergenic potential leads us to raise the problem of the origin of this outbreak after handling infested peas.

B. pisorum is an insect capable of biting with its teeth, but due to its small size it is unlikely to damage the skin causing the welt-like lesion that our patients presented. After analyzing the results, it seems more likely that the symptoms reported in these workers are caused by contact with the body of B. pisorum, either directly on the skin of the abdominal area when insects fell when shaking the bags of peas and with the skin of the fingertips (Figure 3), or by the inhalation of allergenic particles that were found in the ambient air breathed by these workers. However, the possibility that the skin lesions were caused by the penetration of parts of the parasite in affected patients cannot be ruled out.

No patient had hypersensitivity to other legumes or cross-reactive allergens, but had a positive bronchoprovocation and skin test for parasite-infested peas. However, controls 9 and 11, who presented asthma / anaphylaxis due to lentil and urticaria due to lentil respectively, were positive for the bronchoprovocation test for infested peas, which may suppose a positive response due to a cross-reaction due to allergens from the lentil (Lenc 1) and the pea (Pis s1).

Taking into account that our patients had worked for approximately 10 years in the cultivation of peas with different insecticides, it was suggested that these insecticides had a role in relation to the injuries suffered by our patients since at the time of the outbreak, the insecticide had changed from magnesium phosphide to aluminum phosphide for the control of the vole plague that transmits tularemia in our region according to the regulations of the Ministry of Agriculture at the time of the study.

This hypothesis that was raised at the time of the study has been clarified due to several reasons: firstly, all the patients were negative for the patch test with insecticides that can cause symptoms of delayed type IV hypersensitivity, although it could not be done with phosphide. aluminum due to its

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high organic toxicity after inhalation, contact or ingestion [25], which was to be expected since the skin lesions of our workers were not compatible with contact eczema caused by insecticides but rather with urticarial lesions after bite, inhalation or penetration of the skin by the pointed hairs of B. pisorum (Figure 5).

Therefore, patients were recommended to wear individual protection with masks, gloves, goggles, clothing with a closed collar and cuff to avoid contact by air. Those patients with symptoms of anaphylaxis were instructed to use intramuscular autoinjectable epinephrine in the lateral part of the quadriceps [26], and those with symptoms of contact urticaria were treated with descending doses of corticosteroids and antihistamine. Thanks to all these measures, our patients have been able to continue working since their symptoms subsided, although their asthma is still persistent but milder.

Attending to different scientific studies, we can observe that there are numerous strategies to reduce the infestation capacity of the pea beetle (B. pisorum), among some of them it is worth mentioning that the use of chemical markers for the resistance of new pea crops it can be an effective method of defense against B. pisorum, but at the same time they are very toxic and can even alter the allergenic power of its proteins [27]. Also in a recent study, he declared that the creation of neoplasm in peas has the ability to obstruct the entry of weevil larvae in the absence of ultraviolet light, which was a problem for field cultivation but it was significantly demonstrated that by means of the Intercropping of pea with maize can facilitate neoplasm formation by preventing the entry of weevil larvae into developing seeds [28]. Likewise, it was found that the expression of alpha-A1 in greenhouse-grown transgenic peas can offer complete protection against the pea weevil and even be safer for those who work in pea cultivation [29].

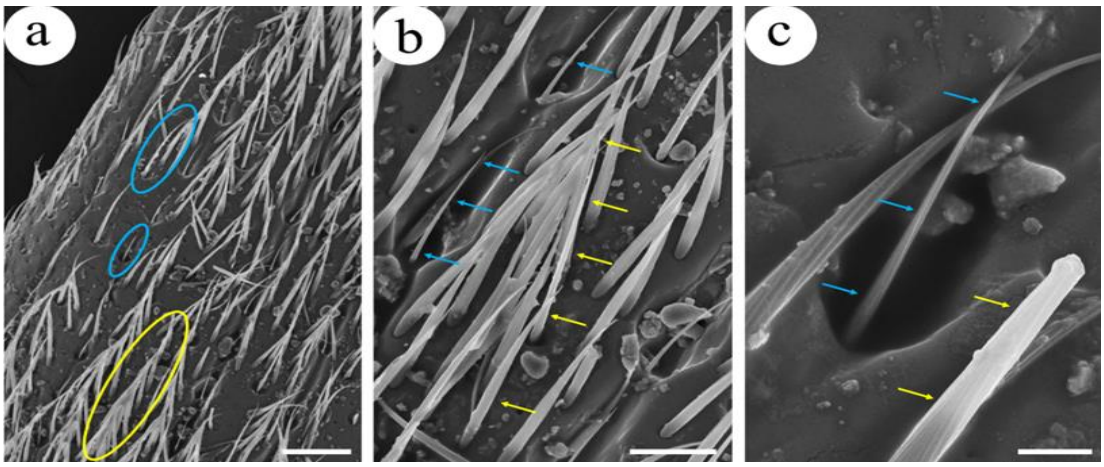

Figure 5: Elitra from B. pisorum. A: Two types of hairs in B. pisorum elytra: grouped to form prominent spikes (in yellow). Others solitary and pointed (in blue). B: Details of the morphology of the hair types (yellow and blue arrows). C: Detail of a solitary and pointed hair (blue arrow). A-C: Scanning electron microscopy. Scales: a: 100µm; b: 40µm; c: 10µm.

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Conclusion

Immune-allergic contact urticaria requires prior exposure to an allergen. All the patients were atopic and worked for many years in pea cultivation, consistent with the development of this rare occupational manifestation.

Therefore, the proteins of the pea weevil (B. pisorum) are capable of causing Ig E-mediated contact urticaria, anaphylaxis and asthma in those patients who inhale particles of the infested pea or by contact with the hairs of the parasite.

Therefore, the knowledge of the pests that infest the legumes responsible for immediate allergy symptoms is important to evaluate the possible allergic manifestations in farmers and agronomists who work with legumes.

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