



# Are results of environmental exposure units transferable to real-life exposure?

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## Purpose of review

To reflect on and discuss the recent evidence of reliability of the challenge chamber results for real life, especially focusing on the validation of allergen concentrations being 'physiologic' and generating symptoms in the exposed volunteers comparable to the natural exposure during pollen season and to the home environment.

## Recent findings

Publications of the last 18 months covered by this review focus on the validation of allergen chamber systems, especially on the quantitative comparison of environmental allergen exposure with allergen challenge chamber (ACC) exposure. It highlights new evidence on the question of relevance of environmental and experimental priming effects and discusses the selection of relevant parameters for the quantification of treatment effects.

## Summary

Benchmarks for validating the ACC systems have been defined and are referred to in clinical ACC trials in increasing amount. So the level of evidence concerning the validity and reliability of allergen chamber systems is steadily growing, especially in allergen-specific models, priming effects and reproducibility of results in the same study population at different time points and with different challenge systems.

## Keywords

allergen challenge chamber, allergen load, priming, validation

## INTRODUCTION

Challenge chamber models for the investigation of mechanisms and treatment of allergic rhinoconjunctivitis were developed in the 1980s [1]. Studies using this challenge model can not only investigate the treatment effects of antiallergic therapies like antihistamines, corticosteroids or immunotherapy, but also evaluate the mechanisms of allergic inflammatory reaction like mediator release and time course of cell and mediator synergism. The allergen challenge chamber (ACC) model is approved and accepted by the Food and Drug Administration and European Medicines Agency (EMA) at least for phase II trials and further supporting phase III trials. EMA suggests ACC trials for conducting dose finding trials of immunotherapeutic products [2]. Grass and ragweed pollen, tree pollen, Japanese cedar pollen, house dust mite and cat epithelia serve as routine allergen sources for challenge. Currently, there are several challenge chambers in use in Europe, North America and Japan.

ACCs enable the investigators to generate a stable and reproducible allergen climate to which volunteers can be exposed. This offers advantage

compared with the natural seasonal pollen exposure, as confounding conditions like rain, temperature and wind influencing the pollen counts can be excluded. Low pollen counts during seasons generate fewer symptoms in the pollen allergic volunteers, which consecutively diminishes the possibility to detect a treatment effect of an investigational compound during a clinical trial. Several advantages and disadvantages of the ACC model are discussed in a European review dated 2011 [3<sup>••</sup>].

## VALIDATION OF ALLERGEN CHALLENGE CHAMBER MODELS

Challenge chambers in use normally are validated in terms of reproducibility of ACC conditions, stability

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## KEY POINTS

- Validation of allergen challenge chamber systems do allow comparability and reproducibility of trial data generated in an ACC trial.
- Real-life allergen exposure is comparable to the experimental exposure in ACC by means of clinical symptoms.
- Environmental as well as experimental priming effects can be detected, but are usually not of clinical relevance.
- The allergen load of pollen, house dust mite or cat epithelia required to cause clinical symptoms is defined for each ACC separately and is not transferable from one ACC system to the other.

of allergen load and reproducibility of symptoms in patients [4,5].

A short article published in 2012 provides a summary of the needs for the validation of ACC systems which have been defined during a U.S. American NIH workshop in Bethesda, Maryland, USA, in 2010 [6<sup>¶</sup>]. It is as follows:

- (1) the standardization of ACC conditions (uniformed distribution of pollens, amount of pollen, air temperature and humidity);
- (2) the determination of the reproducibility of ACC clinical trial data for evaluating treatments for allergic rhinitis; and
- (3) the determination of reproducibility of allergen exposure and treatment outcomes between different ACCs.

It was postulated to compare patients on natural allergen exposure to ACC exposure first, then to compare the treatment effects of established anti-allergic medications in patients on natural allergen exposure and on ACC exposure in a multicentre mode. Furthermore, it was recommended to investigate the variabilities between different ACC models [7]. The author additionally mentioned to document the reproducibility of symptoms in the same population at different time points [6<sup>¶</sup>].

