Allergens II: Cross-reactivity Classes
What to test

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AAOA Basic Course — 2013
Disclosures

Sunovion- Advisory Board, Speaker
Teva- Advisory Board
Objectives

Participants will be able to:

1. list most clinically important occurrences of cross-reactivity

2. divide the most clinically relevant inhalant allergens into classes based on biologic origin

3. apply knowledge of allergens in formulating a rational testing strategy in their region.
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Cross-Reactivity: Occurrences

- Can occur between inhalants, foods, and between foods and inhalants
- Cross-reactivity increases with closeness of biologic relationship: Order → family → tribe → genus → species
  - Cross-reactivity between closely related species is common
  - Distantly related species occasionally cross-react, due to conserved antigens

Degrees of separation. In the above comparison of proteins from apples (left) and celery (right) to that from birch pollen, red areas show where surface structure has been conserved across the proteins. Given these two proteins' relative structural similarity to that of birch pollen, people allergic to birch pollen are more likely to also be allergic to apples than to celery. image: John Jenkins and Clare Mills, JACI, 2005
Oral Allergy Syndrome

- Oral allergic symptoms in response to eating certain (usually fresh) fruit, vegetables, nuts
- Due to cross-reactivity between inhalant and food allergens
- Clinical implications:
  - Concomitant reactions during pollen/food season
  - (+) inhalant allergies suggest foods to be wary of

<table>
<thead>
<tr>
<th>Inhalant allergy</th>
<th>Food allergy</th>
</tr>
</thead>
<tbody>
<tr>
<td>Birch pollen</td>
<td>hazelnut, apple, pear, peach, plum, cherry, strawberry, carrot, orange, persimmon, zucchini</td>
</tr>
<tr>
<td>Ragweed pollen</td>
<td>Melon, banana</td>
</tr>
<tr>
<td>Mugwort pollen</td>
<td>Celery, coriander</td>
</tr>
<tr>
<td>Grass pollen</td>
<td>Tomato, peanut, pea, wheat, rye, apple, carrot</td>
</tr>
<tr>
<td>Latex</td>
<td>Banana, chestnut, kiwi, avocado</td>
</tr>
<tr>
<td>Chironomids (midges)</td>
<td>Crustaceans</td>
</tr>
</tbody>
</table>

N. Eriksson et al (1947)
Some knowledge of cross-reactivity is necessary for skin testing, and for immunotherapy

- Improves safety
- Test efficiency improved by eliminating redundant allergens
- Helps predict additional sensitivities
## Cross-reactivity: Clinical Utility

### Inhalants

<table>
<thead>
<tr>
<th>Item</th>
<th>Rating</th>
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<tbody>
<tr>
<td>Grasses</td>
<td>++++++</td>
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<tr>
<td>Weeds</td>
<td>++++</td>
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<tr>
<td>Trees</td>
<td>+++</td>
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<tr>
<td>Insects</td>
<td>++</td>
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<tr>
<td>Dust Mites</td>
<td>++</td>
</tr>
<tr>
<td>Animals</td>
<td>-</td>
</tr>
<tr>
<td>Molds</td>
<td>?</td>
</tr>
</tbody>
</table>
"I'm sorry, Dear, but I wasn't listening. Could you repeat everything you've said since we've been married."
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Inhalant Allergens

- **Seasonal**
  - Pollen
    - Spring Trees
    - Summer Grass
    - Fall Weeds
  - Mold
    - Fall

- **Perennial**
  - Mold
  - Dust Mites
  - Animals
  - Insects
    - Roach
Rotorod (Sampling Technologies, Minnetonka, MN)
Pollen in grains/m3
Published on internet
Pollen

• Male germinal cell used for plant reproduction
  – Entomophilous: insect-pollinated heavy, sticky pollen, tend **not to cause allergy**
  – Anemophilus: wind-pollinated tree/grass/weed

• Produced in the microsporangium

• Vegetative and generative cells surrounded by intine and exine
Pollen

- Size: Large 10-100 microns
- Antigenicity: Variable
- Major Antigens: Ragweed and grasses standardized
- Cross-Reactivity: Variable

- Thommen’s postulate (1931)
  - Wind born and buoyant
  - Produced in large quantities
  - Abundantly distributed
  - Allergenic
2 major categories of pollen-producing plants:

**Gymnosperms** - ovules are carried naked on scales of a cone
- Non-flowering Trees (e.g., ginkgo)
- Conifers - thin leaves (needles)
  produces cones

