ISSN: 2577 - 8005

Original Article

Medical & Clinical Research

Evaluation of local nasal immunotherapy to Dermatophagoides sp. in patients with allergic rhinitis

Luiz Werber-Bandeira[1,2*], Thiago Luiz Bandeira[1], Isabela Mansur Bandeira[1], Selma Sabra[3], Aderbal Sabra Filho[4], Juliana Vasconcellos[1], Bruno de Brito Braga[1], Jorge Oliveira[1], Carla Goulart[1], Alexandre Monteiro[1], Aderbal Sabra[5]

[1]*Clinical and Experimental Immunology and Allergy Service, Santa Casa da Misericórdia do Rio de Janeiro/Imunoderm Clinic, Rio de Janeiro, RJ, Brazil.*

[2]*Estacio de Sa University / IDOMED. Rio de Janeiro, RJ, Brazil.*

[3]*Pediatric Endoscopy Service, Hospital Universitário Antônio Pedro, Universidade Federal Fluminense, Niteroi, Rio de Janeiro, RJ, Brazil.*

[4]*Universidade do Grande Rio, School of Medicine, Professor Jose de Souza, Rio de Janeiro, RJ, Brazil.*

[5]*Food Allergy and Autism Unit of the Clinical Immunology Service, Santa Casa da Misericórdia do Rio de Janeiro, Rio de Janeiro, RJ, Brazil.*

[*]Corresponding Author

Luiz Werber-Bandeira, Clinical and Experimental Immunology and Allergy Service, Santa Casa da Misericordia do Rio de Janeiro, Santa Luzia Street, 206-Centro, Rio de Janeiro, RJ, Brazil.

Submitted:24 Jan 2023;**Accepted**: 09 Feb 2023;**Published**:15 Feb 2023

Citation: *Luiz Werber-Bandeira, Thiago Luiz Bandeira, Isabela Mansur Bandeira, Selma Sabra, Aderbal Sabra Filho (2023) Evaluation of local nasal immunotherapy to* Dermatophagoides sp. *in patients with allergic rhinitis. Medical & Clinical Research 8(2): 01-07.*

Abstract

Introduction: *The aim of this study was to evaluate the clinical efficacy of allergen-specific nasal immunotherapy (LNIT) by observing the improvement in the patient's quality of life, and the side effects of this route of immunotherapy.*

Methods: *From a cohort of 2687 patients with perennial rhinitis treated at the Clinical and Experimental Immunology Service-Hospital Geral-Santa Casa da Misericórdia do Rio de Janeiro for 5 years, a total of one hundred thirty six patients positive in the prick test for Dermatophagoides pteronyssinus (Dp) and Dermatophagoides farinae (Df), were divided into two groups of cases (n=108) and controls (n=28) undergoing or not LNIT, both submitted to the same type of control treatment for the same amount of time. Both groups were classified into mild, moderate and severe rhinitis. Quality of life was assessed based on a questionnaire for Rhinoconjunctivitis, carried out at each visit, with a rating from 0 to 6 according to the increasing degree of difficulty in performing common tasks or nasal, ocular or other symptoms.*

Results: *No systemic side effects or bronchospasms were observed in the cases. Both patients and controls with moderate and severe rhinitis had quality of life grades 3 and 4; 5 and 6 respectively, before immunotherapy. Comparison of cases with controls during the controller medication phase associated with nasal immunotherapy (cases) showed an improvement in quality of life for both (Grades: 0-2, after 5 weeks). Full use of the controller medication was 15 weeks followed by more eighteen weeks with half doses. Patients under LNIT, when the control medication was withdrawn after the sixth series of nasal immunotherapy, maintained the improvement in quality of life with grades of 0-1, not requiring regular and frequent use of symptomatic therapy. Until the final evaluation time, three years and two months, the patients who remained until the end of the immunotherapy regimen (n=89) did not present or significantly reduced the need for control medication, remaining with a degree of quality of life: Degree: 0 and 1.The controls, in the period of 33 weeks of return for consultations, with the withdrawal of the controller medication, reported that they needed the frequent use of controller medicines due to the recurrence of symptoms. The quality of life questionnaire showed a worsening, with grades ranging from 3 to 5, when evaluated in this phase without regular symptomatic medication.*

