

Cross-reactivity of pollen allergens: recommendations for immunotherapy vaccines

Richard W. Weber

Purpose of review

This review will summarize recent research on pollen allergen and epitope cross-reactivity. Knowledge of these relationships aids in the rational formulation of allergen immunotherapy vaccines.

Recent findings

There has been further clarification of panallergens and their roles as both major and minor allergens. Recent studies have targeted non-specific lipid transfer proteins and calcium-binding proteins (polcalcins), as well as pathogenesis-related protein families and profilins. Polcalcins and non-specific lipid transfer proteins are responsible for pollen–fruit interactions as well as pollen cross-reactivity, in some cases, but not all, accounting for major allergenicity. Delineation of the enzymatic activity of certain allergens explains the ubiquitous nature of these pollen proteins.

Summary

Characterization of specific pollen allergens and their protein families has provided insight into the grounds for cross-reactivity. Continuing clarification of these relationships will allow the substitution and consolidation of inhalant extracts as described in the conclusion.

Keywords

cross-reactivity, immunotherapy, pollen allergen, vaccine

Introduction

The proper formulation of allergen vaccine immunotherapy for inhalant allergy is impacted by the relationships of pollen allergens, which become crucial at a time of diminishing availability of pollen extracts. Also, as optimal allergen doses become clarified, cross-reactivity will impact on formulation. Ignorance of cross-reacting allergens will increase the likelihood of adverse reactions as a result of inadvertent dosing with larger amounts of the same allergen. In the past few years, much more information has become available on pollen cross-reactivity and plant systematics, with amino acid sequencing, gene sequencing, and the cloning of recombinant allergens. Also with the functions of these allergenic proteins being clarified, and homologous proteins identified, it has been suggested that protein family content (molecular classification) rather than botanical taxonomy is a superior way to address cross-reactivity issues [1^{••}]. For the purposes of this review, however, it remains useful to discuss pollen cross-reactivity and the implications on allergen vaccine formulation in terms of botanical groupings. This review will emphasize reports published since mid-2004. Although much research has been published recently on pollen and plant–food interactions secondary to shared allergen families, as this topic is less germane to vaccine formulation, such articles will not be discussed here in great detail. Recently published reviews that provide additional background are by Weber [2,3^{••}] and Gadermaier and colleagues [4^{••}].

Conifers: gymnosperms

Cupressaceae is the most allergenic family in the order Coniferales, with the pine family, Pinaceae, being of little clinical relevance. The cypress family, including the subfamily Taxodioideae, contains cedar, juniper, cypress, bald cypress, redwood, sequoia, and Japanese red cedar. Members are strongly cross-reactive, with most studies demonstrating consistent cross-inhibition with both animal antisera and human IgE antibodies. There is strong homology demonstrated between the major (group 1) allergen of the various species, an approximately 40 000 M_r glycoprotein [2]. Whereas amino acid sequences show high homology among group 1 allergens, glycosylation sites may differ [5]. Midoro-Horiuti and co-workers [6] mapped two of three major linear IgE epitopes of Jun a 1 to a highly conserved region of a pectate lyase catalytic site. This enzyme is important during fertilization, and is highly conserved. A carbohy-

Curr Opin Allergy Clin Immunol 5:563–569. © 2005 Lippincott Williams & Wilkins.

National Jewish Medical and Research Center, University of Colorado Health Sciences Center, Denver, Colorado, USA

Correspondence to Richard W. Weber, MD, 1400 Jackson Street, Room J326, Denver, CO, USA
Tel: +1 303 398 1991; fax: +1 303 270 2189; e-mail: weberr@njc.org

Current Opinion in Allergy and Clinical Immunology 2005, 5:563–569

Abbreviations

nsLTP non-specific lipid transfer protein
RAST radioallergosorbent test

© 2005 Lippincott Williams & Wilkins
1528-4050

Table 1. Formulation suggestions for conifers and monocots.

Single <i>Juniperus</i> adequate for Cupressaceae
Treat relevant Pinaceae separately
One or two temperate grasses (Pooideae) adequate coverage – timothy (<i>Phleum pratense</i>) good choice; consider sweet vernal (<i>Anthoxanthum odoratum</i>) where prevalent
Bermuda (<i>Cynodon dactylis</i>) covers prairie grasses such as buffalo, salt, and grama
Despite some cross-reactivity with temperate grasses, treat Johnson (<i>Sorghum halepense</i>) and bahia (<i>Paspalum notatum</i>) grasses separately where relevant

drate epitope is found in high density in the pollen exine, and is found in unrelated pollens [7]. However, van Ree [8] has recently suggested that only a minority of sensitized patients recognizes the carbohydrate moiety as an IgE epitope. Cry j 2, from Japanese cedar, has been identified as a polymethylgalacturonase, a member of an enzyme family found in both pollen and fruits [9]. Tinghino and coworkers [10] reported that Jun o 4, from *Juniperus oxycedrus*, is a calcium-binding protein. Emphasizing the ubiquity of these proteins, immunoblotting studies revealed inhibition by family members and the unrelated pollens olive and ryegrass, with lesser inhibition by pellitory (see Table 1).

Earlier literature has demonstrated the interchangeability of Cupressaceae member extracts [2,3^{••}]. Therefore, a single family member, perhaps *Juniperus ashei*, would be adequate coverage in vaccine formulation. Pinaceae sensitivity, if relevant, needs to be treated separately.

