

EDITORIAL

Peanut allergy—Individual molecules as a key to precision medicine

Peanut allergy is one of the most severe food allergies affecting millions of individuals worldwide. In German-speaking countries, peanut allergy is the most important cause of anaphylaxis in children and adolescents.¹ This Editorial highlights the impact of individual peanut allergens on the severity of symptoms and summarizes the allergens' biological characteristics and usefulness as tools for precision diagnostic tests. While peanuts are consumed cooked, roasted, or as peanut butter, roasting is the one processing method that considerably increases the allergenicity of peanuts (Figure 1).

Atopic children with impaired skin barrier functions may experience severe allergic reactions to peanuts without prior consumption illustrating the importance of the cutaneous route for sensitization.² Sensitization via the gastrointestinal tract, breastfeeding, or inhalation is also highly probable (Figure 1).³ Symptoms range from oral allergy syndrome, mostly related to pollen-associated peanut allergy, to severe dyspnoea, anaphylaxis, and sometimes death.⁴ If peanut allergy starts in early childhood, it will persist for life in the majority of cases. To date, no safe and efficacious causative treatment for peanut allergy has been authorized. However, epicutaneous immunotherapy with peanut extract is in a phase III trial.⁵ Currently, early exposure is intensely discussed as a preventive measure.⁶

During recent years, diagnostic tests have been improved considerably driven by molecular allergology research (Figure 1). Besides aqueous peanut extracts, six individual peanut allergens are already available for routine in vitro diagnostic tests in single- and multiplex formats (Figure 1).

The systematic allergen nomenclature provided by the WHO/IUIS Allergen Nomenclature Sub-Committee (<http://www.allergen.org>) is the only allergen nomenclature accepted by the World Health Organization (WHO) and the International Union of Immunological Societies (IUIS). At present (09/2018), 16 peanut allergens are officially recognized (Table 1). The designation Ara h 4 is no longer in use; it was renamed Ara h 3.02. The biology and immunology of individual peanut allergens was reviewed in detail recently.⁷

Ara h 1 and Ara h 3 are seed storage proteins. Both allergens possess two beta-barrel cupin domains and are members of the cupin superfamily and the cupin 1 protein family. The protein family database Pfam (<https://pfam.xfam.org/>) refers to protein superfamilies as clans, abbreviated CL. While Ara h 1 monomers aggregate to form trimers, known as vicilins, Ara h 3 monomers form hexamers, known as legumins. Cupin seed storage proteins serve as source of amino acids and energy during seed germination. In many plant



Protein Family	Allergen	Clinical Relevance	References
Profilin	Ara h 5	n/a	n/a
Bet v 1	Ara h 8*	Marker for pollen-associated peanut allergy. Dose-dependent anaphylactic reactions are rare. Lipid-binding seems to protect its structure from digestion, thus possibly increasing allergenicity.	Mittag, J Allergy Clin Immunol. 2004;114:1410 Glaumann, J Allergy Clin Immunol Pract. 2013;1:698 Petersen, Biol Chem. 2014; 395:239
Lipid transfer protein (LTP)	Ara h 9* (Ara h 16, 17)	Causes severe reactions in patients predominantly in the Mediterranean area.	Krause, J Allergy Clin Immunol. 2009;124:771
Defensin	Ara h 12 Ara h 13	Potential marker allergens for severe reactions. Defensins have antifungal activities.	Petersen, J Allergy Clin Immunol. 2015;136:1295
Oleosin	Ara h 10 Ara h 11 Ara h 14 Ara h 15	Marker allergens for severe reactions. Roasting increases allergenic potential, shown on a molecular basis.	Schwager, J Allergy Clin Immunol. 2017;140:1331
Cupin 1 and 2S albumin seed storage proteins	Ara h 1* Ara h 2* Ara h 3* Ara h 6* Ara h 7	Ara h 1, 2, 3, and 6: roasting increases allergenic potential, shown on a molecular basis. Ara h 2 is a marker for severe reactions, with 42.2 KU/L being a 95% predictive value. Ara h 2 and 6 were shown to be transferred into breast milk (sensitization route?).	Maleki, J Allergy Clin Immunol. 2003;112:190 Beyer, J Allergy Clin Immunol. 2001;107:1077 Beyer, Allergy 2015;70:90 Visser, Clin Exp Allergy 2011;41:1631 Schocker, Pediatr Allergy Immunol. 2016;27:348 Bernard, Allergy 2014;69:888