Conclusions: *The of LNIT performed with full concentrations, did not show secondary reactions with risks to patients and that the effect of inducing tolerance to the antigens of Dermatophagoides sp. was achieved, based on the observation of the decrease in the use of control medications for signs and symptoms and mainly by the improvement in the patients' quality of life.*

Keywords: Immunotherapy, Allergic Rhinitis, Allergen, Dermatophagoides pteronyssinus, Dermatophagoides farinae

Introduction

Allergic rhinitis (AR) is the IgE-mediated inflammatory disease that affects mainly the nasal mucosa.AR is usually classified according to etiology into Seasonal AR (SAR), induced periodically by airborne allergens, such as grass, tree or weed pollen or Perennial AR (PAR) in response to persistently present indoor allergens such as house dust mite (HDM), the major source of aeroallergens for patients with AR, animal dander, insects according to severity, assessed by frequency and intensity of symptoms into severe AR, moderate AR, and mild AR [1-3]. AR is a highly prevalent disease and can also be considered the most common type of respiratory disease affecting 40% of population worldwide with an increasing prevalence [4,5]. In Brazil, it affects around ¼ of the population [6].

The treatment of AR is based in a pharmacotherapy that include antihistamines and local corticosteroids as the first line that are normally effective for controlling symptoms and suppress inflammation [7,8]. However, in cases of uncontrolled symptoms despite of medication and allergen avoidance allergen-specific immunotherapy (AIT), is indicated [9-11]. AIT, a personalized treatment approach for the allergic airway disease is associated with a reduction of symptoms, a reduction in the use of rescue medications, improvement of quality of life and allergen-specific immune tolerance [12-18].

Still today, in spite of AIT effectiveness, a lot of discussion about some critical points such as the duration of the whole therapy, the time of remission after discharge, the use of one or multiple allergens, the ideal amount of protein in the antigens preparation, the administration routes, the percentage of efficacy and safety, the ways of using immunotherapy and its limitations still remain [11]. It is of interest to achieve and standardize allergen preparations, with ideal concentration of proteins, and the most suitable administration route, with less local or systemic adverse reactions, that allow patients to more easily adhere to treatment. The objective of AIT is an allergen-specific immunomodulation, leading to a specific tolerance, through the control of Th2CD4+ lymphocytes; inducers of IgE production and inflammatory substances in allergic-atopic individuals mainly through induction of various functional regulatory cells such as regulatory T cells (Tregs), follicular T cells (Tfr), B cells (Bregs), dendritic cells (DCregs), innate lymphoid cells (IL-10^{+} ILCs), and natural killer cells (NKregs) [19].

Use of AIT involves administration of repeated high-doses of allergens for at least 3 years to confer clinical benefits. The administration of allergens for AIT can be done by different injectable (subcutaneous (SCIT), until recently, used as the standard administration route for AIT, except for the rush protocol where increase of the risk of anaphylaxis was observed, epicutaneous, or non-injectable (sublingual (SLIT), ORAL (OIT), bronchial (LBIT) and nasal (LNIT) routes [10, 20-22]. Being SCIT and SLIT the most commonly used routes in spite of their differences, indirect comparisons are made and the results are diverging depending on the study design, patients, schedules, and other aspects. For example, SCIT showed to be effective for children and adults with AR. Comparisons using Meta-analysis methodology revealed both to be effective for SAR but not for PAR with HDM where only SICT confirmed the efficacy [23]. Regarding safety SLIT showed to be safer than SCIT alternative [24].

In this context, other options of alternative non injection AIT route, has become available. The Local Nasal Immunotherapy (LNIT), conceived by Dunbar in 1913 and investigated since the 1970s by researchers from United States and later in Italy, proved to be effective in desensitizing children with symptoms of rhinitis and reducing medication consumption, as well as decreasing allergen-specific nasal reactivity after 18 months of immunotherapy without significant side effects, but with local adverse events [25,26]. Today, several advances are being considered including new intranasal delivery systems that overcome limitations mainly of difficulties in dosing and local adverse events.