**Angiosperms** - sex organs within flowers, and seeds are fruit
- Grasses
- Weeds
- Flowering Trees
Trees

Greatest diversity (300+ pollens in some areas) and

Least antigenic cross-reactivity.
Gymnosperms: 4 families

- Cypressaaceae (Cyprus family)
  - Cypress, False Cypress, Junipers, Cedar, Arbovitae
  - Mid to late winter
  - Good cross-reactivity in this family

- Pinaceae (Pine family)
  - Firs, Larch, Spruce, Hemlock
  - Pollen low antigenicity

- Taxodiaceaea (Bald Cypress Family)
  - Redwoods, Bald cypress

- Ginkgoaceae (Ginko Family)
  - Little known
Trees: Gymnosperms

- Airborne pollens copious, but typically not very potent.

Exceptions:
- Cypress family: very antigenic pollens.
- Pine family: not antigenic but increasingly planted by timber/paper companies, grows close to human habitats.
Trees : Gymnosperms

- Little cross-reactivity between families
- All cypress family members (Cupressaceae) share major cross-reactivity
- Always test / treat a prevalent local cypress (arborvitae, cedar, cypress, juniper, etc.)

Angiosperms -- Families

- Maple Family
- Cashew Family
- Birches
- Oaks/Beeches/Chesntnuts
- Witch Hazel/Sweet Gums
- Walnuts/Hickories
- Magnolia/Tulip Trees
- Sycamores
- Roses/Spirea
- Citrus Family
- Willow/Aspens
- Elms
Trees: Angiosperms

- Oak
  - single most important
  - (Fagaceae)
  - Substantial cross-reactivity between species
  - April/May Peak (*Quercus alba* & *Quercus rubra*)
  - High volume, moderate antigenicity
- Birch
  - among most potent of tree pollens
  - Strong cross-reactivity within family
- Maple / Box Elder (3rd big player)
- Olive / Ash (4th important family)
Trees: Angiosperms

- Most diverse, least cross-reacting plants
- Usually strong cross-reactions only within genus
- Some family cross-reactivities:
  - Birch, alder, hazelnut
  - Oak and beech
  - Maple
  - Olive, ash, lilac, privet (ligustrum)

Trees: Angiosperm Families

- Mulberries
- Cottonwoods, aspens, poplars, willows
- Walnut, butternut, hickory, pecan
- Elms, hackberries
- Legumes (Acacia sp.)
- Eucalyptus
- Australian pines
- Palms
  - As little cross-reactivity, test / treat individually

Locus
Tree Pollen - Summary

When starting to test in new area, always test:

- Fagaceae - oak (local variety)
- Juniperaceae - juniper or cedar
- Aceraceae - maple (box elder)
- Alnus - alder (if in West & North)
- Betula - birch (if in North & Southeast)
- Oleaceae - olive (if in South & West)
- Tropical Trees – if major relevance locally
Grass Pollens

• Among most **POTENT** allergens
  – 20-50 identified antigens
  – 10 allergen groups
  – Expansins, Calcium binding proteins
• Cross-reactivity = strong
• Some food cross-reactions
Grasses

- Common grasses: 5 subfamilies
- Most important in North America
  - Pooideae
  - Cloridoideae
  - Panicoideae
- Local importance
  - Arundinoideae
  - Bambusoideae
Pooideae subfamily

- Most of familiar cultivated and wild grasses, world-wide
  - Brome, fescue, june, orchard, perennial rye, redtop, sweet vernal, timothy, kentucky blue
- Most cereals
  - Barley, oats, rye, spelt, wheat
- Epitopes are widely shared
  - Strong cross-reactions within subfamily
- Timothy pollen appears to contain all of antigenic epitopes

Mixed “field” Grass

The American Academy of Otolaryngic Allergy
Cloridoideae subfamily

- More important in plains and subtropics, but present in most regions of North America
  - Bermuda, buffalo, grama, zoysia grasses
- Strong cross-reactivity within subfamily
  - Bermuda has most of antigenic epitopes
  - Very minor cross-reactivity between other grasses and cloridoids
Panicoideae subfamily

- Common in southern North America
- Bahia, barnyard, Johnson (sorghum) grasses, crabgrass
- Edible corn / maize, millet, sorghum (molasses), and sugarcane
- Limited cross-reactivity with other grasses
- Bahia is most antigenically unique

Depending on local prevalence: test Bahia or Johnson
Grass Pollen Summary

When starting to test in a new area, always test:

Pooideae – timothy
Choridoideae – bermuda
Panicoideae - bahia or johnson
Weeds:

2 groups of major importance in N.A.:

• Composites
  – 3 large tribes of common, wind pollinating, highly allergenic weeds

• Chenopods & Amaranths
  – Two related allergenic families

• Other weeds can be of local importance

Urban Ragweed
Weeds: Composites

• Heliantheae tribe:
  – burro-brush, cocklebur, marsh elder, parthenium, ragweed
  – Usually test / treat ragweed

• Anthemideae tribe:
  – mugwort, sage, sagebrush, wormwood, chrysanthemum, dandelion
  – partly cross-reacts with ragweed Test / treat separately if prevalent

• Astereae tribe:
  – Baccharis species, aster (daisy), goldenrod
  – Test / treat separately if prevalent

Ragweed Pollen
3500 x
Weeds : Heliantheae - Ragweed

- Most important weed (except Pacific coast)
- Cross-reactivity extensively studied
- 7 allergens (Amb a 1)
  - False, giant, short, and western ragweed all strongly cross-react
  - Test / treat with predominant of these 4 species in your area

Short Ragweed

Leiferman KM, Gleich GJ, Jones RT. The cross-reactivity of IgE antibodies with pollen allergens. II. Analyses of various species of ragweed and other fall weed pollens. J Allergy Clin Immunol. 1976; 58:140-8
Weeds : Anthemideae

- Sage tribe –
  - very important
  - worldwide distribution
- Mugwort
  - 80% cross-reactive with ragweed
- Edible: sage, crysanthenum

Weeds: Astereae

- Daisy tribe - (Baccharis sp.), important in coastal U.S. and Western N.A.
- Baccharis tall-growing with small, thistle-like flowers
- Cross-reactivity not fully characterized

Weeds : Other Composites

• Not major pollinators, but strong contact sensitizers
• Dandelion pollen may be airborne
• Discoid tribes
  – Thistles
  – Edible: stevia, artichoke
• Ligulate tribes
  – Edible: dandelion, chicory
Weeds: Chenopods

- Chenopodiaceae family
- Common, heavy pollinators (esp. West), on saline / arid soil
  - Kochia (burning bush), goosefoot, lamb's quarters, mexican tea, orach
  - Edible: beet, spinach, sugar beet
- Some Amaranth cross reactivity, but many strong unique epitopes
- Test / treat separately except for Atriplex sp., (saltbush, shadescale, spearscale, wingscale, etc.)
Weeds: Amaranths

- Amaranthaceae family - closely related to Chenopods
- Common, major pollinators
  - cockscob, careless weed, cottonweed, pigweed, Tidestromia, waterhemp
  - Edible: quinoa
- Strong cross reactivity, but
  - data incomplete
  - can test / treat individually
Sporulating Pteridophytes

Local importance (esp. tropics)

• Ferns
• Horsetails
• Club Mosses (Lycopodium)

Weed Pollen - Summary

When starting to test in a new area, always test:

Heliantheae - a ragweed
Anthemidae - a sage
Amaranthaceae - amaranth or pigweed
Chenopodaceae - russian thistle, Kochia, or lamb’s quarters
Any other weed very common in your area
Inhalant allergens: non-pollens

- Pet sensitivity due to proteins in the **pet's dander** (dead skin that is shed), saliva or urine
- Dust: proteins in the excretion of dust mites
- Fungi: airborne spores
Animal Dander

- **Size:**
  - Small (asthma and rhinitis)
  - 75% Cat Dander 5 to 10 microns

- **Antigenicity:** Cat and Rodent -- Strong

- **Distribution:** Cat antigen routinely sampled in school dust. “sticky”

- **Major antigenic determinants:** Fel d 1

- **Cross reactivity:** Low
Dander: Size/Antigenicity

• 80% of Swedish children with cat allergies have never lived with a cat.

• Homes with cat can have high enough levels to induce tolerance.

• Many allergic children who live with a cat are not sensitized to cat.