Grasses: monocot angiosperms

Results from crude extract inhibition studies can be summarized as follows: Pooideae subfamily members (northern pasture grasses) strongly cross-react; Chloridoideae members, Bermuda, grama, and salt grass, are cross-inhibitory, with Bermuda being the most potent; Panicoideae members Johnson and Bahia grasses show a mixed pattern [3^{••}]. Ten grass allergen groups have now been characterized, and the review of these allergens by Andersson and Lidholm [11] is outstanding. Groups 1, 2/3, and 5, and probably groups 4 and 11 as well, are major allergens. Group 1 allergens are beta-expansins, papain-related cysteine proteinases [12]. Earlier work demonstrated homology within group 1 allergens, as well as homology to group 2/3 allergens. In addition, T-cell epitopes are shared between group 1 and group 5 allergens [2,3^{••}]. Meadow fescue Fes p 4 has been shown to cross-react with a kiwi fruit allergen [13]. Research by Smith and associates [14] helped to explain the lack of cross-reactivity between temperate and southern grasses. The authors found group 1 allergens in both the Pooideae and Panicoideae; however, they did not find group 5 allergens in other than the Pooideae subfamily, some epitopes were not even found outside the tribe Poeae.

Davies and colleagues [15[•]] examined ryegrass, canary, Bahia, and Bermuda grasses with IgE antibodies to Lol p 1 and Lol p 5, and demonstrated reactivity with the former, but not the latter two grasses. Timothy, canary, and ryegrass could inhibit ryegrass and Bahia; Bahia, Johnson, and Bermuda did not inhibit ryegrass.

Group 7 minor allergens are 2 EF hand calcium-binding proteins. Homologues are also seen in other pollens: Phl p 7, Cyn d 7, Bet v 4, Aln g 4, Ole e 3, and Bra n 1, with timothy allergen Phl p 7 showing the greatest degree of cross-reactivity [16,17]. Lol p 11 shows 32% homology with soybean trypsin inhibitor without the active enzyme site [18]. Despite no homology with other grass allergen groups, similarity was found with *Dactylis glomerata*, *Festuca rubra*, and *Phleum pratense*, with corn and tomato pollen proteins, and 44% homology with the major olive allergen, Ole e 1. Bermuda grass profilin, Cyn d 12, shares B-epitopes with sunflower profilin [19]. Niederberger and associates [20], using recombinant timothy Phl p 1, Phl p 2, Phl p 5, and birch profilin Bet v 2, found IgE to the four recombinant proteins accounted for a mean 59% of grass pollen-specific IgE. The lower inhibition of IgE binding to Bermuda grass and corn, *Zea mays*, was attributed to the lack of detectable groups 2 and 5 in these two species.

Other monocots show within-family cross-reactivity, but little to no cross-inhibition with grasses [2]. Asturias and coworkers [21[•]] characterized a date palm major allergen, Pho d 2, as a profilin. IgE cross-reactivity was shown with perennial ryegrass, olive, and annual mercury. See Table 1 for vaccine recommendations. Timothy has been shown to be a very adequate choice to represent the Pooideae subfamily members, and may possess some unique allergens on the basis of earlier work. The use of additional members may be appropriate for the optimal concentration of allergen groups. Consider sweet vernal where prevalent, because earlier work suggested some unique allergens; otherwise, June or perennial ryegrass are good choices. Southern grasses such as Bermuda, Bahia, and even Johnson grass, need to be treated separately. Bermuda covers certain prairie grasses well [2,3^{••}].

Trees and weeds: tricolpate angiosperms

The taxonomy of this large group has recently been reviewed [2,3^{••}]. Review of the earlier literature concerning cross-allergenicity within the superorder Caryophyllanae, especially between chenopod and amaranth weeds, shows almost complete allergenic identity among the *Amaranthus* species redroot pigweed and Palmer's amaranth, and among *Atriplex* scales. Other members of Chenopodioideae show greater diversity, and Russian thistle appears to possess significant unique allergens [3^{••}]. Lamb's quarter allergen Che a 1 has been characterized by Barderas and associates [22]. Despite 27–45%

Table 2. Formulation suggestions for amaranth family and rosidae class.

Single <i>Amaranthus</i> species adequate
Single <i>Atriplex</i> species adequate
Treat <i>Salsola</i> and <i>Kochia</i> separately
Lamb's quarter (<i>Chenopodium album</i>) cross-reactive with <i>Amaranthus</i> and <i>Atriplex</i> and may cover these adequately
Prevalent Betulaceae member adequate
Where oak (<i>Quercus</i>) common, may adequately cover birch (<i>Betula</i>)
Treat boxelder (<i>Acer negundo</i>) and other maples (<i>Acer</i>) separately
Treat pellitory (<i>Parietaria</i>) and nettle (<i>Urtica</i>) separately

identity with members of the Ole e 1-like protein family, there was no significant IgE cross-reactivity between Che a 1 and Ole e 1, and only low-level IgG cross-reactivity was demonstrated. The same researchers have now isolated and sequenced Che a 2, a profilin, and Che a 3, a polcalcin, which they believe are of greater relevance than Che a 1 [23^{••}]. There was strong cross-reactivity between corresponding allergens of olive (Ole e 2 and Ole e 3), and 65–82% identity with similar proteins from birch, alder, rapeseed, and timothy pollens. In addition, recombinant Che a 2 showed 95% cross-inhibition with latex, more than 80% with pollens, and variable inhibition up to 95% with plant-food profilins [24[•]].