It is interesting that, since the nasal route is easily accessible, the study of eosinophilic cell dynamics and the local presence of specific IgE, IgG4 antibodies could be performed without difficulties, being also evaluators of the immunotherapy efficacy. These parameters, associated with the improvement or worsening of the patients' quality of life, would provide data to guide the ideal concentrations of allergens introduced into the nasal mucosa, evaluating, in this case, the presence or absence of the specific IgE that triggers the allergic mechanisms in the nasal mucosa [27]. Together, important information would be provided, in a non-invasive, easy-to-perform way, both to find the ideal immunotherapy concentrations for each patient, to avoid inducing risks to patients undergoing treatment, and to evaluate the clinical efficacy of nasal allergen immunotherapy considering that it is well known that *Dermatophagoides pteronyssinus* (Dp) and *Dermatophagoides farinae* are among the most frequent house dust mites clinically important and able to sensitize patients [28-31].

The objective of this study was to evaluate the clinical efficacy of allergen-specific nasal immunotherapy (LNIT) for AR induced by Dermatophagoides sp. (Der p and Der f) by observing the improvement in the patient's quality of life, the reduction of AR symptoms and control medication requirements, the side effects and its safety with full concentrations of allergens.

Materials and Methods

Study Design and Patients Selection

This is an open interventionist study. The patients enrolled in this study originally belong to a cohort of 2687 patients with perennial Allergic rhinitis (AR) treated at the Clinical and Experimental Immunology Service-Hospital Geral-Santa Casa da Misericórdia do Rio de Janeiro for 5 years. Eligibility and enrollment were based in the inclusion, (perennial rhinitis and single positive skin prick test responses only to *Dermatophagoides pteronyssinus* (Dp) and *Dermatophagoides farinae* (Df) and exclusion criteria, (patients with severe systemic diseases, such as diabetes mellitus

and hypertension, or using any medication capable of affecting sensitivity to the allergen). A total of 136 subjects comprising 65 male and 71 females were selected and enrolled after signing an informed consent. Patients responded to a standard clinical questionnaire and were divided into two groups being cases, undergoing LNIT (n=108) and controls, not undergoing LNIT (n=28). Controls were patients removed from this total who, for personal reasons, did not wish to undergo immunotherapy. Both groups were submitted to the same type of control treatment for the same amount of time and were classified into mild, moderate and severe AR. The effects of interventions on Health-Related Quality of Life (HRQOL) of patients was assessed according to the validated questionnaire carried out at each visit, with a rating ranging from 0 to 6 according to the increasing degree of difficulty in performing common tasks or nasal, ocular or other symptoms [32-34].

Allergen Preparation and Skin Prick Test

The allergen concentrate was produced by Alk-Abello A/S Horsholm, Denmark, with 130 µg/mL of Dermatophagoides pteronyssinus (Der p) and 140 µg/mL of Dermatophagoides farinae (Der f). With effective therapeutic dose ranging from 7 to 15 µg per month, during the maintenance period. The tests were performed by puncture with reading after 15 mm, in the 1/10 dilution, weight/volume, of the concentrate. The positive test pattern: histamine 1mg/mL and the negative test pattern: 50% glycerin phenolic saline solution was used. A positive result, representing IgE sensitization, is a papule diameter that is 3 mm larger than the diameter of the negative pattern. Routine international protocol conduct.

All research subjects, patients and controls were placed without nasal medication for thirty days prior to the start of work. They were followed up and only used oral antiallergic medication when necessary.

Sensitization-LNIT Conduction

The method used was the intranasal application of the allergen, by spray of 0.1cc per jet diluted from the concentrate for different periods depending on the series. Two evaluators measured the amount expelled by the spray bottle separately.

The whole protocol involved 19 series of sensitizations with different concentrations of the allergen as described in detail in figure 1. The medication to control symptoms, also administered locally, was fluticasone 50 µg, 2 sprays in each nostril, once a day and azelastine 0.14 mg, 1 spray in each nostril twice a day. The full dose of these medications was for 15 weeks, considered First Phase, stage 1a, and then reduced to half for another 18 weeks; First Phase, stage 1b. After the two stages from phase I. For the second phase of LNIT lasted from 7th to 13th series controlling drugs were removed and only used if necessary. In this phase, only the specific allergen Nasal Immunotherapy was used under environmental control rules.

Clinical and quality of life assessments were performed at each new consultation, initially every five weeks until reaching a return every four months, all times with concentration of immunotherapy. During the first consultation and the other clinical evaluations, quality of life questionnaires were performed.