FIGURE 1 Synopsis of clinical relevance of individual peanut allergens regarding allergenicity, sensitization, and grade of symptom severity. *Allergens which are already available for component-resolved diagnostic testing; n/a, not available

TABLE 1 Peanut allergens—protein family membership and sequence accession numbers

Cupin (CL0029) Cupin 1 (PF00190)	WHO/IUIS	MW (kDa) ^a	Genbank nucleotide	Genbank protein/Uniprot	References
Ara h 1.0101 (=Ara h 1 clone 41B)	✓	68.75	L34402	AAB00861 P43238	Burks, J Clin Invest. 1995;96:1715
Ara h 1 clone P17	-	67.70	L38853	AAA60336 P43237	ibid.
Ara h 3.0101	✓	58.70	AF093541	AAC63045 O82580	Rabjohn, J Clin Invest. 1999;103:535
Ara h 3.0201 (formerly Ara h 4)	✓	58.85	AF086821	AAD47382 Q9SQH7	Kleber-Janke, Int Arch Allergy Immunol. 1999;119:265
iso-Ara h 3	-	56.24	AY618460	AAT39430	Boldt, Proteomics 2005;5:775
Prolamin (CL0482) Protease inhibitor/seed storage/LTP (PF00234)					
2S albumins					
Ara h 2.0101	✓	16.05	L77197	AAK96887	Stanley, Arch Biochem Biophys. 1997;342:244
Ara h 2.0201	✓	17.99	AY158467	AAN77576 Q6PSU2	Chatel, Int Arch Allergy Immunol. 2003;131:14
Ara h 6.0101	✓	14.85	AF092846	AAD56337 Q647G9	Kleber-Janke, Int Arch Allergy Immunol. 1999;119:265
Ara h 7.0101	✓	16.32	AF091737	AAD56719 Q9SQH1	ibid.
Ara h 7.0201	✓	17.37	EU046325	ABW17159 B4XID4	Schmidt, J Proteome Res. 2010;9:3701
Ara h 7.0301	✓	16.49	AY722691	AAU21496 Q647G8	Yan, Plant Sci. 2005;169:439
nsLTP					
Ara h 9.0101	✓	9.14	EU159429	ABX56711 B6CEX8	Krause, J Allergy Clin Immunol. 2009;124:771
Ara h 9.0201	✓	9.05	EU161278	ABX75045 B6CG41	ibid.
Ara h 16.0101	✓	7	n/a	n/a	Derksen, Allergy 2015;Suppl. 101:8
Ara h 17.0101	✓	9	n/a	n/a	ibid.
Profilin-like (CL0432) Profilin (PF00235)					
Ara h 5.0101	✓	13.91	AF059616	AAD55587 Q9SQI9	Kleber-Janke, Int Arch Allergy Immunol. 1999;119:265
iso-Ara h 5	-	13.97	GU354312	ADB96066	Cabanas, Protein Expr Purific. 2010;73:36
Bet v 1-like (CL0209) Bet v 1 (PF00407)					
Ara h 8.0101	✓	16.82	AY328088	AAQ91847 Q6VT83	Mittag, J Allergy Clin Immunol. 2004;114:1410
Ara h 8.0201	✓	16.28	EF436550	ABP97433 BOYIU5	Riecken, Biol Chem. 2008;389:415
No superfamily defined Oleosins (PF01277)					
Ara h 10.0101	✓	17.62	AY722694	AAU21499 Q647G5	Schwager, PLoS One 2015;10(4):e0123419
Ara h 10.0102	✓	15.40	AY722695	AAU21500 Q647G4	ibid.
Ara h 11.0101	✓	14.18	DQ097716	AAZ20276 Q45W87	ibid.

