



Molecular profiles: A new tool to substantiate serum banks for evaluation of potential allergenicity of GMO

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ARTICLE INFO

Keywords:

Component-resolved diagnosis

Profilin

Lipid-transfer protein

Food allergy

Olive pollen allergy

ABSTRACT

Assessment of the allergenicity of GMOs involves performing a test with a panel of sera obtained from allergic donors. However, there is no clear indication of how to characterize the above-mentioned panel. The patient selection criteria should take into account the geographical location of patients, the intensity and nature of the environmental allergens in the area and the potential cross-reactivity among allergenic molecules. Sera for serum banks, obtained from patients with demonstrated food allergy, should be subjected to a further characterization by screening with a panel of relevant allergenic molecules. A representative panel of these sera should be used in the allergenicity assessment. Finally, the “in vitro” methodologies should have the adequate specificity and sensitivity, and the integrity of the molecules tested should be guaranteed.

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1. Introduction

Assessing the allergenicity of a new genetically modified (GM) food is a complex task. Since 1996, there have been several consensus documents and scientific opinions regarding such allergenicity assessment (FAO/WHO, 2001; Guidance document of Scientific Panel on GMO, 2004; Goodman et al., 2005; Spök et al., 2005). When introducing a new gene, there are three possible outcomes linked to allergenicity. First of all, the product of the transgene could have allergenic properties as such. To minimize this risk, a careful assessment of sequence similarities with a well-established allergen sequence data bank should be performed. Secondly, the new protein(s) expressed might sensitize the exposed population. The pepsin stability test should at least minimize this possibility, as it is well known that severe food allergens are, in most cases, stable to proteases (Astwood et al., 1996). However, extensive degradation following simulated gastric fluid digestion has been found in some food allergens (Fu et al., 2002; Díaz-Perales et al., 2003). Finally, the introduction of the transgene can cause a change in the expression of existing genes. The use of a representative panel of allergic patients' sera is the most adequate tool to address this possibility. The aim is first to check if there is an unexpected IgE bind-

ing to the newly expressed gene product and then, if the genetic variation affects or causes a quantitative or, more important, a qualitative difference in the accumulation of known or potential allergens. Each vegetable system contains a full array of potential allergenic molecules. Some of the genes coding for allergens can remain silent or under-expressed (Asero et al., 2005). A significant change in the gene expression, either in storage tissues or in other plant parts, such as pollen, and the interaction with a particular population will determine the allergenic impact of a GMO. Ideally, at least in the most relevant systems, main potential allergens might be identified, specific quantification methods might be developed and an accepted reference value and expression risk levels might also be defined. However, there may still be the possibility that a previously silent gene is now expressed.

Most of the known plant food allergens can be classified into a limited number of protein families (Breiteneder et al., 2004), and most are either storage or defense-related proteins. In the latter case, significant changes in accumulation occur under plant stress conditions, such as induced maturation, storage or by selection of resistant varieties.

In the characterization of a panel of sera to be used for screening new GMOs, two main aspects must be taken into account: the geographical origin of the allergic patients and the specific IgE profile of the serum sample.

Over the last years, we have characterized a wide panel of food and pollen allergens to which the Spanish population is sensitized. Spain has a wide climatic variation ranging from the Atlantic, to

Abbreviations: LTP, non-specific lipid-transfer protein; sIgE, specific IgE; GMO, genetically modified organisms.

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the Continental, Mediterranean and Subtropical climates. This contributes to a unique model for the study of inhaled allergen sensitization and the interaction with food allergies.

2. Materials and methods

2.1. Populations studied

Selected cases from the following studies were used. All subjects provided written informed consent and approval from Ethics Committees was obtained.

Patients' data were collected according to the daily practice of each clinical group. Each patient was identified by means of a numeric bar-coded label.

2.1.1. EXPO study

This study included 891 pollen-allergic patients in the olive pollen exposed area of Spain. Patients were selected consecutively over a period of 3 months out of the pollen season. All patients with a compatible clinical history of pollinosis, not having undergone previous immunotherapy and who had been living at least during the last five consecutive years in the same location, were included in the survey. In order to avoid any bias in patient selection, neither specific IgE nor skin prick tests were performed before inclusion of patients.

Serum samples were collected from the different subjects, identified by such bar-coded labels, stored at -40°C and thawed immediately before using.

2.1.2. RED Vegetalia

Four hundred and fifty patients with clinically proven vegetable food allergy, from nine different locations in Spain, with a parallel control sample of 450 patients allergic to pollen but not to food, were included.