See Table 2 for vaccine recommendations for Amaranthaceae family members. *Amaranthus* species can be represented by a single member, with the ubiquitous redroot pigweed a good choice. Any *Atriplex* species should be adequate. Because of strong cross-reactivity, Lamb's quarter could adequately cover both *Amaranthus* and *Atriplex*. Russian thistle and burning bush need to be treated separately.

Earlier enzyme-linked immunosorbent assay inhibition studies suggested that Russian olive, *Elaeagnus angustifolia*, in Elaeagnaceae, cross-reacts with members of Oleaceae [25]. Sastre and coworkers [26[•]] in Spain recently showed that *E. angustifolia* could not inhibit olive, *Olea europaea*, whereas the latter inhibited the former up to 40%. Crude extracts of pellitory and nettle, despite botanical proximity, do not cross-react. The major *Parietaria judaica* pollen allergens, Par j 1 and Par j 2, are non-specific lipid transfer proteins (nsLTPs), and exhibit significant sequence homology [27]. Another member of Urticaceae, *Holoptelea integrifolia*, although not containing a Par j 1 homologue, does show immunoblot cross-inhibition with pellitory [28[•]]. Asero and colleagues [29] were not able to not show cross-inhibition between profilins of pellitory, grass, or birch, whereas Asturias and associates [30[•]] did report cross-reactivity with recombinant profilin Par j 3.

A member of Euphorbiaceae is annual mercury, whose major allergen, Mer a 1, is a profilin [31]. Recombinant Mer a 1 shares B-epitopes with sunflower profilin, reacting with annual mercury, olive, and castor bean pollens. *Hevea brasiliensis* latex profilin is highly cross-reactive with ragweed, but not annual mercury profilin [32]. Immunoblot studies by Fuchs and colleagues [33] showed that ragweed, mugwort, and timothy extracts could inhibit IgE binding to latex allergens, but Bet v 1 and birch profilin Bet v 2 could not. Earlier studies utilizing skin tests, Prausnitz–Küstner neutralization, and passive hemagglutination inhibition [3^{••}] suggest strong cross-reactivity within Salicaceae, and moderate cross-reactivity between this family and Fagales members.

Earlier data showed high skin test correlations between shagbark hickory and pecan, and moderate correlations with these and black walnut [3^{••}]. Skin test correlations, radioallergosorbent tests (RASTs), Prausnitz–Küstner neutralization, and RAST inhibition support the cross-reactivity of the beech and birch families, Betulaceae and Fagaceae, respectively. RAST inhibition studies support some cross-reactivity between Fagales members and ash, elm, and willow [3^{••}].

Recombinant DNA studies of Betulaceae pollen allergens show strong homology between Bet v 1 and the major allergens of other members [34,35]. Bet v 1 belongs to the pathogenesis-related protein group-10 family (PR-10), and has been linked to a large number of foods and spices inducing food allergy [36[•],37^{••}]. Bet v 2, is a profilin, Bet v 3 and Bet v 4 are calcium-binding proteins [38,39]. Bet v 5 is an isoflavone reductase-related protein, with cross-reactivity to pear and lychee food allergens [40]. Bet v 2, Bet v 3, Bet v 4, and Bet v 5, are all minor allergens. Cross-reactivity between birch, mugwort, and celery has been demonstrated with Bet v 1, Bet v 2, and 46 000–60 000 M_r allergens [41]. Recombinant Aln g 2 cross-reacts with an oilseed rape allergen, whereas the polcalcin Aln g 4 shares IgE epitopes with allergens from various pollens [42,43]. Neudecker and coworkers [44^{••}] have demonstrated that structural similarity accounts for the strong IgE cross-reactivity between Bet v 4 and Phl p 7, despite differences in oligomerization. Hazel pollen Cor a 10 is an Hsp70 family chaperon luminal binding protein, showing 88–92% sequence identity with similar proteins from soy, tobacco, corn, and tomato [45].

See Table 2 for formulation suggestions. The prevalent Betulaceae member should be adequate to cover the family. Allergen characterization suggests that oak, in areas of predominance, may also cover birch family members.

Two oilseed rape pollen allergens are a calcium-binding protein homologous to Aln g 2 and a profilin [46]. Higher

Table 3. Formulation suggestions for olive and aster families.

Use olive (<i>Olea europaea</i>) where prevalent, ash (<i>Fraxinus</i>) otherwise
One or two major <i>Ambrosia</i> (short, giant, western, or false ragweed) adequate coverage
Single <i>Artemisia</i> adequate – <i>A. tridentata</i> in western USA, <i>A. vulgaris</i> in eastern USA and Europe
Other Asteraceae members need to be treated separately where relevant

molecular mass allergens are glycoproteins inhibiting timothy grass group 4 allergens. Oleaceae shows strong cross-reactivity between family members [3^{••}]. At least 10 olive allergens have been characterized: Ole e 2 is a profilin, Ole e 5 is a copper/zinc superoxide dismutase; Ole e 3 and Ole e 8 are calcium-binding proteins, 2 and 4 EF hand, respectively; and Ole e 9 and Ole e 10 have 1,3- β -glucanase activity [47,48^{••},49[•]]. Study of the calcium-binding motifs of Ole e 3 and Ole e 8 did not reveal any significant IgE cross-reactivity [47]. Ole e 9 and Ole e 10 have 53% identity, the latter sharing IgE B-cell epitopes with several pollen and vegetable proteins [49[•]]. Numerous ash allergens have been demonstrated by Niederberger and colleagues [50]: Fra e 1, the Ole e 1 homologue; a profilin homologue of Bet v 2; a 2 EF hand calcium-binding protein cross-reactive with Phl p 7; a 35 000 M_r allergen cross-reactive with birch, ragweed, and olive; and high molecular weight components.