(Continues)

TABLE 1 (Continued)

Cupin (CL0029) Cupin 1 (PF00190)	WHO/IUIS	MW (kDa) ^a	Genbank nucleotide	Genbank protein/Uniprot	References
Ara h 11.0102	✓	14.22	DQ097717	AAZ20277 Q45W86	ibid.
Ara h 14.0101	✓	18.30	AF325917	AAK13449 Q9AXI1	Pons, Plant Physiol Biochem. 2005;43:659 Schwager, PLoS One 2015;10(4):e0123419
Ara h 14.0102	✓	18.33	AF325918	AAK13450 Q9AXIO	ibid.
Ara h 14.0103	✓	18.32	AY605694	AAT11925 Q6J1J8	Schwager, PLoS One 2015;10(4):e0123419
Ara h 15.0101	✓	16.74	AY722696	AAU21501 Q647G3	ibid.
Scorpion toxin-like knottin (CL0054) Plant defensin (PF00304)					
Ara h 12.0101	✓	8.04	EY396089	n/a	Petersen, J. Allergy Clin Immunol. 2015;136:1295
Ara h 13.0101	✓	8.22	EY396019	n/a	ibid.
Ara h 13.0102	✓	8.18	EE124955	n/a	ibid.

Definitions of protein superfamilies (=clans, CL) and protein families (PF) follow the Pfam—protein family database (<https://pfam.xfam.org/>) nomenclature.

MW, Molecular weights; n/a, not available.

^aMolecular weights were calculated for the mature but unprocessed proteins.

species, cupins are involved in the defense against fungi and insects. Cupin storage proteins play an important role as allergens for primary peanut allergy, particularly in children, who develop allergy at a young age.⁸ Individual allergens are already used as tools to elucidate the sensitization process.⁹

The prolamin superfamily comprises several protein families with limited sequence similarities, among them two families of low molecular weight proteins, the 2S albumins and the nonspecific lipid transfer proteins (nsLTPs). These are heavily disulfide-bonded resulting in the high stability of the allergenic peanut 2S albumins Ara h 2, 6, and 7. 2S albumins in seeds store amino acids, but they also possess antifungal and antibacterial properties. nsLTPs are divided into the 9 kDa nsLTP1 and the 7 kDa nsLTP2 subfamilies. Both nsLTP1s and nsLTP2s are found in seeds where they play important roles in resistance to biotic and abiotic stress, plant growth, and development. Ara h 9 and 17 are nsLTP1s, and Ara h 16 is an nsLTP2. Due to their high stability, allergens of the prolamin superfamily are associated with severe symptoms (Figure 1). Among the 2S albumins, Ara h 2 has a prominent role as marker allergen for the severity of the reaction, followed in significance by Ara h 6 (Figure 1).

Peanut is an oilseed crop, and its oil content is around 50%. Oleosins are the major oil body stabilizing proteins. Eight allergenic peanut oleosins have been identified and classified into four allergen groups, namely Ara h 10, 11, 14, and 15. All patients with severe peanut allergy have IgE to oleosins (Figure 1). This is in contrast to defensins, to which not all patients with severe allergic reactions have IgE antibodies. Roasting increases the allergenicity of oleosins which was also observed for Ara h 1, 2, and 6 (Figure 1).

Plant defensins are a family of peptides involved in the protection of seeds and reproductive tissues. Defensins are disulfide-rich antimicrobial peptides. Their activity is primarily directed against fungi but also against bacteria and insects. Membrane targeting via specific membrane phospholipids and membrane disruption are functions of many defensins. Ara h 12 and 13 are peanut defensins. They are not yet available for component-resolved diagnosis, but preliminary data indicate their high significance for patients with severe reactions to peanuts.

The profilin family comprises proteins that are present in all eukaryotic cells and that are highly conserved in higher plants. Profilins are involved in the turnover and restructuring of the actin cytoskeleton. Ara h 5, the peanut profilin, cross-reacts with many other plant profilins and, hence, can decrease the test specificity.

Proteins with the typical Bet v 1 architecture are found in all kingdoms of life. The Bet v 1-like superfamily comprises 17 families including the Bet v 1 family with its several subfamilies. Most of the Bet v 1-homologous allergens known belong to the PR-10 subfamily. Ara h 8, the Bet v 1 homolog from peanut, is linked to pollen-associated peanut allergy with mostly mild or moderate symptoms (Figure 1). As Ara h 8 associates with lipids via its hydrophobic cavity, thereby protecting the protein from degradation, it may also be responsible for severe allergic reactions.