The use of standardized pretreatment and post-treatment allergen challenges in an ACC allows precise determination of efficacy, including onset of action, magnitude and duration of action of any antiallergic treatment. A precisely designed clinical ACC trial with a carefully selected patient population can reduce the cost of drug development substantially by requiring significantly less time than a field study and smaller numbers of

individuals per trial. Treatment effects of immunotherapy could be clearly determined [8]; evaluations of naturally exposed patients do often show less efficacy, as shown by Howarth *et al.* [9] recently. The authors could demonstrate that treatment effects of allergen immunotherapy are correlated with the symptom severity in patients. Limiting factors for the power of immunotherapy studies usually are the natural pollen exposure and the corresponding symptom severity of the patients, retrospective symptom scoring for baseline evaluation and the combined evaluation of patients with different symptom severity. The authors conclude with the suggestion to divide patients into three groups of different symptom severity and evaluate separately to enhance the power of the trial.

## SELECTION OF PARAMETERS FOR ALLERGEN CHALLENGE CHAMBER TRIALS

To avoid the production of unfeasible data during a clinical trial, there would be some restrictions to select patients suitable for allergen challenge trials: skin prick test results do not necessarily correlate with clinical reactivity; therefore one important inclusion criterion for a challenge chamber trial should be proven clinical reactivity during the challenge session. Recent data on the correlation between skin test reactivity and clinical rhinitis symptoms are heterogeneous. In a study published by Ellis *et al.* [10], an investigation of 123 patients with ragweed pollen allergy showed no correlation between skin prick test reactivity and clinical rhinitis symptoms. A trial of Huss-Marp *et al.* [11] investigating the correlation between different challenge tests, skin prick test reactivity and specific IgE levels in 104 grass pollen allergic patients could show a correlation between skin test reactivity and sIgE levels, but not of clinical reactivity and specific IgE levels.

## PRIMING EFFECTS

ACC trials do not provide long-term exposure with generation and direct evaluation of the late-phase reactions, but evaluation of priming effects can be done with ACC trials. Differences in clinical reactivity immediately after season and out of season, corresponding to a priming effect, were newly investigated by Yuki *et al.* [12<sup>¶</sup>]. In the present trial, the authors stated that the symptoms were induced more rapidly after the end of pollen season than with out-of-season exposure. During pollen season with variable amounts of pollen counts, patients often exhibit symptom-free intervals during phases of low pollen exposure. But

nevertheless, upregulation of inflammatory cells and mediators occur. This minimal persistent inflammation leads to increased sensitivity and inflammatory response to allergens. The result of such a priming effect is a more rapid onset of symptoms, but not a higher extent of symptoms, as shown by Yuki *et al.* [12<sup>¶</sup>]. That priming effects do not influence the overall amount of symptoms could be demonstrated also by Badorrek *et al.* [13]. The effect of a combination of 10 mg cetirizine and 120 mg pseudoephedrine was investigated in a randomized, placebo-controlled, double-blind, four-way crossover study in 70 patients with grass pollen allergy. The preparation, which has been established since the 1990s, was administered 2 h after the start of each 6-h ACC session. In this study, there was no difference in the clinical efficacy of the preparation, unless given in seasonally primed patients or outside the season. This indicates that ACC trials can be conducted not only outside the pollen season, but also inside the pollen season, as results are not influenced by the environmental priming effects.

## DETERMINATION OF ALLERGEN LOAD

ACC pollen models are widely accepted. Natural exposure levels are almost impossible to determine because of high variabilities, and therefore transferability into ACC loads is no option. Determination of pollen allergen load within ACC systems is usually done with the help of symptom response [3<sup>¶¶</sup>].

