

Correspondence

Intralymphatic immunotherapy: Time interval between injections is essential

To the Editor:

Witten et al¹ recently published an extensive Letter to the Editor in the *Journal of Allergy and Clinical Immunology* questioning that intralymphatic immunotherapy (ILIT) with grass pollen, at the present state of knowledge, is ready for clinical use. The results of their double-blind placebo-controlled clinical trial in 38 adult patients with hay fever revealed that 3 or 6 intralymphatic injections with grass pollen allergen extract induced some promising immunological changes, such as a regulatory T-cell response and elevated IgG₄ levels. However, no improvement in clinical parameters such as symptom or medication scores was observed. The authors perceived their study design to be similar with our studies, where symptom amelioration was observed after 3 intralymphatic injections.²⁻⁴

Witten et al used a similar allergen extract, the same dose, and the same route as we did; however, a very important difference in their and our protocols must be noted. In our trials, the time interval between injections was 4 weeks, whereas Witten et al injected every 2 weeks, which is a major difference that we regard as the most likely explanation for the different clinical outcome.

We chose a 4-week time interval between injections to be in accordance with general recommendations in vaccine immunology. A 4-week interval allows for the development of successive waves of antigen-specific immune responses without interference.⁵ Memory B-cell formation and affinity maturation require phases in which little antigen is present in lymph follicles so that competition for the antigen can positively select high-affinity B cells. There is not a single Food and Drug Administration–approved vaccine with a shorter time interval than 4 weeks between injections,⁶ and the Centers for Disease Control and Prevention guidelines consider a vaccination “invalid” if the time interval is shortened to less than the recommended minimum: “For example, if the first and second doses of *Haemophilus influenzae* type b (Hib) were administered only 14 days apart, the second dose would be invalid and need to be repeated because the minimum interval from dose 1 to dose 2 is 4 weeks. The repeat dose should be administered ≥ 4 weeks after the invalid dose (in this case, the second). The repeat dose is counted as the valid second dose.”⁶

Much less is known about the effect of the time interval between injections on the affinity of functionality of the T-cell response, because T-cell avidity cannot readily be assessed. However, John Kappler and Philippa Marrack, leading experts in this field, conclude that “competition for limited resources is a common theme throughout nature, and immunology is no exception,” stating that antigen is one of the resources CD4 T cells are competing for and that higher affinity T cells have a competitive advantage.⁷ In line with this, there is recent evidence that low-dose peptide immunization generates higher affinity CD8 T cells than does high-dose peptide immunization.⁸ A more recent example using CD4 T-cell peptides for allergen-specific immunotherapy may also be interpreted along these lines. Patel et al⁹ observed that 4 injections with Fel d 1 derived peptides in monthly intervals successfully resulted in symptom amelioration in patients with cat allergy–induced rhinoconjunctivitis,

whereas immunotherapy with 8 injections in 2-week intervals was ineffective.

That a 2-week time interval between injections would produce only low-affinity B- and T-cell responses may seem against current clinical practice in allergen-specific immunotherapy, because classical subcutaneous allergen immunotherapy regimens up-titrate the doses in weekly intervals. However, it should not be forgotten that these “invalid” injections are followed by “valid” maintenance doses that are administered with a minimum 4-week interval in accordance with the Centers for Disease Control and Prevention guidelines.

Moreover, pulsatile antigen kinetics, such as achieved by our monthly injections, tend to induce a T_H1-polarized immune response, whereas prolonged antigen exposure produced by frequent injections tends to induce a T_H2-polarized response. This was demonstrated by Guery et al¹⁰ who showed that uninterrupted ovalbumin delivered by the use of a micro-osmotic pump induced IL-4 and IgE production, whereas a bolus injection generated T_H1-type immune responses with IFN- γ and IgG.

Finally, the reason why Witten et al observed a higher rate of allergic side effects in their study may also be explained by the shorter time interval between injections. After 2 weeks, the lymph nodes are more likely to still be inflamed because of the aluminum-based adjuvant in the extract, increasing the risk for side effects.