2.1.3. Food allergy in the North of Spain

Thirty patients from the North of Spain (Bilbao and Navarra), who visited the Allergy Services due to potential food allergy further confirmed by clinical evaluation as described by Gamboa et al. (2007), were also considered.

2.1.4. Positive sIgE to fruits

A serum bank from allergic patients in Northern Europe was screened for Peach sIgE titers ($>10\text{ kU/l}$) by CAP (Phadia, Barcelona, Spain), following the manufacturer's instructions.

2.2. Specific IgE determination to single allergens

sIgE to the different allergens was tested on the ADVIA Centaur® platform (Bayer HealthCare Diagnostics Division, Tarrytown, NY, USA). The principle of the sIgE assay is based upon a reverse-sandwich assay. The platform is a continuous and fully automated system. Bar-coded controls and samples are loaded onto the apparatus and the system runs 120 tests per hour. The methodology has been previously described for whole allergen extracts (Petersen et al., 2004). Single allergens were biotinylated with NHS-LC-biotin (Pierce, Rockford, IL, USA) at a 7:1 (w/w) ratio following the manufacturer's instructions. Dose-response curves of biotinylated allergen were obtained and the optimal allergen dose per test was selected. This dose ranged from 10 to 40 ng of allergen per test. Biotinylated allergens were stored 50%-glycerinated at -20°C . Prior to the assay, allergens were diluted to the optimal dose and used. No stability problems during storage of allergens under these conditions were seen. The amount of sIgE, in kU/l, in the serum samples was determined by a two-point calibration system. A separate "reference" assay was simultaneously performed alongside the sIgE assay. The assay employs recombinant Bet v 1 as standard biotinylated allergen, a high calibrator which is a serum pool with a specified amount of Bet v 1 sIgE, traceable to the total IgE WHO reference preparation, and the low calibrator, which is an IgE-depleted serum sample. Reference reagents were obtained from ALK-Abelló A/S (Stenløse, Denmark).

The sample dose was calculated by applying the formula:

$$D = D_{\text{hical}} \times [(B - B_0) / (B_{\text{hical}} - B_{\text{local}})]$$

where D is the sample dose to a single allergen in kU/l, D_{hical} is the dose of high calibrator in kU/l, B is the sample response (relative light units) to the single allergen, B_0 is the low calibrator response to the single allergen, and B_{hical} and B_{local} are responses of high and low calibrators to Bet v 1.

All samples were run in duplicates and confirmed in triplicates when needed (CV above 12% for positive samples, and above 20% for negative samples). For reference samples, five replicates were run. The detection limit of the method was 0.1 kU/l. A cut-off value of 0.35 kU/l was used following the standard criterion.

Dilution recovery experiments were performed to evaluate the assay's dynamic range with the different single allergens and the degree of parallelism between them and the reference Bet v 1 allergen. A linear response was obtained with the different single allergens from doses as high as 400 kU/l. Parallel responses were obtained with the single allergens and Bet v 1, validating the use of the two-point calibration system for dose determination.

3. Results and discussion

3.1. Some examples of geographical variations in allergy

3.1.1. The Rosaceae case

Populations are exposed to a wide variety of potentially allergenic molecules. For instance, in Northern Europe, one of the most important inhalant allergies is caused by birch pollen. It is known that the main birch allergen, Bet v 1, is homologous with different proteins present in numerous vegetables. Thus, Bet v 1-related plant food allergy syndromes have been described and multiple allergens belonging to this group, PR10 (Pathogenesis Related proteins, group 10), have been characterized, the most representative being Mal d 1, purified from apple (Vieths et al., 2002; Fernández-Rivas et al., 2003, 2006). In Southern Europe, another group of allergenic molecules, LTPs (lipid-transfer proteins), has been described as major allergens (Salcedo et al., 2004, 2007; Fernández-Rivas et al., 2003, 2006). Through epidemiological studies, it has been demonstrated that whereas the North European population allergic to *Rosaceae* fruits is sensitized to Mal d 1, LTPs are the clinically relevant allergens in Spain. The reason for this difference is unknown, as a clear mediation by particular pollens to LTP allergy has not been demonstrated. In fact, LTP-mediated vegetable allergy prevails in all Spanish areas studied, regardless of the different climatic conditions (Red Vegetalia, unpublished). Within the Spanish population, LTP-allergic patients from Mediterranean areas seem to present a more complex profile than those living in continental regions (García-Sellés et al., 2002). LTPs display limited cross-reactivity. In general, sequence homology is only high ($>80\%$) within a particular taxonomic group (Salcedo et al., 2007). In Mediterranean populations, patients are exposed to different pollens containing LTPs (*Parietaria*, *Olea*, *Artemisia*, *Platanus*). Whether these pollen-linked LTPs play a role in the spreading of LTP allergic syndrome remains to be proven. A practical consequence, for the evaluation of GMOs for LTPs, is that sera from LTP-allergic patients in the Spanish Mediterranean areas will allow a better assessment of LTP allergenicity in a new vegetable system.