See Table 3 for formulation details. Olive family members are strongly cross-reactive. Therefore, use European olive to cover in areas of prevalence, and an ash species where the latter are common.

The allergenic similarity of short, giant, western, and false ragweed crude extracts has been supported by a large variety of techniques over a great number of years [2]. Short ragweed allergens Amb a 1, 2, 3, 4, 5, 6, and 7 have been characterized: the most important is Amb a 1. Ninety to 95% of ragweed sufferers will react to Amb a 1 and Amb a 2; the other allergens are minor [3^{••}]. Amb a 1 has been found in short, giant, southern, and false ragweed, with minimal amounts in slender ragweed; and no Amb a 1 was found in cocklebur, marshelder, sages and mugwort, or plantain [51]. Lee and Dickinson [52] found decreasing amounts of Amb a 1 in short, western, southern, canyon, slender, giant, and false ragweeds, respectively, as well as in cosmos, but not in sunflower, narrowleaf marshelder, or spiny cocklebur.

Using enzyme-linked immunosorbent assay and immunoblot inhibitions, Katial and colleagues [53] found overlapping inhibition curves in nine *Artemisia* species, and virtually complete immunoblot inhibition. Brandys and coworkers [54] showed strong cross-reactivity between giant sagebrush, *Artemisia tridentata*, and five Eurasian

sages. Hirschwehr and associates [55] demonstrated that Art v 1 was shared by short ragweed, and there was cross-reactivity with Bet v 2. Structural analysis of Art v 1 showed it to be a 10 800 M_r defensin-like glycoprotein [56]. The molecule contains two unique O-glycans, one is felt to represent a potentially cross-reactive carbohydrate determinant [57[•]]. Art v 2 is a 35 000 M_r glycoprotein, and Art v 3 is a 13 000 M_r nsLTPs with cross-reactivity to peach allergen Pru p 3 [58[•]]. *Artemisia* and Rosaceae fruit major allergen cross-reactivity is accounted for by such nsLTPs [59]. The importance of nsLTP allergens in both food allergy and pollinosis has recently been reviewed [60[•],61^{••}]; Art v 4 is a profilin [62].

Helianthus annua pollen possesses 13 allergens, with two 24 000–25 000 M_r proteins reacting with 95–100% of patient sera [63]. RASTs and immunoblot inhibitions revealed the greatest cross-reactivity with mugwort, followed by oxeye daisy, dandelion, goldenrod, and short ragweed having the least. A strongly cross-reactive profilin has been identified as Hel a 2 [64].

See Table 3 for recommendations on formulation. The single most prevalent major ragweed (short, giant, western, or false) should be appropriate coverage. However, common usage is to utilize a mix, for example, short and giant ragweed. Minor ragweeds appear to be less potent, and should be covered by the appropriate major species. Other Aster family members need to be treated separately if important; cross-reactivity between ragweed and mugwort is not complete. Any *Artemisia* species is adequate: *A. tridentata* is appropriate in western north America, and *Artemisia vulgaris* is a good choice for eastern states and Europe.

Three allergens from London planetree have been identified: the major allergen Pla a 1 is a 18 000 M_r invertase inhibitor; Pla a 3 is a 10 000 M_r lipid transfer protein; and Pla a 2 is a 43 000 M_r polygalacturonase [65[•]]. These latter pectin-degrading enzymes are important allergens in grass pollen, and have been isolated as food allergens. Pla a 2 is reactive with 84% of planetree-sensitized patient sera, representing 52% of total IgE binding capacity of *Platanus acerifolia* extract. Cross-reactivity of IgG antibodies has been shown with tomato and Cupressaceae pectate lyases. Studies of *Platanus acerifolia* profilin showed that it cannot account for the cross-inhibition of planetree pollen extracts and food allergens [66].

Conclusion

Table 4 lists the scientific and common names of plants discussed in detail above. In addition, other pollen sources that may commonly be used in allergen vaccines are included.

Table 4. Selected pollen sources for allergen vaccines.

Scientific name	Common name(s)	Comments
Conifers		
<i>Juniperus ashei</i>	Mountain cedar	Single choice adequate
<i>Juniperus virginiana</i>	Eastern red cedar	
<i>Cryptomeria japonica</i>	Japanese (red) cedar	
<i>Pinus strobus</i>	White pine	
Grasses		
<i>Phleum pratense</i>	Timothy	Representative northern all pasture grasses (Pooideae) all strongly cross-reactive; timothy and sweet vernal may possess unique allergens
<i>Lolium perenne</i>	Perennial ryegrass	
<i>Poa pratensis</i>	Kentucky blue (June)	
<i>Anthoxanthum odoratum</i>	Sweet vernal	
<i>Dactylis glomerata</i>	Orchard (cock's foot)	
<i>Cynodon dactylon</i>	Bermuda	
<i>Sorghum halepense</i>	Johnson	
<i>Paspalum notatum</i>	Bahia	
Trees and weeds		
<i>Amaranthus retroflexus</i>	Redroot pigweed	Representative species
<i>Chenopodium album</i>	Lamb's quarter	Strongly cross-reactive
<i>Kochia scoparia</i>	Burning bush	Representative species
<i>Atriplex canescens</i>	Wingscale	
<i>Rumex acetosella</i>	Sheep sorrel	Presumed cross-reactive
<i>Populus deltoides</i>	Common cottonwood	
<i>Salix nigra</i>	Black willow	Strong cross-reactivity among group 1 allergens
<i>Mercurialis annua</i>	Annual mercury	
<i>Acacia dealbata</i>	Mimosa	
<i>Prosopis glanulosa</i>	Mesquite	
<i>Quercus alba</i>	White oak	Moderate cross-reactivity
<i>Betula verrucosa</i>	White birch	
<i>Alnus glutinosa</i>	European alder	Strong cross-reactivity among group 1 allergens
<i>Corylus avellana</i>	Hazel	
<i>Carya illinoensis</i>	Pecan	
<i>Juglans nigra</i>	Black walnut	
<i>Acer rubrum</i>	Red maple	Major four ragweeds strongly cross-reactive
<i>Acer negundo</i>	Box elder	
<i>Olea europaea</i>	Olive	
<i>Fraxinus excelsior</i>	European ash	
<i>Plantago lanceolata</i>	English plantain	Non-Ambrosia Aster family members treated separately
<i>Ambrosia artemisiifolia</i>	Short ragweed	
<i>Ambrosia trifida</i>	Giant ragweed	
<i>Iva xanthifolia</i>	Burweed marshelder	
<i>Xanthium communis</i>	Cocklebur	Single species adequate
<i>Artemisia vulgaris</i>	Mugwort	
<i>Artemisia tridentata</i>	Giant sagebrush	
<i>Platanus acerifolia</i>	London planetree	