House dust mite models were first established in the 1990s [14,15]. The authors used an allergen load of 40–100 ng/m<sup>3</sup>, referring to a home exposure level of 0.03–30 ng/m<sup>3</sup> with a mean particle size of 20 µm. This allergen concentration could be shown to induce allergic rhinitis symptoms as well as biphasic asthmatic symptoms in sensitive individuals [15] and is still used [5]. At the moment, several trials are running focusing on the house dust mite model to be revalidated.

Using topical intranasal or intrabronchial challenge fluids, the individual reaction threshold has to be determined for each patient [16], which is very complex. And also for challenge chamber systems, the minimum pollen count per m<sup>3</sup> of air, that induces symptoms in the majority of patients, has to be determined. In Japan, ACCs investigating Japanese cedar pollen allergics have been in use since 2005 and have been validated systematically [17]. In a recent North American trial, Japanese cedar pollen counts three to four times the average seasonal counts were required to elicit significant symptoms [18<sup>¶</sup>]. Two ACCs of different size were

validated. The trial not only determined the necessary amount of Japanese cedar pollen to cause significant symptoms in allergic patients, but also quantified the priming effects by repeated ACC sessions up to four consecutive 2-h exposures.

A cat challenge model was established in 1993 [19]. In a recent trial, the treatment effect of omalizumab in 69 cat asthmatic patients was investigated by Corren *et al.* [20<sup>¶¶</sup>]. In this randomized, double-blind, placebo-controlled parallel group study conducted in three different study centres, cat allergic patients were challenged before and after 16 weeks of treatment with subcutaneous omalizumab or placebo. Cat challenges were performed in a room comparable to a living room with upholstered furniture and a blanket. The room was contaminated with cat allergens with the help of two neutered adult cats, which were kept in the closed room at all times except during challenges. The trial is very instructive for two reasons: investigators argued that one main criterion to use an ACC instead of a topical allergen provocation test was the different allergenic particle size leading to different clinical pictures of upper and lower airway symptoms. Explanation on that point is provided by De Blay [21]. The median level of Fel d 1 at home capable of inducing symptoms in 50 samples was evaluated. The author mentioned that a positive bronchial challenge test was achieved with 13.5 ng/m<sup>3</sup> of Fel d 1 with 10.3 µm particle size. The median time for the onset of symptoms was 30 min. Thus, at home, the minimum amount of Fel d 1 needed to induce symptoms was 4.2 ng. This indicated that bronchial challenges with particles greater than 10 µm of cat allergen are comparable to the levels of Fel d 1 that induce symptoms at home. Furthermore, they investigated the house dust mite exposure at home in 37 mite allergic patients. There were about 60 ng/m<sup>3</sup> of Der p 1 found in the air after air turbulences, 80% had a particle size greater than 10 µm. The authors estimated the amount of Der p 1 to induce symptoms at home was 12 ng, with an onset time of around 30 min. In their bronchial challenge tests, they used 23 ng of Der p 1, so the levels were comparable to the environmental exposure.

Furthermore in the omalizumab trial [20<sup>¶¶</sup>], cat allergen levels were highly variable within and amongst the three study centres; the levels of Fel d 1 allergen ranged between 0 (undetectable) and around 23 000 ng/m<sup>3</sup>. The median level of Fel d 1 allergen was 425 ng/m<sup>3</sup> at baseline challenge and 429 ng/m<sup>3</sup> at week 16. The highest level of 22 631 ng/m<sup>3</sup> was measured once; no other measurement exceeded 6445 ng/m<sup>3</sup>. Only at very high continuous exposure levels, FEV1 decrease was more pronounced in the placebo group. A high variability

in cat allergen levels was reported in the past in several trials investigating environmental exposure. Levels of airborne cat allergens are described between around 2 and 600 ng/m<sup>3</sup> in homes with cats and around 3–90 ng/m<sup>3</sup> in homes without cats (see [20]). A cumulative effect with increasing allergen concentrations over time of cat persistence could be shown.