Peanut-allergic patients react to different peanut allergens and, therefore, require individualized allergy diagnostic testing. Marker allergens will allow molecular phenotyping in the future. While improved component-resolved diagnosis will help to identify patients who will profit from specific immunotherapy, only a thorough understanding of the mechanistic processes of sensitization and symptom

elicitation based on studies using the same individual allergens will enable us to cure the disease.

ACKNOWLEDGMENTS

Author UJ gratefully acknowledges the funding by the German Research Foundation (DFG) grant JA-1007/2-1 and the German Center for Lung Research (DZL). Author HB acknowledges the support of the Austrian Science Fund (FWF) Doctoral Program MCCA W1248-B30.

CONFLICT OF INTEREST

Author UJ reports grants from the German Research Foundation, the German Center for Lung Research, and from Phadia ThermoFisher Scientific, and personal fees from the Fraunhofer Zukunftsstiftung. Author HB declares that he has no conflict of interest in relation to this manuscript.

Keywords

Allergen structure, component-resolved diagnostics, food processing, marker allergens, peanut allergens, sensitization routes

ORCID

Heimo Breiteneder  <http://orcid.org/0000-0003-2022-8689>

Uta Jappe^{1,2,3}

Heimo Breiteneder^{3,4} 

¹Division of Clinical and Molecular Allergology, Research Center Borstel, Airway Research Center North (ARCN), German Center for Lung Research, Borstel, Germany

²Interdisciplinary Outpatient Clinic, University of Lübeck, Lübeck, Germany

³WHO/IUIS Allergen Nomenclature Sub-Committee

⁴Institute of Pathophysiology and Allergy Research, Medical University of Vienna, Vienna, Austria

Correspondence

Uta Jappe, Division of Clinical and Molecular Allergology, Research Center Borstel, Borstel, Germany.
Email: ujappe@fz-borstel.de

and
Heimo Breiteneder, Institute of Pathophysiology and Allergy Research, Medical University of Vienna, Vienna, Austria.
Email: heimo.breiteneder@meduniwien.ac.at

REFERENCES

1. Worm M, Eckermann O, Dolle S, et al. Triggers and treatment of anaphylaxis: an analysis of 4,000 cases from Germany, Austria and Switzerland. *Dtsch Arztebl Int*. 2014;111:367-375.
2. Lack G, Fox D, Northstone K, Golding J, Avon Longitudinal Study of Parents and Children Study Team. Factors associated with the development of peanut allergy in childhood. *N Engl J Med*. 2003;348:977-985.
3. Trendelenburg V, Ahrens B, Wehrmann AK, Kalb B, Niggemann B, Beyer K. Peanut allergen in house dust of eating area and bed—a risk factor for peanut sensitization? *Allergy*. 2013;68:1460-1462.
4. Ahrens B, Niggemann B, Wahn U, Beyer K. Organ-specific symptoms during oral food challenge in children with food allergy. *J Allergy Clin Immunol*. 2012;130:549-551.
5. Wang J, Sampson HA. Safety and efficacy of epicutaneous immunotherapy for food allergy. *Pediatr Allergy Immunol*. 2018;29:341-349.
6. Hindley JP, Filep S, Block DS, King EM, Chapman MD. Dose of allergens in a peanut snack (Bamba) associated with prevention of peanut allergy. *J Allergy Clin Immunol*. 2018;141:780-782.
7. Palladino C, Breiteneder H. Peanut allergens. *Mol Immunol*. 2018;100:58-70.
8. Ballmer-Weber BK, Lidholm J, Fernandez-Rivas M, et al. IgE recognition patterns in peanut allergy are age dependent: perspectives of the EuroPrevall study. *Allergy*. 2015;70:391-407.
9. Palladino C, Narzt MS, Bublin M, et al. Peanut lipids display potential adjuvant activity by triggering a pro-inflammatory response in human keratinocytes. *Allergy*. 2018;73:1746-1749.