The Third Phase of the LNIT, considered the Maintenance and Reinforcement phase, consisted of six more series of the specific allergen. The total treatment lasted 38 months as recommended by the *American Academy of Allergy* Asthma & Immunopathology.

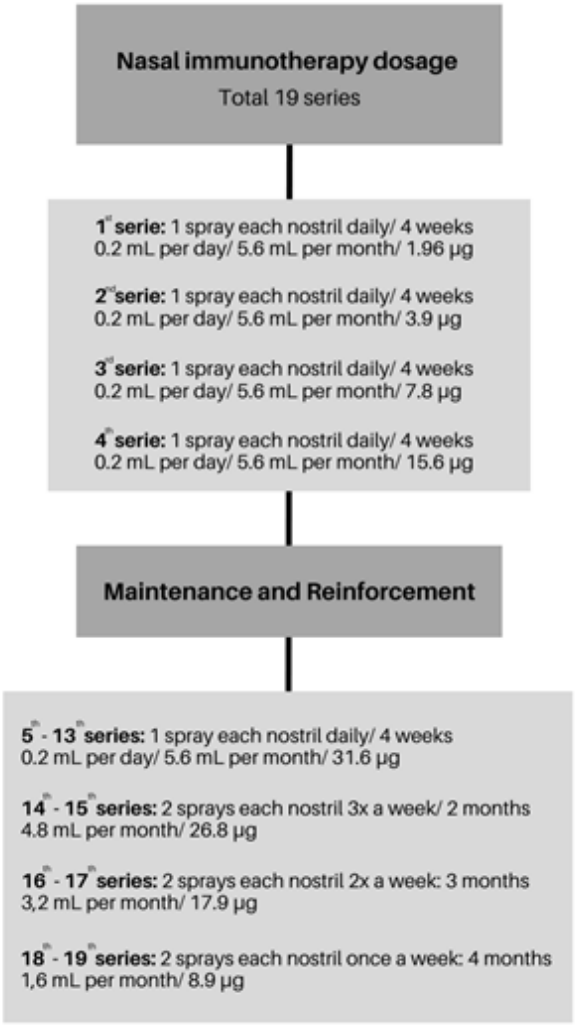

Figure 1: LNIT scheme used in the study.

Results

The clinical and demographic characterizations of the research subjects by group are described in table 1. Both (cases and controls) were clinically classified as having mild, moderate or severe rhinitis before AIT.

Table 1: Classification of cases and controls according to the severity of the disease.

	Cases (n=108)	Controls (n=28)
Males	54	11
Moderate Rhinitis	44	6
Quality of Life	3 - 4	3 - 4
Severe Rhinitis	10	5
Quality of Life	5-6	5-6
Females	54	17
Moderate Rhinitis	38	10
Quality of Life	3-4	3-4
Severe Rhinitis	16	7
Quality of Life	5-6	5-6

No systemic side effects or bronchospasms were observed in the cases during or after AIT. Five patients had mild nasal symptoms and signs, effectively when the vial was changed for a higher concentration, being controlled with the use of systemic antihistamines. The symptoms disappeared quickly as the immunotherapy continued; there was no need to discontinue it. Both, patients and controls with moderate and severe rhinitis had quality of life grades 3 and 4; 5 and 6 respectively, before immunotherapy. Comparison of cases with controls during the controller medication phase associated with nasal immunotherapy (cases) showed an improvement in quality of life for both (Grades: 0-2, after 5 weeks). Full use of the controller medication was for 15 weeks followed by more 18 weeks with half doses.

Patients under LNIT, when the control medication was withdrawn after the sixth series, maintained the improvement symptoms, without rhinorrhea, stuffiness, sneezing and nasal itching, that is, in response to the quality of life questionnaire they ranged from Grade 0 to 1, not requiring regular and frequent use of symptomatic therapy.

Eight patients showed signs and symptoms when they were subjected to extreme contact with house dust mites, or those who started using feather/down pillows, despite being instructed to use polyester pillows with replacement every three months.

Finally, until the final evaluation time, three years and two months, the patients who remained until the end of the immunotherapy regimen (n=89) did not present symptoms and significantly reduced the need for control medication, remaining with a degree of quality of life ranging from 0 to 1. The controls, in the period of 33 weeks of return for consultations, with the withdrawal of the controller medication, reported that they needed the frequent use of controller medicines due to the recurrence of symptoms. The quality of life questionnaire showed a worsening, with grades ranging from 3 to 5, when evaluated in this phase without regular symptomatic medication.