A third group of allergens involved in *Rosaceae* fruit allergy are profilins (Radauer et al., 2004). Profilins are panallergens present in both pollens and vegetables. After LTPs, profilins are the second most relevant allergens involved in plant food allergy in Spain. An interesting hypothesis is that the different pollens that are present in Spain, or the difference in the intensity of pollinosis, might account for the higher prevalence of profilin allergy in Spanish plant food-allergic patients. The significant association found between grass Group 5 allergen sIgE and profilin, especially in areas of high grass pollen exposure (Barber et al., unpublished) can support this.

3.1.2. Minor pollen allergens

The population allergic to olive pollen in heavily exposed areas represents an adequate group to study the food allergens linked to minor pollen allergens.

Minor allergens are so named when the prevalence of sensitization of the studied population is below 50%. However, significant differences in exposure might lead to different sensitization rates, and as several of the minor pollen allergens might be linked to food proteins, this might have a clear impact on some types of food allergy. Over the last years, we have characterized the most relevant olive (*Olea europaea*) pollen allergens (Rodríguez et al., 2001; Huecas et al., 2001; Barral et al., 2004). Among the minor allergens described, profilin (Ole e 2), β -glucanase (Ole e 9) and lipid-transfer protein (Ole e 7) are especially relevant, as they may be associated with food allergies in patients sensitized to them via inhalation.

In Spain, olive pollen is the second cause of pollen allergy. The levels of olive pollen have been reported to reach peaks of more than 14,000 pollen grains/m³ in the most exposed areas of Andalucía. The broad range of olive pollen exposure makes this pollinosis a particularly adequate model to study the prevalence of minor allergens and the clinical implications in pollen-allergic populations.

We have observed a gradient of prevalence of Ole e 7 and Ole e 9 reactivity ranging from less than 5% (in Madrid) to more than 50% (in Jaén) of olive pollen-allergic patients. Apart from the relevance of these data for allergen standardization (Duffort et al., 2006), there is a significant risk of food allergies linked to minor olive sensitization. Glucanases have been identified in multiple vegetable systems (Palomares et al., 2005), and as previously mentioned, LTPs are important food allergens. We have observed no cross-reactivity between Pru p 3 (LTP from peach) and Ole e 7, although we have found patients where Ole e 7 seems to be related to nut allergy, independently of Pru p 3.

3.2. Molecular profiles of sera

From the data previously presented, the importance of knowing the specific profile of allergens to which a particular patient is sensitized is evident. In order to evaluate the interaction of allergens with populations and the relationship between inhalant and food allergy, we developed a panel of 28 allergenic molecules (Table 1). This panel includes the most relevant pollen and food allergens that affect the Spanish population. Two epidemiological studies are currently being conducted. Nine clinical groups and four basic research groups are participating in the first one, called Red Vegetalia (Theme Networks of Cooperative Research, Spanish Health Ministry: Law SCO/709/2002). Sera from 900 patients allergic to either plant foods or pollens without concomitant food allergy have been screened against the panel of 28 allergens. In the second study called EXPO, sponsored by ALK-Abelló, 891 pollen-allergic patients included by 33 clinical groups from olive pollen exposed areas are being screened against a panel of 16 pollen allergens, including

several minor olive allergens and other relevant major pollen allergens.

The analytical platform used had a reverse configuration assay, where IgE is captured first and the allergen is added biotinylated. The main advantages of this technology are the lack of interference of IgG, the high dynamic range, the high specificity and, last but not least, the low amount of allergen required in the assay. Consequently, a combination of both recombinant and purified native allergens covers the main allergens involved in vegetable allergies (both inhaled and food) in our population. Some examples of the usefulness of this strategy to evaluate allergic sensitization are presented here.