Dander: Major Antigen
Fel d 1

- One of the more dominant major antigens
- Saliva, Dander, Tears.
- Function unknown.
- >90% decrease in skin-test reactivity in cat-allergic patients was found after absorption of Fel d 1 from cat dander extract.
- In RAST, a 76% reduction of IgE binding was found after depletion of Fel d 1 (medline 89054671).

http://www.allergen.org/Allergen.aspx
Dander:
Cross-Reactivity

- Numerous epitopes in mammalian / avian epidermal allergens, but main belong to single protein family = lipocalins
  (cat – Fel d1, dog – Can f1)
- Only minor cross-reactivity between species
- Dogs have both common and breed-specific allergens; Cat allergens are less variable
- Test / treat important mammals & birds individually
- Always test cat
Dust mite

- **Size:**
  - Small (asthma and rhinitis)
  - Feces averages 10 microns

- **Antigenicity:** Strong (very prevalent in allergic asthmatics)

- **Distribution:** Widely distributed if humidity adequate

- **Major antigenic determinants:**
  - Two species dust mite, each with two major antigens
  - Der f 1, Der f 2, Der p 1, Der p 2

- **Cross reactivity:** Moderate
House Dust Mites:

- Temperate climate house dust mites: Dermatophagoides farinae, D. pteronyssinus, D. microceras, and Euroglyphus have partial cross-reactivity; less so Blomia tropicalis

- Most common in U.S. are D. farinae and D. pteronyssinus

- Preferred to test / treat dust mites individually
Arthropod

Cockroaches:
- American, German, Oriental, others
- Size: 10-200 microns
- Major roaches have significant cross-reactivity & unique antigens
- 23-60% sensitization urban asthma*
- Test / treat with American or roach mix

Krouse, Chadwick, Gordon, Derebery, Eds. Allergy and Immunology, an Otolaryngic Approach. 2002. Lippincott Williams Wilkins, Philadelphia, p. 47

*NEJM 1997;336:1356-1363
Mold

• **Size:** 3 to 40 microns
• **Antigenic determinants:**
  – Vary with strains. Vary daily in cultures.
  – Stimulates cell mediated late reactions
• **Cross-reactivity:** Complex
• **Taxonomy:** Classifying molds difficult.
  >1 million species. 80 in respiratory dz.

Mold Allergy – Taxonomy issues

- DNA tests reclassifying molds
- “For example, the old genus Helminthosporium has been divided into at least 5 different genera including Drechslera, Bipolaris, Exserohilum, Leptosphaeria, and Cochliobolus. To complicate matters even further, these genera now also include certain species from Curvularia”

Average person inhales about 3 cubic meters daily.
Mold Cross-Reactivity

- Studies are limited
- Several related species have cross-reactivity as well as significant antigenic differences
- Cross-reactivity has also been demonstrated between unrelated genera: Alternaria and Cladosporium
- Antigenic mold polysaccharides may cross-react with fungi, bacteria, and vegetable dusts
- **Test / treat major molds separately**
Mold - Summary

• When starting to test in a new area, always test:
  – Alternaria
  – Aspergillus
  – Cladosporium (Hormodendrum)
  – Helminthosporium
  – Penicillium

• Also, consider < 5 other molds anticipated to be common such as Curvularia, Epicoccum, Fusarium, Mucor, Phoma, Pullularia, Rhodotorula, and smuts / rusts
Non-pollen Antigens - Summary

Always test:

- Cat
- Dog
- Cockroach
- Dermatophagoides farinae &/or pteronyssinus
- Mold mix or Selected molds

Consider testing other common animals or allergens unique to area
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Antigen Selection - Goals

• Identify major allergens locally prevalent during all seasons
  – From references
  – From observation
  – From local practitioners
• Decide on supplier(s)
• Use standardized extracts when possible
• Periodically review, and revise as needed, your office’s allergen test battery
Resources for Antigen Preparation, Geographic Distribution and Cross-reactivity

- Antigen supply houses
- Textbooks, regional flora guide, local conservation district or university botany department
- Allergy Practitioners (otolaryngic or general)
## Antigen Selection

### Testing Guidelines

- Begin with 9-12 antigen screen
- Often ~ 20-24 suffice
- Rarely test > 40 inhalant allergens

### Seasonal
- Spring - tree
- Summer - grass
- Fall - weed

### Perennial
- Mite
- Cockroach
- Animal Epidermal
- Mold
Principles of Screening

• (Nearly) all testing for presence of inhalant allergy should begin with an allergy screen.

• Screening techniques have proven to provide a rapid, efficient (96%), and cost-effective method to assess the presence or absence of allergy.

In vivo allergy screens

Prick testing (multiple prick device) has been demonstrated to have sensitivity of 68% to 100% and specificity of 95% to 97%

The screening battery should represent classes of antigens to which the patient is exposed and which are common geographically.