Cupressaceae members are markedly cross-reactive; in practice, a single *Juniperus* pollen extract should be suitable for immunotherapy. Members of the Pinaceae should be treated separately in the unusual case of clinical importance. There is no significant cross-reactivity between conifers and flowering plants; the minor reactivity seen, caused by panallergens such as profilins, is not clinically important.

Members of the temperate pasture grass subfamily Pooideae are strongly cross-reactive based on the marked homology of groups 1, 2/3, 4, and 5. Inhibition differences reflect quantitative rather than qualitative differences. One or two northern pasture grasses can achieve adequate coverage; there is no need for multiple species in either testing or immunotherapy. Panicoideae and Chloridoideae subfamilies have greater diversity, and the lack of group 2 and group 5 allergens accounts for differences with northern pasture grasses. Therefore, members of

these tribes should be tested for and treated separately. Chloridoideae members are cross-reactive, with Bermuda grass being most potent, and the appropriate choice to cover other members. The cross-reacting profilins and calcium-binding proteins are minor allergens of grasses, of little practical consequence in vaccine formulation. Grass allies, such as palm or sedge, if felt to be clinically relevant, need to be treated separately.

Diversity is the rule among tricolpate angiosperms, and the lack of cross-reactivity is the rule down even to the level of tribe. There are exceptions, with significant cross-reactivity across family boundaries. In Amaranthaceae weeds, *Amaranthus* and *Atriplex* species can be represented by single members, but the other locally relevant members need to be addressed separately. The order Fagales shows strong cross-allergenicity within the birch family and extending to the beech family. Use of the locally prevalent Betulaceae member should also

cover other family members. In areas where oaks are predominant, *Quercus* would be expected to cover birch. With other members of the Rosidae subclass, generally, one member of a family can be expected to be adequate for immunotherapy, although there are exceptions such as nettle and pellitory.

Oleaceae members are strongly cross-reactive: in areas where European olive is grown, it seems the proper choice, whereas in areas where ash is prevalent, it should be adequate.

Asteraceae members show variable cross-reactivity. The four major ragweeds (short, giant, western, and false) strongly cross-react, and one or two are quite adequate. In areas where the sages or marshelders are common, these need to be handled separately. *Artemisia* species are strongly cross-allergenic; it is not necessary to skin test or treat with multiple members. In the midwest and west USA, choose common sagebrush, *A. tridentata*, while in the eastern states and Europe, mugwort, *A. vulgaris*, seems most appropriate.

References and recommended reading

Papers of particular interest, published within the annual period of review, have been highlighted as:

- of special interest
- of outstanding interest

Additional references related to this topic can also be found in the Current World Literature section in this issue (p. 579).