Discussion

The universe of immunotherapy is wide and controversial, not only because of possible adverse effects, especially the systemic ones, but because of doubts about clinical efficacy. The treatment time is long, an average of three years, and the classic introduction is injectable, reducing the patient's adherence to the treatment, and therefore, the search for other routes is essential.

Although the effectiveness of AIT for AR has been shown, assessing this effectiveness over the years has not been a simple task [35]. Several studies have already been carried out in different populations as well as numerous meta-analyses, but the number of parameters involved and problems such as ethnic heterogeneity of the populations studied, the different allergen products and protocols used, and the clinical outcomes used to document efficacy and safety makes this analysis difficult, which ends up being done separately. However, although with variable effectiveness, depending on the different factors, such as administration routes, allergen preparation, treatment duration, etc, AIT has been shown to be useful after administration for 3 to 4 years not only for patients with seasonal rhinitis but also in perennial allergy caused by house dust mites [26, 36-44].

In this study, we specifically evaluated the clinical efficacy of specific local allergen nasal immunotherapy, used in the routine of the Immunology and Allergy Clinical and Experimental Service of Santa Casa de Misericórdia do Rio de Janeiro in patients with AR by comparing it with patients who did not use it.

The nasal mucosa is considered to be the entry point for numerous pathogens and since a large number of lymphoid organs are located in this area, LNIT has been considered a favorable method to trigger immune tolerance, especially when targeting a single immunodominant peptide from an allergen [45]. Furthermore, LNIT is less invasive than injection immunotherapy, with fewer systemic reactions, presents the convenience of the route of introduction and, for this reason, could result in greater adherence to the treatment in any age, as well as offering a lower risk of adverse reactions to the patient when compared to the subcutaneous route. It is of interest that, as the nasal route is easily accessible, the study of eosinophilic cell dynamics, the local presence of specific antibodies IgE, IgG4 could be carried out without difficulties,

being evaluators of the effectiveness of immunotherapy. These parameters, associated with the improvement or worsening of the patients' quality of life, would provide data that would guide the ideal concentrations of allergens introduced into the nasal mucosa, assessing, in this case, the presence or absence of specific IgE that trigger allergic mechanisms in the mucosa nasal. Together, important information can be provided, in a non-invasive way, both for determining the ideal immunotherapy concentrations in a personalized way for each patient, avoiding risks, and for evaluating the clinical efficacy of LNIT.

Our results corroborate the recently data from the literature evaluating the LNIT for AR. In a systematic review and Meta analysis a pooled data collected from a total of 20 studies involving almost 700 patients. Results obtained from these pooled data clearly demonstrate the immunological efficacy and safety of LNIT for AR was demonstrated by the observation of improvement in the presentation of clinical symptoms and reduction in the use of control medication [46].

In our assessment, there were also no systemic or important local symptoms that would prevent the use of nasal immunotherapy. The reason for the withdrawal of patients who did not fully perform our evaluation, for those we were able to contact, was the long treatment time, about three (3) years. This time was chosen to follow the international consensus [18, 47-49]. Data from the literature on LNIT using aqueous extracts have shown that symptoms of rhinitis appear with high doses whereas low doses are well tolerated but lack of clinical efficacy [26, 50, 51]. In our study we used the maximum possible concentration of the antigen, starting daily and finally weekly, with gradual concentrations, the final dose being around 8.9 µg/month, based on what is recommended by the producer for subcutaneous immunotherapy (ALK-Abello) without causing side effects, therefore, as international Immunotherapy protocols seek to use the most concentrated monthly doses possible, without inducing side effects, we could conclude that we have achieved this important objective [50-53]. Suboptimal doses are considered clinically ineffective, that is, they would not induce tolerance.

The clinical efficacy of this study was proven mainly when compared to the control group, during the moment of withdrawal of controller medications, the controls, who did not use immunotherapy, showed a return of symptoms, while the patients, who underwent immunotherapy, when using only of nasal immunotherapy, remained without control medication or had it rarely, remaining in Grade: 0 and 1 of quality of life. Controls returned to grades 3, 4 and 5.