<table>
<thead>
<tr>
<th>Pollens</th>
<th>Dust mites</th>
<th>Epidermals</th>
</tr>
</thead>
<tbody>
<tr>
<td>– one or two trees</td>
<td>– use at least one, if not both species</td>
<td></td>
</tr>
<tr>
<td>– one or two grasses</td>
<td></td>
<td></td>
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<tr>
<td>– one or two weeds</td>
<td></td>
<td>– should include cat</td>
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<td></td>
<td></td>
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<tr>
<td>Molds</td>
<td></td>
<td></td>
</tr>
<tr>
<td>– include at least two families of mold</td>
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</tbody>
</table>

The American Academy of Otolaryngic Allergy

Pollens – one or two trees, one or two grasses, one or two weeds
Dust mites – use at least one, if not both species
Epidermals – should include cat
Initial Antigen Screen: Temperate Zone

- Trees (2 or 3)
  - Cedar
  - Oak
  - Olive or Birch
- Weeds (1 or 2)
  - Ragweed, Cheno-Am (pigweed, lamb’s quarters) or Sage
- Grasses (1 or 2)
  - Timothy, Bahia or Bermuda
- Arthropods
  - D. Pteronyssinus
  - D. Farinae
  - Cockroach
- Epidermals
  - Cat
  - Dog
- Molds (2 or 3)
  - Alternaria
  - Aspergillus
  - Cladosporium
  - Helminthosporium

9-12 antigens representative of major groups; if any ↑+ do full test
Negative and positive screens

- The absence of reaction to several common antigens suggests that the likelihood of the patient demonstrating sensitivity to a broader panel of antigens is low.

- Therefore no further testing is necessary with a negative allergy screen

- If inhalant screens demonstrate positive reactions, a relevant antigen survey is indicated, and is necessary prior to the provision of immunotherapy
<table>
<thead>
<tr>
<th>Screening Panel Examples</th>
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</thead>
<tbody>
<tr>
<td><strong>P. Fass (FL)</strong></td>
</tr>
<tr>
<td>Bahia grass</td>
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<tr>
<td>Bermuda grass</td>
</tr>
<tr>
<td>Short ragweed</td>
</tr>
<tr>
<td>Oak</td>
</tr>
<tr>
<td>Australian Pine</td>
</tr>
<tr>
<td>Queen Palm</td>
</tr>
<tr>
<td>Melaleuca</td>
</tr>
<tr>
<td>Alternaria</td>
</tr>
<tr>
<td>Cladosporium</td>
</tr>
<tr>
<td>Dust mite</td>
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<tr>
<td>Cockroach</td>
</tr>
<tr>
<td><strong>R. Mabry (TX)</strong></td>
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<tr>
<td>Bermuda grass</td>
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<tr>
<td>Timothy grass</td>
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<tr>
<td>Short ragweed</td>
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<tr>
<td>Oak</td>
</tr>
<tr>
<td>Mountain cedar</td>
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<tr>
<td>Alternaria</td>
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<td>Helminthosporium</td>
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<tr>
<td>Dust mite</td>
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<tr>
<td>Cat</td>
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<tr>
<td><strong>B. Gordon (MA)</strong></td>
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<tr>
<td>Timothy grass</td>
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<tr>
<td>Short ragweed</td>
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<tr>
<td>Oak</td>
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<tr>
<td>Alternaria</td>
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<td>Cladosporium</td>
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<td>Johnson smut</td>
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<tr>
<td>Dust mite</td>
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<tr>
<td>Cat</td>
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<tr>
<td>Dog</td>
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<tr>
<td>Cockroach</td>
</tr>
</tbody>
</table>
Screening Panel Examples

W. Mims (NC)
IgE testing
  Timothy grass
  Tree Mix
  Weed Mix
  Mold Mix
  Cat
  Dog
  Dust Mite x 2
      (Df, Dp)
  Cockroach

M. Zacherek (MI)
SPT
  kentucky bluegrass
  bermuda grass (for all of our snowbirds that fly down to florida)
  feather mixture
  dog hair
  cockroach
  cat hair
  mite farinae
  mite pteronyssinus alternaria
  hormodendrum
  english plantain
  box elder
  short ragweed
  white oak.

S. Lin (Baltimore)
SPT
  Cat
  Dog
  d. pter & farinae
  white oak
  Ragweed
  timothy
  Alternaria
  american elm
  Bermuda
  english plantain
  Hickory
  aspergillus
  cockroach
Do not use Screening Panel

D. Palmer (Utah)
Does not do screens

S. Houser (OH)
(SPT screen only for kids and polyps, usually full 30 antigen IDT)

- tree mix
- grass mix
- weed mix
- dust mites (both mixed together)
- cat
- dog
- mold mix

A. Patel (Baltimore)
MQT x 30 allergens
Thank you

Moss Glen Falls
Vermont