- 1 Mothes N, Horak F, Valenta R. Transition from a botanical to a molecular classification in tree pollen allergy: implications for diagnosis and therapy. *Int Arch Allergy Immunol* 2004; 135:357–373.
- An important discussion of the value of characterized allergens as markers in diagnosis, and ultimately, treatment. Represents a paradigm shift away from classic plant systematics.
- 2 Weber RW. Patterns of pollen cross-allergenicity. *J Allergy Clin Immunol* 2003; 112:229–239.
- 3 Weber RW. Cross-reactivity of pollen allergens. *Curr Allergy Asthma Rep* 2004; 4:401–408.
- This is an up to date broader review of pollen allergens. There is a tabulation of grass pollen allergen groups and a cladogram of the grass family, clarifying relationships.
- 4 Gadermaier G, Dedic A, Obermeyer G, *et al.* Biology of weed pollen allergens. *Curr Allergy Asthma Rep* 2004; 4:391–400.
- Provides a very nice discussion of weed pollen allergens and a good up to date review. There is an excellent summary table delineating aspects of characterized weed allergens.
- 5 Midoro-Horiuti T, Goldblum RM, Kurosky A, *et al.* Molecular cloning of the mountain cedar (*Juniperus ashei*) pollen major allergen, Jun a 1. *J Allergy Clin Immunol* 1999; 104:613–617.
- 6 Midoro-Horiuti T, Mathura V, Schein CH, *et al.* Major linear IgE epitopes of mountain cedar pollen allergen Jun a 1 map to the pectate lyase catalytic site. *Mol Immunol* 2003; 40:555–562.
- 7 Canini A, Giovannazzi J, Iacovacci P, *et al.* Localisation of a carbohydrate epitope recognised by human IgE in pollen of Cupressaceae. *J Plant Res* 2004; 11:147–153.
- 8 van Ree R. Carbohydrate epitopes and their relevance for the diagnosis and treatment of allergic diseases. *Int Arch Allergy Immunol* 2002; 129:189–197.
- 9 Ohtsuki T, Taniguchi Y, Kohno K, *et al.* Cry j 2, a major allergen of Japanese cedar pollen, shows polymethylgalacturonase activity. *Allergy* 1995; 50:90–93.
- 10 Tinghino R, Barletta B, Palumbo S, *et al.* Molecular characterization of a cross-reactive *Juniperus oxycedrus* pollen allergen, Jun o 2: a novel calcium-binding allergen. *J Allergy Clin Immunol* 1998; 101:772–777.
- 11 Andersson K, Lidholm J. Characteristics and immunobiology of grass pollen allergens. *Int Arch Allergy Immunol* 2003; 130:87–107.
- 12 Grobe K, Becker W-M, Schlaak M, Petersen A. Grass group I allergens (beta-expansins) are novel, papain-related proteinases. *Eur J Biochem* 1999; 263:33–40.
- 13 Gavrovic-Jankulovic M, Cirkovic T, Burazer L, *et al.* IgE cross-reactivity between meadow fescue pollen and kiwi fruit in patients' sera with sensitivity to both extracts. *J Invest Allergol Clin Immunol* 2002; 12:279–286.
- 14 Smith PM, Ong EK, Knox RB, Singh MB. Immunological relationships among group I and group V allergens from grass pollens. *Mol Immunol* 1994; 31:491–498.
- 15 Davies JM, Bright ML, Rolland JM, O'Hehir RE. Bahia grass pollen specific IgE • is common in seasonal rhinitis patients but has limited cross-reactivity with ryegrass. *Allergy* 2005; 60:251–255.
- Little attention has been paid to Panicum grasses, and this paper offers an early insight into this group.
- 16 Tinghino R, Twardosz A, Barletta B, *et al.* Molecular, structural, and immunologic relationships between different families of recombinant calcium-binding pollen allergens. *J Allergy Clin Immunol* 2002; 109:314–320.
- 17 Smith PM, Xu H, Swoboda I, Singh MB. Identification of a Ca²⁺ binding protein as a new Bermuda grass pollen allergen Cyn d 7: IgE cross-reactivity with oilseed rape allergen Bra r 1. *Int Arch Allergy Immunol* 1997; 114:265–271.
- 18 van Ree R, Hoffman DR, van Dijk W, *et al.* Lol p XI, a new major grass pollen allergen, is a member of a family of soybean trypsin inhibitor-related proteins. *J Allergy Clin Immunol* 1995; 95:970–978.
- 19 Asturias JA, Arilla MC, Gomez-Bayon N, *et al.* Cloning and high level expression of Cynodon dactylon (Bermuda grass) pollen profilin (Cyn d 12) in *Escherichia coli*: purification and characterization of the allergen. *Clin Exp Allergy* 1997; 27:1307–1313.
- 20 Niederberger V, Laffer S, Froschl R, *et al.* IgE antibodies to recombinant pollen allergens (Phl p 1, Phl p 2, Phl p 5, and Bet v 2) account for a high percentage of grass pollen-specific IgE. *J Allergy Clin Immunol* 1998; 101:258–264.
- 21 Asturias JA, Ibarrola I, Fernández J, *et al.* Pho d 2, a major allergen from date palm, is a profiling: cloning, sequencing, and immunoglobulin E cross-reactivity with other pollens. *Clin Exp Allergy* 2005; 35:374–381.
- An excellent characterization of a major allergen from a non-grass monocot – little work has been published in this arena.
- 22 Barderas R, Villalba M, Lombardero M, Rodriguez R. Identification and characterization of Che a 1 allergen from *Chenopodium album* pollen. *Int Arch Allergy Immunol* 2002; 127:47–54.
- 23 Barderas R, Villalba M, Pascual CY, *et al.* Profilin (Che a 2) and polcalcin (Che a 3) are relevant allergens of *Chenopodium album* pollen: isolation, amino acid sequences, and immunologic properties. *J Allergy Clin Immunol* 2004; 113:1192–1198.
- Earlier work has suggested a high degree of cross-reactivity of lamb's quarter with other family members. This work may help explain the nature of that relationship.
- 24 Barderas R, Villalba M, Rodriguez R. Recombinant expression, purification • and cross-reactivity of chenopod profilin: rChe a 2 as a good marker for profilin sensitization. *Biol Chem* 2004; 385:731–737.
- Profilins were described as the earliest panallergen, this report characterizes an important member of that group of allergens.
- 25 Kernerman SM, McCullough J, Green J, Ownby DR. Evidence of cross-reactivity between olive, ash, privet, and Russian olive tree pollen allergens. *Ann Allergy* 1992; 69:493–496.
- 26 Sastre J, Lluch-Bernal M, Bustillo AMG, *et al.* Allergenicity and cross-reactivity • of Russian olive pollen (*Eleagnus angustifolia*). *Allergy* 2004; 59:1181–1186.
- This report follows up on earlier work on a puzzling cross-reactivity.
- 27 Stumvoll S, Westritschnig K, Lidholm J, *et al.* Identification of cross-reactive and genuine *Parietaria judaica* pollen allergens. *J Allergy Clin Immunol* 2003; 111:974–979.
- 28 Sharma S, Panzani RC, Gaur SN, *et al.* Evaluation of cross-reactivity between • *Holoptelea integrifolia* and *Parietaria judaica*. *Int Arch Allergy Immunol* 2005; 136:103–112.
- This is a recent report of relationships between botanically related but geographically disparate plants.
- 29 Asero R, Mistrello G, Roncarolo D, Amato S. *Parietaria* profilin shows only limited cross-reactivity with birch and grass profilins. *Int Arch Allergy Immunol* 2003; 133:121–124.
- 30 Asturias JA, Ibarrola I, Euseverri JL, *et al.* PCR-based cloning and immunological characterization of *Parietaria judaica* pollen profilin. *J Invest Allergol Clin Immunol* 2004; 14:43–48.
- Further work on allergens from a very important inducer of pollinosis.