Studies carried out in adults allergic to Dermatophagoides report clinical improvement in patients after six months of therapy [38]. In our evaluation, rhinitis symptoms were significantly reduced after 8 months and remained so until the end of the monotherapy - the nasal local allergen specific immunotherapy, for 3 years.

Conclusion

The present work shows that the results of LNIT performed with full allergen concentrations, did not show secondary reactions with risks to patients and that the effect of inducing tolerance to the antigens of *Dermatophagoides sp*. would have been achieved, based on the observation of the decrease in the use of control medications for signs and symptoms and mainly by the improvement in the patients' quality of life. Several studies are assessing this route for AIT and the conclusions are that the nasal mucosa has the capacity to absorb allergenic molecules, which remain antigenically active in the bloodstream [26, 54]. As bronchospasm was not induced at any time, the assessment of LNIT in patients with allergic asthma may represent an innovative and promising study for this type of disease.

Ethical Aspects

All procedures for enrollment and conduction of this research project was reviewed and approved by the Institutional Review Board of the General Hospital of Santa Casa da Misericórdia do Rio de Janeiro

Funding

This study was financial supported by IMUNODERM Clinic. This research received no external funding.

Conflict of Interest

Authors have declared that no competing interests exist.

Acknowledgement

We thank the Health and Sustainability Association – ASAS by the administrative and technical support.

References

1. Bousquet J,Khaltaev N, Cruz A, Denburg J, Fokkens W, et al. (2008) Allergic Rhinitis and its Impact on Asthma (ARIA) 2008 update (in collaboration with the World Health Organization, GA(2)LEN and AllerGen). Allergy 63(86):8-160.
2. Brożek JL, Bousquet J, Agache I, Agarwal A, Bachert C, et al. (2017) Allergic Rhinitis and its Impact on Asthma (ARIA) guidelines-2016 revision. J Allergy ClinImmunol 140:950-958.
3. Muraro A, Lemanske RF, Hellings PW, Akdis CA, Bieber T, et al. (2016) Precision medicine in patients with allergic diseases: airway diseases and atopic dermatitis-PRACTALL document of the European Academy of Allergy and Clinical Immunology and the American Academy of Allergy, Asthma & Immunology. J Allergy ClinImmunol 137:1347-1358.
4. Passali D, Cingi C, Staffa P, Passali F, Muluk NB, et al. (2018) The International Study of the Allergic Rhinitis Survey: outcomes from 4 geographical regions. Asia Pac Allergy 8(1):e7.
5. https://jornal.usp.br/atualidades/rinite-alergica-afeta-cerca-de-40-da-populacao-mundial/
6. https://fbh.com.br/rinite-alergica-atinge-1%E2%81%844-da-populacao-dos-brasileira-afirmam-especialistas-do-hospital-