3.2.1. Specific IgE responses without clinical relevance

A validated serum bank to screen a particular allergen source, such as peach, should include samples from patients with actual clinical symptoms to that food. Sera with high specific IgE levels to peach, obtained from North European subjects showing no clinical symptoms when eating this fruit, can illustrate the rationale for this requirement. The three main allergens of *Rosaceae* fruits belong to the Bet v 1 (PR-10 proteins), LTP and profilin families. Peach Pru p 1 (Bet v 1-like) seems to be present in low amounts and has not been reported as a relevant allergen (Ahrazem et al., 2007). Therefore, the above-mentioned sera were screened with rPru p 3 (peach LTP), rMal d 4 (apple profilin) and horseradish peroxidase, a potential marker of cross-reactive carbohydrate determinants (CCDs) (Altmann, 2007). The results are shown in Table 2. It is interesting to see that most of the IgE responses are directed to cross-reactive carbohydrate determinants that have been found to have uncertain clinical relevance. Despite of the fact that major peach allergens are unglycosylated molecules, other non-allergenic molecules which are glycoproteins cause unspecific positive results when testing sIgE to peach extract with sera from patients reactive to CCDs. This supports the need of using sera obtained from patients with a positive double-blind, placebo-controlled food challenge, but also supports the relevance of appropriate molecular screening to validate the sera before use in the evaluation of GMOs.

3.2.2. CCDs interference

As previously demonstrated, one of the most important confounding factors for the evaluation of allergenicity is the IgE directed against CCD epitopes. The abundance of such glycosylations in plant proteins leads to the frequent measurement of non-relevant IgE titers to different foods and pollens (Altmann, 2007). We have shown that Ole e 1 possesses a specific IgE-binding carbohydrate determinant (Batanero et al., 1994). This epitope binds IgE in practically all of the patients sensitized to Ole e 1 (Batanero et al., 1999). The technology configuration in this study is different from that of the most widely used specific IgE methodologies. Low-affinity interactions are then expected to have a lower impact in sIgE measurement (Fromberg, 2006).

Data reflecting serum profiles of different patients with high IgE titers to purified native Ole e 1 are shown in Table 3. The patients were tested with different purified natural allergens that are all glycoproteins, as well as with horseradish peroxidase (as specific CCD marker). Whereas some sera (less than 10% of the total sera analyzed) of the patients studied had low titers of IgE against peroxidase, (patients 1 and 4), no IgE binding directed to the other glycosylated allergens is seen in some sera (patient 3). This may be due to specific recognition of the different glycosylated determinants, since a great variability in CCDs structures is known to exist (Altmann, 2007). These data also suggest that it is possible to design more specific IgE assays, where the influence of low-affinity IgE antibodies will not hamper an allergenicity analysis of a new GMO. On the other hand, as shown in Table 2, there is also a strong

Table 1
Panel of allergens developed for molecular screening of sera from plant foods allergic patients

Pollens	
<i>Artemisia</i>	nArt v 1 Major allergen. Glycosylated
<i>Cupressus</i>	nCup s 1 Major allergen. Glycosylated
<i>Olea</i>	nOle e 1 Major allergen. Glycosylated rOle e 9 e. Minor allergen. Glucanase
<i>Parietaria</i>	nPar j 1. Major allergen. LTP family
<i>Gramineae</i>	nPhl p 1 Major allergen. Glycosylated nPhl p 5 Major allergen. Non-glycosylated nCyn d 1 Major allergen. Glycosylated (Phl p 1 related)
<i>Plantago</i>	nPla l 1 Major allergen Glycosylated. Ole e 1 family
<i>Chenopodium</i>	rChe a 1 Major allergen.. Ole e 1 family rChe a 3 Minor allergen. Calcium binding protein
<i>Salsola</i>	nSal k 1 Major allergen. Glycosylated
<i>Platanus</i>	rPla a 1; nPla a 2 Major allergens ^a
Plant foods	
Wheat flour:	nWTAI CM3 + nWTAI CM16 (α -amylase inhibitors)
Lentil:	nLen c 1 (vicilin)
Peach:	rPru p 3 (LTP)
Avocado/ chestnut:	nPrs a 1 + rCas s 5 (Class I chitinases)
Mustard:	nSin a 1 (2S Albumin)
Foods/pollens	
Profilin family	rChe a 2 + rMal d 4 (isoforms A + B) + rCuc m 2
PR-10 family	nBet v 1 + rMal d 1
Cross-reactive carbohydrate determinants	Horseradish peroxidase ^a

^a Commercial reagents; n = natural, r = recombinant.