- 31 Vallverdú A, Asturias JA, Arilla C, *et al.* Characterization of recombinant *Mercurialis annua* major allergen Mer a 1 (profilin). *J Allergy Clin Immunol* 1998; 101:363–370.
- 32 Vallier P, Ballard S, Harf R, *et al.* Identification of profilin as an IgE-binding component in latex from *Hevea brasiliensis*: clinical implications. *Clin Exp Allergy* 1995; 25:332–339.
- 33 Fuchs T, Spitzauer S, Vente C, *et al.* Natural latex, grass pollen, and weed pollen share IgE epitopes. *J Allergy Clin Immunol* 1997; 100:356–364.
- 34 Valenta R, Breiteneder H, Pettenberger K, *et al.* Homology of the major birch-pollen allergen, Bet v I, with the major pollen allergens of alder, hazel, and hornbeam at the nucleic acid level as determined by cross-hybridization. *J Allergy Clin Immunol* 1991; 87:677–682.
- 35 Niederberger V, Pauli G, Gronlund H, *et al.* Recombinant birch pollen allergens (rBet v 1 and rBet v 2) contain most of the IgE epitopes present in birch, alder, hornbeam, hazel, and oak pollen: A quantitative IgE inhibition study with sera from different populations. *J Allergy Clin Immunol* 1998; 102:579–591.
- 36 Mittag D, Vieths S, Vogel L, *et al.* Soybean allergy in patients allergic to birch pollen: clinical investigation and molecular characterization of allergens. *J Allergy Clin Immunol* 2004; 113:148–154.
- Another example of cross-reactivity between pollen and foods because of yet another shared allergen.
- 37 Jahn-Schmid B, Radakovics A, Lüttkopf D, *et al.* Bet v 1_{142–156} is the dominant T-cell epitope of the major birch pollen allergen and important for cross-reactivity with Bet v 1-related food allergens. *J Allergy Clin Immunol* 2005; 116:213–219.
- The characterization of the dominant T-cell epitope on a very important and ubiquitous allergen. This work carries immense potential for tailoring vaccine immunotherapy in the future.
- 38 Seiberler S, Scheiner O, Kraft D, *et al.* Characterization of a birch pollen allergen, Bet v III, representing a novel class of Ca²⁺ binding proteins: specific expression in mature pollen and dependence of patients' IgE binding on protein-bound Ca²⁺. *EMBO J* 1994; 13:3481–3486.
- 39 Engel E, Richter K, Obermeyer G, *et al.* Immunological and biological properties of Bet v 4, a novel birch pollen allergen with two EF-hand calcium-binding domains. *J Biol Chem* 1997; 272:28630–28637.
- 40 Karamloo F, Schmitz N, Scheuer S, *et al.* Molecular cloning and characterization of a birch pollen minor allergen, Bet v 5, belonging to a family of isoflavone reductase-related proteins. *J Allergy Clin Immunol* 1999; 104:991–999.
- 41 Bauer L, Ebner C, Hirschwehr R, *et al.* IgE cross-reactivity between birch pollen, mugwort pollen and celery is due to at least three distinct cross-reacting allergens: immunoblot investigation of the birch–mugwort–celery syndrome. *Clin Exp Allergy* 1996; 26:1161–1170.
- 42 Focke M, Hemmer W, Hayek B, *et al.* Identification of allergens in oilseed rape (*Brassica napus*) pollen. *Int Arch Allergy Immunol* 1998; 117:105–112.
- 43 Hayek B, Vangelista L, Pastore A, *et al.* Molecular and immunologic characterization of a highly cross-reactive two EF-hand calcium-binding alder pollen allergen, Aln g 4: structural basis for calcium-modulated IgE recognition. *J Immunol* 1998; 161:7031–7039.
- 44 Neudecker P, Nerkamp J, Eisenmann A, *et al.* Solution structure, dynamics, and hydrodynamics of the calcium-bound cross-reactive birch pollen allergen Bet v 4 reveal a canonical monomeric two EF-hand assembly with a regulatory function. *J Mol Biol* 2004; 336:1141–1157.
- A well-performed work not only on the primary function of polcalcins, but an investigation of the similarities across botanical borders despite structural differences.
- 45 Gruehn S, Suphioglu C, O'Hehir R, Volkmann D. Molecular cloning and characterization of hazel pollen protein (70 kD) as a luminal binding protein (BiP): a novel cross-reactive plant allergen. *Int Arch Allergy Immunol* 2003; 131:91–100.
- 46 Welch J, Jones MG, Cullinan P, *et al.* Sensitization to oilseed rape is not due to cross-reactivity with grass pollen. *Clin Exp Allergy* 2000; 30:370–375.
- 47 Ledesma A, González E, Pascual CY, *et al.* Are Ca²⁺-binding motifs involved in the immunoglobulin E-binding of allergens? Olive pollen allergens as model of study. *Clin Exp Allergy* 2002; 32:1476–1483.
- 48 Butteroni C, Afferni C, Barletta B, *et al.* Cloning and expression of the *Olea europaea* allergen Ole e 5, the pollen Cu/Zn superoxide dismutase. *Int Arch Allergy Immunol* 2005; 137:9–17.