paulista/
7. Bousquet J, Schunemann H, Samolinski B, Demoly P, Baena-Cagnani C, et al. (2012) Allergic Rhinitis and its Impact on Asthma (ARIA): achievements in 10 years and future needs. J Allergy Clin Immunol 130:1049-1062.
8. Wheatley L, Togias A (2015) Clinical practice Allergic rhinitis. N Engl J Med 372:456-463.
9. Matsuoka T, Shamji MH, Durham SR (2013) Allergen immunotherapy and tolerance. Allergol Int 62:403-413.
10. Komlosi ZI, Kovacs N, Sokolowska M, Van de Veen W, Akdis M, et al. (2020) Mechanisms of subcutaneous and sublingual aeroallergen immunotherapy: what is new? Immunol Allergy Clin North Am 40:1-14.
11. Kucuksezer UC, Ozdemir C, Cevhertas L, Ogulur I, Akdis M, et al. (2020) Mechanisms of allergen-specific immunotherapy and allergen tolerance. Allergol Int 69:549-560.
12. Calderon M, Alves B, Jacobson M, Hurwitz B, Sheikh A, et al (2007) Allergen injection immunotherapy for seasonal allergic rhinitis. Cochrane Database Syst Rev 1:CD001936.
13. Compalati E, Braido F, Canonica G (2014) An update on allergen immunotherapy and asthma. Curr Opin Pulm Med 20:109-117.
14. Malling H, Bousquet J (2008) Subcutaneous immunotherapy for allergic rhinoconjunctivitis, allergic asthma, and prevention of allergic diseases. Clin Allergy Immunol 21:343-358.
15. Burks A, Calderon M, CasaleT, Cox L, Demoly P, et al. (2013) Update on allergy immunotherapy: American Academy of Allergy, Asthma & Immunology/European Academy of Allergy and Clinical Immunology/PRACTALL consensus report. J Allergy Clin Immunol 131:1288-1296.
16. Canonica G, Cox L, Pawankar R, Baena-Cagnani C, Blaiss M, et al. (2014) Sublingual immunotherapy: World Allergy Organization position paper 2013 update. World Allergy Organ J 7:6.
17. Bousquet J, Lockey R, Malling H (1998) Allergen immunotherapy: therapeutic vaccines for allergic diseases. A WHO position paper. J Allergy Clin Immunol 102:558-562
18. Durham S, Walker S, Varga E, Jacobson M, O'brien F, et al. (1999) Long-term clinical efficacy of grass-pollen immunotherapy. N Engl J Med 341:468-475.
19. Boonpiyathad T, Satitsuksanoa P, Akdis M, Akdis CA (2019) Il-10 producing T and B cells in allergy. SeminImmunol 44:101326.
20. Passalacqua G, Canonica GW (2016) Allergen immunotherapy: history and future developments. Immunol Allergy Clin North Am 36:1-12.
21. Feng S, Xu Y, Ma R, Sun Y, Luo X, et al. (2014) Cluster subcutaneous allergen specific immunotherapy for the treatment of allergic rhinitis: a systematic review and meta-analysis. PLoSONE 9:e86529.
22. Scheurer S, Toda M (2017) Epicutaneous immunotherapy. Allergologia et Immunopathologia 45:25-29.
23. Durham SR, Penagos M (2016) Sublingual or subcutaneous immunotherapy for allergic rhinitis? J Allergy Clin Immunol 137:339-349.
24. Sharif H, Singh I, Kouser L, et al. (2019) Immunologic mechanisms of a short-course of Lolium perenne peptide immunotherapy: A randomized, double-blind, placebo - controlled trial. J Allergy Clin Immunol 144:738-749.
25. Dunbar WP (1913) The present state of knowledge of hayfever. J Hyg 13:105.
26. Marcucci F, Sensi LG, Caffarelli C, Cavagni G, Bernardini R, et al. (2002) Low-dose local nasal immunotherapy in children with perennial allergic rhinitis due to Dermatophagoides. Allergy 57:23-28.
27. Shamji MH, Valenta R, Jardetzky T, Verhasselt V, Durham SR, et al. (2021) The role of allergen-specific IgE, IgG and IgA in allergic disease. Allergy 76:3627-3641.
28. Arlian LG, Morgan MS (2003) Biology, ecology, and prevalence of dust mites. Immunol Allergy Clin North Am 23:443-468.
29. Fernandez-Caldas E, Iraola V, Carnes J (2007) Molecular and biochemical properties of storage mites (except Blomia species). Protein Pept Lett 14:954-959.
30. Tsai JJ, Shen HD, Chua KY (2000) Purification of group 2 Dermatophagoides pteronyssinus allergen and prevalence of its specific IgE in asthmatics. Int Arch Allergy Immunol 121:205-210.
31. Liao EC, Ho CM, Lin MY, Tsai JJ (2010) Dermatophagoides pteronyssinus and Tyrophagus putrescentiae allergy in allergic rhinitis caused by cross-reactivity not dual-sensitization. J ClinImmunol 30:830-839.
32. Juniper EF, Thompson AK, Ferrie PJ, Roberts JN (2000) Development and Validation of the Mini Rhinoconjunctivitis Quality of Life. Clinical and Experimental Allergy 30:132-140.
33. Thompson AK, Juniper E, Meltzer EO (2000) Quality of life in patients with allergic rhinitis. Ann Allergy Asthma Immunol 85(5):338-347.
34. Radulovic S, Wilson D, Calderon M, Durham S (2011) Systematic reviews of sublingual immunotherapy (SLIT). Allergy 66:740-752.
35. Nurmatov U, Dhami S, Arasi S, Roberts G, Pfaar O, et al. (2017) Allergen immunotherapy for allergic rhinoconjunctivitis: a systematic overview of systematic reviews. ClinTransl Allergy 7:24.
36. Nickelsen JA, Goldstein S, Mueller U,Wypych J, Reisman RE, et al. (1981) Localintranasal immunotherapy for ragweed allergicrhinitis: clinical response. J Allergy ClinImmunol 68(1):33-40.
37. Welsh PW, Zimmermann EM, Yunginger JW, Kern EB, Gleich GJ (1981) Preseasonal intranasal immunotherapy with nebulized short ragweed extract. J Allergy ClinImmunol 67(3):237-242.
38. Andri L, Senna GE, Betteli C, Givanni S, Andri G, et al. (1992) Local nasal immunotherapy in allergic rhinitis to Parietaria. Allergy 47:318-323.
39. Andri L, Senna GE, Betteli C, Givanni S, Andri G, et al. (1993) Local nasal immunotherapy for Dermatophagoides-induced rhinitis: efficacy of a powder extract. J Allergy Clin