In another recent trial, clinical efficacy of intracutaneous Fel d 1 derived peptides was determined with help of a challenge chamber model [22]. This study was a randomized, double-blind, placebo-controlled, parallel group study evaluating the safety and efficacy of intradermal injections of two dosing regimens of Cat-PAD performed in Canada. The research group used a validated allergen-dispensing system to ensure a mean airborne level of around 50 ng of Fel d 1 per m<sup>3</sup>. The authors determined the chosen allergen amount to be comparable to the previously measured home exposure levels of 10–200 ng/m<sup>3</sup> in homes with cats. A total of 202 individuals were treated with the peptide or placebo and were exposed to the cat allergen ahead of the treatment for baseline evaluation and around 5 months after the start of treatment. Priming effect was induced by 3-h challenges on four consecutive days in the ACC. A subgroup of 89 patients were followed up and rechallenged 1 year after the start of treatment. A significantly better efficacy was shown with four administrations separated by 4 weeks of the 6 nmol dose than for eight administrations separated by 2 weeks of the 3 nmol dose of the preparation.

A strong correlation between the allergic symptoms evoked by natural allergen exposure versus clinical symptomatology in an environmental exposure unit could be demonstrated by Jacobs *et al.* [23]. The authors investigated different reaction types of allergic rhinitis in 31 patients as well as the clinical cross-reactivity of different ragweed types in 26 of the 31 patients with the help of both challenge models. They were not only able to differentiate three reaction types of allergic rhinitis, but also to demonstrate strong correlation of clinical symptoms in both challenge models, the environmental exposure unit and the natural seasonal exposure. So comparability between natural exposure and experimental exposure in an ACC is better provided by clinical symptoms and not by allergen load.

## CONCLUSION

Validation criteria for ACC systems have been established indicating the need for standardizing the system itself, in comparison with other ACCs and against natural allergen exposure. The level of

evidence concerning the validity and reliability of allergen chamber systems is steadily growing, especially for specific allergen models, priming effects and reproducibility of results in the same study population at different time points and with different challenge systems. Evidence on the equivalence of natural pollen exposure versus experimental pollen exposure in an ACC is not easy to achieve because of two main reasons: very heterogeneous intraseasonal pollen counts over the years are not correlating with the allergen amount and the allergenicity of the pollen, which is influenced by humidity, air pollution and other exogenous factors, that are not yet fully elucidated and cannot be mimicked by an ACC. To compare the natural to experimental allergen exposure in an ACC, the use of clinical symptoms as a criterion instead of allergen load is the better option.

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## Conflicts of interest

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## 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 (pp. 316–317).

1. Horak F, Jäger S. The Vienna challenge chamber (VCC) – a new method for allergen exposition test. *Wien Klin Wochenschr* 1987; 99:509–510.
2. Committee for Medicinal Products for Human Use (CHMP) at the European Medicines Agency (EMA). Guideline on the clinical development of products for specific immunotherapy for the treatment of allergic diseases. London, UK; November 2008; Draft CHMP/EWP/18504/2006. <http://www.emea.europa.eu/pdfs/human/ewp/1850406enfin.pdf>. [Accessed March 14th 2013]
3. Devillier P, Le Gall M, Horak F. The allergen challenge chamber: a valuable tool for optimizing the clinical development of pollen immunotherapy. *Allergy* 2011; 66:163–169.
4. Hohfeld 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. *Clin Exp Allergy* 2010; 40:998–1006.
5. Day JH, Horak F, Briscoe MP, *et al.* The role of allergen challenge chambers in the evaluation of anti allergic medication: an international consensus paper. *Clin Exp Allergy Rev* 2006; 6:31–59.
6. Bernstein JA. Correlation between a pollen challenge chamber and a natural allergen exposure study design for eliciting ocular and nasal symptoms: early evidence supporting a paradigm shift in drug investigation? *J Allergy Clin Immunol* 2012; 130:128–129.

This publication paper summarizes the need for validation of ACC systems as defined by NIH and is described in [7].