Table 2

IgE molecular profile of sera from patients with high sIgE to peach but lacking clinical allergenic reactivity

Sample	IgE level		
	rMal d 4	Peroxidase	rPru p 3
1	–	5.23	–
2	–	8.28	–
3	–	9.78	–
4	–	14.80	–
5	–	14.75	–
6	4.11	2.31	–

–, sIgE < 0.35 kU/L.

Two major non-glycosylated *Rosaceae* fruit allergens, Mal d 4 (apple profilin) and Pru p 3 (peach lipid-transfer protein), and horseradish peroxidase as cross-reactive carbohydrate determinant (CCDs) marker were tested. Results are expressed in kU/L.

recognition of CCDs by a small percentage of patients. This might lead to new strategies to verify if CCD binding of high affinity IgE has a clinical significance.

One interesting hypothesis is that high affinity responses to CCDs, which are uncommon, might have clinical relevance, whereas low affinity titers, much more frequent, would lack of clinical significance.

3.2.3. Allergen profile of peach-allergic patients

As previously mentioned, there are marked geographical differences between peach allergic patients from different geographical locations. With the aim of finding out what happens in a transition region, we performed molecular diagnosis to *Rosaceae* fruit by screening sera from patients in the North of Spain, an area with moderate birch pollinosis. The patients were carefully selected by assessing the clinical symptoms and then performing a molecular diagnosis (Gamboa et al., 2007).

Some of the results are summarized in Table 4. As can be seen, completely different profiles are observed, with LTPs (Pru p 3) or the pair Bet v 1- homologues (Mal d 1)/profilins (Mal d 4) being the main sensitizers. Additionally, sera displaying sIgE to all three allergen types (i.e. patient 2) have been also found. Differences that were previously observed between Northern and Southern Europe are now detected in a single population, thus revealing the importance of this type of diagnosis.

Based on the tabulated results, it seems clear that a molecular screening, together with the clinical evaluation, offers the best information on sera characterization for subsequent use in GMO allergenicity assessment.

3.2.4. Assessment of pollen-allergic patients

Olive pollen-allergic patients constitute a relevant model to analyze specific IgE allergy profiles. It is clear that patients from an area heavily exposed to olive pollen might have different profiles from those in other less exposed areas.

While patients living in moderate pollen exposure areas are sensitized mostly to the major allergens (i.e. Ole e 1), an increasing number of patients sensitized to minor allergens are found when searching through regions with progressive higher exposure to olive pollen. Some of these minor allergens, such as Ole e 7 (LTP) and

Table 4

IgE profile to *Rosaceae* fruit allergens of allergic patients from a transition zone in the North of Spain

Sample	rBet v 1	rMal d 1	rMal d 4	rPru p 3	Clinical symptoms
1	–	–	–	12.71	U
2	–	5.35	19.08	16.18	U, GP
3	43.95	28.18	66.41	–	OAS
4	–	–	–	16.40	U, AG
6	–	–	–	6.61	U, AG
7	–	–	–	2.58	ANA
8	4.26	–	–	–	OAS
9	–	–	4.53	–	OAS

–, sIgE < 0.35 kU/L; U (urticaria), OAS (oral allergy syndrome), AG (angioedema), ANA (anaphylaxis), GP (gastric pain).

Results are expressed in kU/L.

Ole e 9 (β -glucanase) are potentially associated with a risk of sensitization to plant foods. Thus, if all potential allergens of GMOs are not assessed with adequate sera there is a risk that their massive introduction might cause severe problems in particular regions that had not been considered for patient's selection. The problem is that the patients from a highly exposed area show an extreme variability as well. This means that without careful screening of the patient's serum, the results obtained in a further analysis using these sera might vary greatly. A hypothetical GMO model containing four different allergen families- PR10 proteins (Bet v 1-like), profilins, LTPs and β -glucanases- could illustrate this point. PR10 allergic patients might be only found in Areas with birch tree allergy, profilin allergic subjects might be more prevalent in grass areas with high pollen counts, patients reacting to multiple LTPs might be found mostly in Mediterranean regions and finally, glucanase allergic patients in highly exposed olive pollen regions of Mediterranean countries. Panels of sera should be defined by selecting the adequate patients of the different locations.

Table 5 shows the molecular profiles of sera from olive pollen-allergic patients from a heavily exposed area in the South of Spain, screened against a panel of olive allergens. Ole e 1 is the major olive pollen allergen sensitizing more than 90% of olive allergic-patients. Three allergens are related to potential food allergens: LTP (Ole e 7), β -Glucanase (Ole e 9), and profilin (Ole e 2). There is also a pollen panallergen, polcalcin, (Ole e 3). Other relevant pollen and food allergens, affecting the allergic patients in the area were also included.