Immunology 91:587-596.
40. Passalacqua G, Albano M, Ruffoni S,Pronzato C, Riccio AM, et al. (1995) Local nasal immunotherapy to Parietaria: evidence of reduction of allergic inflammation. Am J RespirCrit Care Med 152:461-466.
41. Fanales-Belasio E, Ciofalo A, Zambetti G,Ansotegui IJ, Scala E, et al. (1995) Intranasal immunotherapy with Dermatophagoides extract: in vivo and in vitro results of a double-blind placebo-controlled trial. Rhinology 33:126-131.
42. Gaglani B, Borish L, Bartelson BL, Buchmeier A, Keller L, et al (1997) Nasal immunotherapy in weed-induced allergic rhinitis. Ann Allergy Asthma Immunol 79:259-265.
43. Motta G, Passali D, De Vincentiis I, Ottaviani A, Maurizi M, et al. (2000) A multicenter trial of specific local nasal immunotherapy. Laryngoscope 110:132-139.
44. Passali D, Bellussi L, Passali GC, Passali FM (2002) Nasal immunotherapy is effective in the treatment of rhinitis due to mite allergy. A double-blind, placebo-controlled study with rhinological evaluation. Int J Immunopathol Pharmacol 15:141-147.
45. Liu YH, Tsai JJ (2005) Production of salivary immunoglobulin A and suppression of Dermatophagoides pteronyssinus-induced airway inflammation by local nasal immunotherapy. Int Arch Allergy Immunol 138(2):161-168.
46. Kasemsuk N, Ngaotepprutaram P, Kanjanawasee D, Suwanwech T, Durham SR, et al. (2022) Local nasal immunotherapy for allergic rhinitis: A systematic review and meta-analysis. Int Forum Allergy Rhinol 12:1503-1516.
47. Cools M, Van Bever HP, Weyler JJ, Stevens WJ (2000) Long-term effects of specific immunotherapy, administered during childhood, in asthmatic patients allergic to either house-dust mite or to both house dust mite and grass pollen. Allergy 55:69-73.
48. Eng PA, Borer-Reinhold M, Heijnen IA, Gnehm HP (2006) Twelve-year follow-up after discontinuation of pre seasonal grass pollen immunotherapy in childhood. Allergy 61:198-201.
49. Tabar AI, Arroabarren E, Echechipia S, Garcia BE, Martin S, et al. (2011) Three years of specific immunotherapy may be sufficient in house dust mite respiratory allergy. J Allergy Clin Immunol 127:57-63.
50. Welsh PW, Butterfield JH, Yungingerb JW, Agarwalrwal MK, Gleich GJ (1983) Allergen controlled study of intranasal immunotherapy for ragweed hay fever. J Allergy ClinImmunol 71:454-460.
51. Wypich JI, Georgitis JW, Tronolone MJ (1984) Local nasal immunotherapy (LNIT) comparing polymerized and aqueous grass extract (Abstract). J Allergy ClinImmunol 73:140.
52. Cox L, Cohn JR (2007) Duration of allergen immunotherapy in respiratory allergy: when is enough, enough? Ann Allergy Asthma Immunol 98:416-426.
53. Georgitis JW, Nickelsen JA, Kane JH, Reisman RE (1985) Local nasal immunotherapy: efficacy of low dose aqueous extract. J Allergy ClinImmunol 75:496-500.
54. Mistrello G, Rapisarda G, Falagiani P (1991) Detection of IgE-binding activity in serum after intranasal treatment of normal rabbits with Parietaria judaica extract.Allergy 46:52-58.