7. Togias A. Asthma, Allergy, and Inflammation Branch, Division of Allergy, Immunology and Transplantation, NIAID/NIH. Environmental exposure units: clinical trial design for validation. Bethesda: National Institute for Allergy and Infectious Diseases/National Institutes of Health; 2010. pp. 1–12.
  8. Van Overtvelt L, Baron-Bodo V, Horiot S, *et al.* Changes in basophil activation during grass-pollen sublingual immunotherapy do not correlate with clinical efficacy. *Allergy* 2011; 66:1530–1537.
  9. Howarth P, Malling HJ, Molimard M, Devillier P. Analysis of allergen immunotherapy studies shows increased clinical efficacy in highly symptomatic patients. *Allergy* 2012; 67:321–327.
  10. 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.
  11. Huss-Marp J, Darsow U, Brockow K, *et al.* Can immunoglobulin E-measurement replace challenge tests in allergic rhinoconjunctivitis to grass pollen? *Clin Exp Allergy* 2011; 41:1116–1124.
  12. Yuki A, Terada T, Ichihara T, *et al.* Evaluating the effects of testing period on ■ pollinosis symptoms using an allergen challenge chamber. *Allergol Int* 2011; 60:533–539.
- This study newly quantifies the amount of environmental priming for clinical symptoms in pollinosis.
13. Badorrek P, Dick M, Hecker H, *et al.* Antiallergic drug testing in an environmental challenge chamber is suitable both in and out of the relevant pollen season. *Ann Allergy Asthma Immunol* 2011; 106:336–344.
  14. Horak F, Jäger S, Berger U, *et al.* Controlled exposure to mite allergen for a dose-finding of dimethindene maleate (DMM). *Agents Actions* 1994; 41:C124–C126.
  15. Rönborg SM. Exposure chamber for allergen challenge. The development and validation of a new concept. *Allergy* 1996; 51:82–88.
  16. Scadding GW, Calderon MA, Bellido V, *et al.* Optimisation of grass pollen nasal allergen challenge for assessment of clinical and immunological outcomes. *J Immunol Methods* 2012; 384:25–32.
  17. Ito K, Terada T, Yuki A, *et al.* Preliminary study of a challenge test to the patients with Japanese cedar pollinosis using an environmental exposure unit. *Auris Nasus Larynx* 2010; 37:694–699.
  18. Jacobs RL, Ramirez DA, Andrews CP. Validation of the biogenics research ■ chamber for *Juniperus ashei* (mountain cedar) pollen. *Ann Allergy Asthma Immunol* 2011; 107:133–138.
- This study provides fundamental information on connection between pollen load and clinical symptom as well as priming effects induced in an ACC trial.
19. Wood RA, Laheri AN, Eggleston PA. The aerodynamic characteristics of cat allergen. *Clin Exp Allergy* 1993; 23:733–739.
  20. Corren J, Wood RA, Patel D, *et al.* Effects of omalizumab on changes in ■ pulmonary function induced by controlled cat room challenge. *J Allergy Clin Immunol* 2011; 127:398–405.
- This work provides plenty of information on the validation of ACC systems, especially concerning the determination of allergen load and particle size, differences between systems and correlation between allergen load and symptoms.
21. De Blay F. Provocation tests as measure of efficacy and dosage. *Allergy* 2011; 66 (Suppl. 95):47–49.
  22. Patel D, Couroux P, Hickey P. Fel d 1-derived peptide antigen desensitization shows a persistent treatment effect 1 year after the start of dosing: a randomized, placebo-controlled study. *J Allergy Clin Immunol* 2013; 131:103.e7–109.e7.
  23. Jacobs RL, Harper N, He W, *et al.* Responses to ragweed pollen in a pollen ■ challenge chamber versus seasonal exposure identify allergic rhinoconjunctivitis endotypes. *J Allergy Clin Immunol* 2012; 130:122–127.
- This study compares the natural versus experimental allergen exposure effects and discriminates between the different reaction types in allergic rhinitis.