The results reflect the extreme variability in individual responses to allergens within a panel of patients having allergic symptoms induced by olive pollen. In further use of these sera for allergenicity assessment, the results will be dramatically influenced by the specific profiles.

It seems that this can be a general model for inhalant allergy, and that the level of allergen exposure will influence the average sensitization profile of the patient population. Considering this factor in the definition of serum to be included in a serum bank for allergenicity is a key issue.

In the extremely exposed areas, patients with very different sensitization profiles are detected. However, at lower pollen exposure levels, minor allergens scarcely sensitize the pollen-allergic

Table 3

IgE profile of patients with high IgE titers to nOle e 1 screened with a panel of major natural allergens known to be glycoproteins

Sample	nArt v1	nCup s 1	nCyn d 1	nOle e 1	Peroxidase	nPhl p 1	nPla l 1	nSal k 1
1	0.35	0.49	0.45	19.59	1.02	5.27	16.74	2.96
2	–	–	0.38	347.24	–	–	–	1.31
3	–	–	–	212.87	–	–	–	–
4	0.87	0.77	125.41	121.77	0.41	175.99	–	2.03

–, sIgE < 0.35 kU/L.

Results are expressed in kU/L.

Table 5

Natural variability in sIgE responses of a strongly exposed olive pollen population in the south of Spain

Sample	nOle e 1	nOle e 7	rOle e 9	Profilin rMal d 4	Polcalcin rChe a 3	rPru p 3	nPar j 1	nPhl p 1	nPhl p 5	nPla l 1	nArt v 1	nSal k 1
1	12.67	–	–	–	–	0.41	–	44.57	–	–	–	–
2	27.42	7.53	16.97	3.40	3.10	–	–	–	–	–	–	–
3	151.98	–	4.09	–	–	42.81	–	26.59	–	–	–	64.03
4	173.00	27.42	12.12	–	–	1.26	0.38	–	–	34.58	–	6.42
5	–	34.66	–	–	–	–	–	–	–	–	–	–
6	562.36	0.47	–	–	–	–	–	–	–	–	–	–
7	12.36	0.77	–	17.07	–	–	–	11.98	3.55	–	–	22.19
8	245.85	80.97	7.99	–	–	–	–	–	8.87	–	–	2.22

–, sIgE < 0.35 kU/l.

Results are expressed in kU/l. Mal d 4 and Che a 3 have been used as representative pan-allergens due to the high cross-reactivity among the different pan-allergens. Ledesma et al. (2006) and Barderas et al. (2004).

population, and patients are in most cases only reactive to major allergenic components (Barber et al., 2007).

4. Conclusions

Component-resolved allergy diagnosis is a new tool that is increasingly proving its value in the assessment of allergic diseases. It has been demonstrated that the technique can be used routinely by a pragmatic approach that combines the use of purified molecules from natural sources and recombinant allergens and a powerful analytical platform, with a high sensitivity and specificity.

In the current procedures for assessment of GMO allergenicity, specific allergy testing with an appropriate panel of sera is a requirement. Obviously, the proper number of sera comprising the panel can vary according to the transgene and GMO considered. The result of this testing involves a significant economical and social impact. Evidence is provided supporting the fact that serum molecular screening is a useful tool that is becoming increasingly affordable. In the last couple of months, we have participated in epidemiological studies where more than 2000 sera have been screened against panels of up to 28 allergenic components (more than 50,000 individual allergy tests). The results of these studies will be published in the upcoming months, but here we have pinpointed some examples that stress the relevance of this new approach for the generation of adequate serum banks. Allergy in general, and food allergy in particular, is increasingly perceived as a transversal disease, where the allergenic source is less important than the particular allergenic molecule recognized in the particular source, and where families of allergens present in different plant systems might lead to more complex allergy syndromes. At the same time, when an allergic patient is sensitized to food, there is a specific recognition of allergens that will play a significant role in the severity and evolution of the disease. Different clinical risks and cross-reactivities, and consequently different avoidance measures and clinical management, are also determined by these profiles. There are still many issues to be clarified, but it is increasingly evident that this approach represents a new way of understanding allergenic sensitization.

Conflict of interest statement

The authors declare that there are no conflicts of interest.

Acknowledgements

Financial support was provided by Fondo de Investigaciones Sanitarias, Ministerio de Salud y Consumo, Spain (Grant G03/094) and by ALK-Abelló.

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