61 Juniperus Asheii (MC) Pollen Utilized as an Allergen in The Biogenics Chamber: Comparison of Natural and Controlled Exposure

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Abstract

Rationale: Juniperus asheii (MC) pollen allergy is a robust model commonly used in natural studies of pharmacologic efficacy. This study was performed to compare the symptoms elicited by that antigen in the natural season and in a controlled chamber environment outside of the natural season.

Methods: Twenty eight subjects sensitized to Juniperus asheii were monitored for symptoms during the 2009-2010 MC season. These subjects were then exposed in a controlled environment outside of the season. Pollen levels of 12,000 grains/m³ were utilized during 2 hour chamber exposures occurring on consecutive days. Total nasal symptom scores (TNSS) and total ocular symptom scores (TOSS) were compared in the natural and the chamber studies.

Results: To reach meaningful symptoms in 2/3 of subjects during 2 hour chamber exposures, up to four priming runs were required. Although chamber runs utilized higher pollen counts than those during mean natural exposures (3,500/m³), chamber counts were one-half those recorded during maximum natural pollination. Strong correlation was found between the symptoms levels of subjects during natural and chamber exposures.

Conclusions: In brief sensitization chamber runs, Juniperus asheii pollen exposures elicited meaningful symptoms in 2/3 of subjects shown to be symptomatic during the natural season. With consecutive sensitization runs, subjects can be selected for pharmacologic studies utilizing Juniperus asheii pollen in a controlled environment.

Materials and Methods

This study was conducted at the Biogenics Research Chamber. From our previous validation experiments, it was determined that priming exposures with a pollen density of 12,000 grains/m³ would be required.

All study participants were over 18 years of age and received written informed consent before starting the study. The protocol, amendments and informed consent documents were approved by IntegReview IRB, Austin, Texas. The study was conducted according to Good Clinical Practice standards.

Twenty eight patients previously monitored during the 2009-2010 MC winter season in San Antonio, Texas were selected for inclusion in this study. All subjects had a >two year history of seasonal allergic rhinoconjunctivitis to MC and documented sensitivity to MC by prick skin test response. These patients had kept symptom diaries for a one-week period when the pollen counts for MC ranged from 50 to 21,850 gr/m³ (mean 3,500/m³). Patients had scored AM and PM nasal symptoms of a) congestion, b) pruritus, c) sneezing and d) runny nose in a severity scale of 0 to 3. Ocular symptoms were available using a similar score for pruritus, redness and tearing in 15 of the patients.

Priming Visits – Chamber pollen exposure
All subjects were re-evaluated at the first priming visit to determine their eligibility to enter the chamber. Only subjects with low reflective TNSS / TOSS assessment scores were allowed to proceed into the chamber. Participants were exposed to MC pollen for 2 hours in up to 4 consecutive days . Pollen levels were monitored by 8 Allergenco systems placed throughout the chamber (Environmental Monitoring Systems, Inc., Charleston, SC) powered by suction pumps run at 15 liters/minute. Pollen counts were done at 30 minute intervals to ensure uniform distribution of pollen throughout the seating area. Subjects marked their symptom diary forms every 30 minutes for nasal and ocular symptoms. The diaries were processed with an OMR reader.

Study Timeline

Natural Exposure - Mountain Cedar Season

Dec. 2009 - January 2010

28 Patients: TNSS diaries

15 Patients: TNSS+TOSS.

Screening Visit - August 2010

a. Informed Consent.

b. Run–In Diaries issued for one week.

c. Inclusion, exclusion criteria reviewed.

Chamber Exposures - Aug. – Sept. 2010

a. Pollen Concentration: 12,000 gr/m³ +/- 500.

b. Chamber runs: two hours.

c. Up to 4 consecutive days.

d. OMR scan of TNSS/TOSS

e. Data analysis.

Chamber

Photo of the large chamber used in MC studies. All the furniture is movable for different seating configurations and for easy cleaning of the chamber. Not shown are the pollen collecting stations which are distributed throughout the chamber and the pollen dispersal system.

The glass window at the left far end is for observation from the control room.

Conclusions

• Nasal symptoms recorded by subjects during natural exposure correlate well with the symptoms elicited during indoor chamber exposures.

• There is a strong correlation between skin test reactivity to MC pollen and TNSS scores recorded in chamber exposures.