ILIT is a highly potent route for immunization and immunotherapy. This is supported by close to 100 studies using protein- and peptide-based vaccines, naked DNA and RNA vaccines, BCG, and adjuvants. These studies have been performed in mice, dogs, monkeys, cats, ponies, and humans to treat cancer as well as infections, with the first studies dating back to the middle of the last century (reviewed in Senti et al¹¹). Nonetheless, we certainly concur with Witten et al that more and larger studies would be required to optimize protocols for ILIT in allergy. Such optimization should include the dose, the time intervals between injections, and the type of allergens used, that is, native extracts versus allergoids, peptides, modified recombinant molecules, and so on. Also, the types and doses of currently available adjuvants should be evaluated for use in ILIT. Finally, the use of ILIT in patients with asthma has still not been tested, and its applicability in children or juveniles has not been evaluated. Over the last several years, we have been searching for an industrial partner to further clinically develop this program. While we strongly believe that ILIT works, we have never stated that ILIT would be ready for “clinical use.”

Thomas M. Kündig, MD^a

Pål Johansen, PhD^a

Martin F. Bachmann, PhD^a

Lars O. Cardell, MD^b

Gabriela Senti, MD^c

From ^athe Department of Dermatology, University Hospital Zurich, Zurich, Switzerland; ^bthe Division of ENT Diseases, Department of Clinical Sciences, Intervention and Technology, Karolinska Institutet, Stockholm, Sweden; and ^cthe Clinical Trials Centre, University Hospital Zurich, Zurich, Switzerland. E-mail: thomas.kuendig@usz.ch.

Disclosure of potential conflict of interest: T. Kündig is named as an inventor on a patent on intralymphatic immunotherapy owned by the University of Zürich. The rest of the authors declare that they have no relevant conflicts of interest.

REFERENCES

1. Witten M, Malling HJ, Blom L, Poulsen BC, Poulsen LK. Is intralymphatic immunotherapy ready for clinical use in patients with grass pollen allergy? *J Allergy Clin Immunol* 2013;132:1248-52.e5.
2. Senti G, Prinz Vavricka BM, Erdmann I, Diaz MI, Markus R, McCormack SJ, et al. Intralymphatic allergen administration renders specific immunotherapy faster and safer: a randomized controlled trial. *Proc Natl Acad Sci U S A* 2008;105:17908-12.
3. Senti G, Cramer R, Kuster D, Johansen P, Martinez-Gomez JM, Graf N, et al. Intralymphatic immunotherapy for cat allergy induces tolerance after only 3 injections. *J Allergy Clin Immunol* 2012;129:1290-6.
4. Hylander T, Latif L, Petersson-Westin U, Cardell LO. Intralymphatic allergen-specific immunotherapy: an effective and safe alternative treatment route for pollen-induced allergic rhinitis. *J Allergy Clin Immunol* 2013;131:412-20.
5. Siegrist CA. Vaccine immunology. In: Plotkin SA, editor. *Vaccines* (6th ed.). Elsevier; 2013. pp. 14-32.
6. General recommendation on immunization. Recommendations of the Advisory Committee on Immunization Practices (ACIP). 2011. Available from www.cdc.gov. Accessed January 3, 2014.
7. Kedl RM, Kappler JW, Marrack P. Epitope dominance, competition and T cell affinity maturation. *Curr Opin Immunol* 2003;15:120-7.
8. Lovgren T, Baumgaertner P, Wieckowski S, Devere E, Guillaume P, Luescher I, et al. Enhanced cytotoxicity and decreased CD8 dependence of human cancer-specific cytotoxic T lymphocytes after vaccination with low peptide dose. *Cancer Immunol Immunother* 2012;61:817-26.
9. Patel D, Couroux P, Hickey P, Salapatek AM, Laidler P, Larche M, et al. 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;131:103-9.e1-7.
10. Guery JC, Galbiati F, Smiroldo S, Adorini L. Selective development of T helper (Th)2 cells induced by continuous administration of low dose soluble proteins to normal and beta(2)-microglobulin-deficient BALB/c mice. *J Exp Med* 1996;183:485-97.
11. Senti G, Johansen P, Kundig TM. Intralymphatic immunotherapy: from the rationale to human applications. *Curr Top Microbiol Immunol* 2011;352:71-84.

<http://dx.doi.org/10.1016/j.jaci.2013.11.036>