- The first description of a copper/zinc superoxide dismutase as an allergen (manganese superoxide dismutase has been identified as an allergen). Although still considered a minor allergen, over a third of olive-sensitive patients related to this allergen.
- 49 Barral P, Batanero E, Palomares O, *et al.* A major allergen from pollen defines a novel family of plant proteins and shows intra- and interspecies cross-reactivity. *J Immunol* 2004; 172:3644–3651.
- The description and characterization of yet another important olive pollen allergen, with far-reaching cross-reactivity potential.
- 50 Niederberger V, Purohit A, Oster JP, *et al.* The allergen profile of ash (*Fraxinus excelsior*) pollen: cross-reactivity with allergens from various plant species. *Clin Exp Allergy* 2002; 32:933–941.
- 51 Yunginger JW, Gleich GJ. Measurement of ragweed antigen E by double antibody radioimmunoassay. *J Allergy Clin Immunol* 1972; 50:326–337.
- 52 Lee YS, Dickinson DB. Characterization of pollen antigens from Ambrosia L. (Compositae) and related taxa by immunoelectrophoresis and radial immunodiffusion. *Am J Bot* 1979; 66:245–262.
- 53 Katial RK, Lin FL, Stafford WW, *et al.* Mugwort and sage (*Artemisia*) pollen cross-reactivity: ELISA inhibition and immunoblot evaluation. *Ann Allergy Asthma Immunol* 1997; 79:340–346.
- 54 Brandys J, Grimsøen A, Nilsen BM, *et al.* Cross-reactivity between pollen extracts from six *Artemisia* species. *Planta Med* 1993; 59:221–228.
- 55 Hirschwehr R, Heppner C, Spitzauer S, *et al.* Identification of common allergenic structures in mugwort and ragweed pollen. *J Allergy Clin Immunol* 1998; 101:196–206.
- 56 Himly M, Jahn-Schmid B, Dedic A, *et al.* Art v 1, the major allergen of mugwort pollen, is a modular glycoprotein with a defensin-like and a hydroxyproline-rich domain. *FASEB J* 2002; 17:106–108; E-pub: 15 November 2002.
- 57 Leonard R, Petersen BO, Himly M, *et al.* Two novel types of O-glycans on the mugwort pollen allergen Art v 1 and their role in antibody binding. *J Biol Chem* 2005; 9:7932–7940.
- The further characterization of some unique properties of a major mugwort allergen.
- 58 Lombardero M, Garcia-Sellés FJ, Polo F, *et al.* Prevalence of sensitization to *Artemisia* allergens Art v 1, Art v 3 and Art v 60 kDa. Cross-reactivity among Art v 3 and other relevant lipid-transfer protein allergens. *Clin Exp Allergy* 2004; 34:1415–1421.
- Well conducted research especially on the identification of Art v 3 as a nsLTP; again, a central actor in pollen–food cross-reactivity.
- 59 Garcia-Sellés FJ, Diaz-Perales A, Sánchez-Monge R, *et al.* Patterns of reactivity to lipid transfer proteins of plant foods and *Artemisia* pollen: an in vivo study. *Int Arch Allergy Immunol* 2002; 128:115–122.
- 60 Salcedo G, Sanchez-Monge R, Diaz-Perales A, *et al.* Plant non-specific lipid transfer proteins as food and pollen allergens. *Clin Exp Allergy* 2004; 34:1336–1341.
- A useful review of an important class of ubiquitous panallergens, and the role of geographical differences.
- 61 Breiteneder H, Mills C. Nonspecific lipid-transfer proteins in plant foods and pollens: an important allergen class. *Curr Opin Allergy Clin Immunol* 2005; 5:275–279.
- An up to date discussion of the importance of this class of panallergens in both pollen and food sensitivity.
- 62 Wopfner N, Willeroidee M, Hebenstreit D, *et al.* Molecular and immunological characterization of profilin from mugwort pollen. *Biol Chem* 2002; 383:1779–1789.
- 63 Fernandez C, Martin-Esteban M, Fiandor A, *et al.* Analysis of cross-reactivity between sunflower pollen and other pollens of the Composite family. *J Allergy Clin Immunol* 1993; 92:660–667.
- 64 Asturias JA, Arilla MC, Gomez-Bayon N, *et al.* Cloning and immunological characterization of the allergen Hel a 2 (profilin) from sunflower pollen. *Mol Immunol* 1998; 35:469–478.
- 65 Ibarrola I, Arilla MC, Martinez A, Asturias JA. Identification of a polygalacturonase as a major allergen (Pla a 2) from *Platanus acerifolia* pollen. *J Allergy Clin Immunol* 2004; 113:1185–1191.
- The investigation of a major allergen from an aeroallergen source that is recently receiving more attention. Characterization again shows the ubiquitous nature of some of these proteins in nature.
- 66 Enrique E, Alonso R, Bartolome B, *et al.* IgE reactivity to profilin in platanus acerifolia pollen-sensitized subjects with plant-derived food allergy. *J Invest Allergol Clin Immunol* 2004; 14:335–342.