• Most of the very sensitive subjects (89%) developed significant symptoms within the first two days of chamber exposure. Half of the very symptomatic subjects had symptoms the first day.

• To establish a correlation for TOSS between natural and chamber exposures will require a larger population study.

• Mountain cedar pollen can safely be used in an indoor chamber setting for studies of allergic mechanisms and for studies of pharmacologic efficacy.

We have verified the advantages of studies in an allergen challenge chamber :

• a) studies can be done outside of the natural pollen season in the summer;
• b) exposure is controlled and uniform for all study participants at 12,000 grains/m³ ;
• c) there is no impact from the outside weather;
• d) there is ensured compliance with the timeliness and completion of symptom assessments and;
• e) the symptom scoring is easily monitored by the chamber staff.

Introduction

Pollen challenge chambers have been utilized over the past several years to study the effectiveness of medication in controlling signs and symptoms of allergic rhinoconjunctivitis and asthma. Pollens from common airborne allergens, ragweed, birch, Japanese Cedar, or grass, (1-5) are typically introduced into a highly controlled environment to stimulate signs and symptoms of allergic rhinoconjunctivitis in sensitive populations. Concerns have been raised about the validity of chamber studies when compared to 'park studies' or to regular seasonal studies.

The traditional methods of utilizing conventional seasonal or perennial exposure to allergens for the investigation of study medication are fraught with several limitations. These include: a) variations between the timing of a pollen season in the various regions, b) regional differences between pollen counts, and c) differences in personal exposure due to lifestyles and weather conditions. Furthermore, compliance with paper and electronic diaries may be questionable and drop-out rates may be high. Subjects may also be prone to change their diary scores in order to qualify for a research project in which they may receive monetary compensation.

The advantages of allergen chamber studies include: a) they may be done outside of the natural pollen season, b) the exposure is controlled and uniform for all study participants, c) there is no impact from the weather, d) there is ensured compliance with timeliness and completion of symptom assessments and medication administration, and e) the symptom scores are monitored by the chamber staff.

MC pollen has never been used in a indoor pollen chamber setting. The Biogenics Research Chambers have been validated and shown to produce symptoms in MC allergic individuals.

Discussion

We have designed and constructed two exposure chambers to study the responses of sensitive individuals in a controlled environment to MC and other local pollens. No unexpected adverse events were recorded.

These chamber studies were conducted six months after the end of the MC season in San Antonio, Texas when the average outdoor temperature was 93.6º F, there were no significant outdoor airborne allergens, and the subjects were in an unprimed state. Despite the lack of priming, most of the very sensitive subjects (89%) developed significant symptoms within the first two days of chamber exposure. After four consecutive priming days in the chamber, 67% of subjects reached a TNSS of >7.

We found a strong correlation between the nasal allergic rhinitis symptoms achieved during the natural seasonal exposure with the exposures in our indoor chamber (*Figure #1*). The correlation between TOSS scores failed to reach statistical significance (p=0.08) in part because the available sample size was smaller (13 degrees of freedom vs. 26).

We found a strong correlation between MC skin test reactivity and the recorded in-chamber TNSS. Previously published chamber studies using ragweed pollen have not shown this correlation. Our findings may be due to the use of a different allergen (MC vs. ragweed) or to differences in the genetic profiles of the populations studied.

The correlation between symptoms and skin test reactivity was not observed when the skin test results were compared to the natural exposure TNSS. We cannot explain the lack of correlation in the natural exposure setting (*Table #1*).

Results

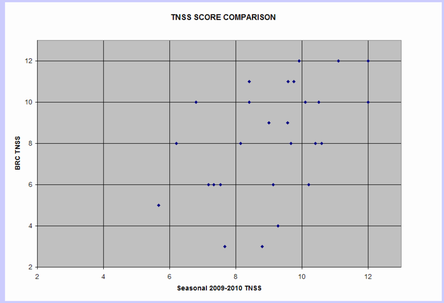

Figure #1: Comparison and correlation of average TNSS during the December 2009 – January 2010 MC season in San Antonio with the TNSS recorded in the chamber priming runs @ 12,000 pollen grains/m³.

r = 0.483 (r^2 = 0.233) P<0.01

PATIENT CHARACTERISTICS				SEASONAL DATA		CHAMBER DATA		
Age	Sex	SUBJ NUM	Skin Test Wheal(mm)	Avg. TNSS	Avg. TOSS	Day Primed	TNSS Max.	TOSS Max
42	F	1002	3	9.27	6.6	---	4	2
22	F	1004	5	5.67		---	5	3
22	M	1006	3	7.17		---	6	5
45	M	1007	6	10.4	7.4	2	8	9
58	M	1018	3	7.67	4.4	---	3	1
44	F	1021	8	8.79		---	3	1
56	F	1022	7	9		1	9	8
48	F	1024	10	9.75		1	11	8
32	F	1028	11	6.2	4.13	4	8	5
38	F	1029	8	10.2	4.13	---	6	1
37	F	1030	12	9.91		1	12	7
26	M	1031	20	11.1		1	12	4
52	M	1032	30	9.58		1	11	9
49	F	1033	7	7.33		---	6	5
25	F	1035	5	7.53		---	6	4
32	F	1036	5	12	7.35	2	10	5
38	F	1037	7	12	9	2	12	6
53	F	1039	8	9.67	5	3	8	4
24	M	1040	4	9.67	6.27	2	8	6
52	F	1042	5	9.13	6.4	---	6	2
48	F	1044	10	10.5	6.93	1	10	7
48	F	1045	6	6.8	5.07	1	10	2
44	F	1046	3	8.15	5.54	2	8	7
55	F	1047	7	8.4	5.33	2	11	8
28	F	2002	12	10.1		2	10	3
51	F	2008	12	8.4	6.87	2	10	6
22	F	2011	3	9.57		1	9	2
48	M	2012	6	10.6		1	8	5

Table #1. Patient characteristics and the seasonal TNSS/TOSS for the 2009-2010 MC season and the corresponding TNSS/TOSS observed in the chamber.

Correlation Coefficient Chamber TNSS and ST wheal: r = 0.512 (r^2 = 0.262) p = 0.005.
Correlation Coefficient Seasonal TNSS ST and wheal : r = 0.222 (r^2= 0.049) p = 0.255.
Correlation Coefficient TOSS: Seasonal vs Chamber: r = 0.462 (r^2= 0.214) p = 0.082.

References

1. Day JH, Horak F, Brisco MP, et al. The role of allergen challenge chambers in the evaluation of anti-allergic medication: an international consensus paper. Clinical and Experimental Allergy Reviews. 2006;6:31-58.
2. Renborg SM, Mosbech H, Johnsen CR, Poulsen LK. Exposure chamber for allergen challenge. The development and validation of a new concept. Allergy 1996;51:82-8.
3. Day JH, Briscoe MP. Environmental exposure unit: a system to test anti-allergic treatment. Ann Allergy Asthma Immunol. 1999;83:83-93.
4. Hashiguchi K, Tang H, Fujita T, et al. Pilot study of Japanese cedar pollen exposure using a novel artificial exposure chamber (OHIO Chamber). Clinical and Experimental Allergy Reviews. 2008;8:30-6.
5. Hashiguchi K, Tang H, Fujita T, et al. Validation study of the OHIO Chamber in patients with Japanese Cedar pollinosis. Int Arch Allergy Immunol. 2009;149:141-9.
6. Ellis AK, Ratz JD, Day AG, Day JH. Factors that affect the allergic rhinitis response to ragweed allergen exposure. Ann Allergy Asthma Immunol. 2010;104:293-298.
7. Hohlfeld JM, Holland-Letz T, Larbig M, et al. Diagnostic value of outcome measures following allergen exposure in an environmental challenge chamber compared with natural conditions. Clinical and Experimental Allergy. 2010; 40: 998-1006.
8. Patel P, D'Andrea C, Sacks HJ. Onset of action of azelastine nasal spray compared with momethasone nasal spray and placebo in subjects with seasonal allergic rhinitis evaluated in an environmental exposre chamber. Am Journal Rhinology 2007; 21: 499-503.
9. Berkowitz RB, Woodworth GG, Lutz C, et al. Onset of action, efficacy, and safety of fexofenadine 60 mg/pseudoephedrine 120 mg versus placebo in the Atlanta allergen exposure unit. Ann Allergy Asthma Immunol. 2002;89:38-45.