

Assessment of Quality of Life Among Patients with Respiratory Allergy, Receiving Sublingual and Subcutaneous Immunotherapy

B. G. Parasuramalu¹, Gangaboraiah¹, R. Balaji¹ and G. M. Someswara²

Department of Community Medicine, Kempegowda Institute of Medical Sciences Hospital and Research Centre¹, Bengaluru and Vijaynagar Institute of Medical Sciences², Bellary, Karnataka.

Abstract

The objective of the present study was to assess the effects of sublingual immunotherapy (SLIT) on quality of life compared to rush subcutaneous immunotherapy (SCIT) among patients suffering from respiratory allergy. The study was conducted from January 2007 to August 2008 and 50 patients with convincing history, positive skin prick test to one or more allergen extracts were recruited, 25 for SLIT and rush SCIT using randomisation technique. Immunotherapy was conducted with antigens (mix), namely dust mites, tree pollens, grass pollens and weed pollens in patients of allergic rhinitis, asthma or both for one year. There was significant improvement in quality of life (QoL) in both SLIT and rush SCIT groups from base line to end of one year. The difference in QoL between the two groups was not found to be statistically significant. Both SLIT and rush SCIT were equally efficacious in improving the QoL among patients with respiratory allergies.

Key words: Sublingual immunotherapy, Subcutaneous (rush) immunotherapy, Quality of life, Skin prick test.

INTRODUCTION

Respiratory allergy is a global public health problem projected to affect 50% of population in future. If the current trends were to continue, it may be an epidemic of the 21st Century.¹ Restriction of physical activity, absenteeism from work due to allergic manifestations is a major factor for loss of productivity, thus lowering quality of life (QoL) and results in socio-economic burden.^{2,3} Respiratory allergies are responsible for 3.5 million lost work-days annually in the United States.⁴ The commonest manifestations of respiratory allergies include allergic rhinitis and allergic bronchial asthma.

The main strategy in the management includes patient's education, environmental control, pharmacotherapy and immunotherapy. Immunotherapy is gaining more importance than pharmacotherapy because of its long lasting effect. There are various types of immunotherapies viz. sublingual, subcutaneous, intranasal and oral. Immunotherapy reduces the symptoms and use of relief medications thereby improving patient's QoL within the first treatment year. In contrast with symptomatic treatment with anti-allergic drugs, immunotherapy is the only available therapy that treats the underlying cause of the disease with proven long-term benefits.⁵⁻⁸

Sublingual immunotherapy (SLIT) has a favourable safety profile than the injection route, thereby enhancing the benefit-risk ratio in favour of a broader indication for allergen immunotherapy for use in the patient's home.⁵ Previous studies did not show on QoL compared

Correspondence and reprint requests: Dr B.G. Parasuramalu, Professor and Head, and In-charge Allergy Centre, Department of Community Medicine, Kempegowda Institute of Medical Sciences Hospital and Research Centre, K.R. Road, Bengaluru - 560 004, India; Phone: 9986003467 Email: bgparasuramalu@yahoo.co.in

to subcutaneous immunotherapy (SCIT) among patients suffering from respiratory allergies⁹. Benefit of conventional SCIT schedule consists of once or twice weekly injections usually achieving a maintenance dose after 4 to 6 months, whereas in rush immunotherapy the maintenance dose can be achieved in two days. It could reduce cost, time, and discomfort to the patient and potentially increase compliance. Further, it was shown that pre-medication reduces the incidence of systemic reactions during rush immunotherapy.¹¹ Because of advantages of rush SCIT over conventional SCIT, in the present study we used rush SCIT to compare with SLIT on the QoL of the patients.

MATERIAL AND METHODS

The present study was conducted in Allergy Centre, Department of Community Medicine, Kempegowda Institute of Medical Sciences Hospital and Research Centre, Bangalore, Karnataka, from January 2007 to August 2008. Fifty patients in the age group of 10-60 years suffering from respiratory allergy (allergic rhinitis, asthma or both) who were found positive on skin prick test were included. The clinical diagnosis of allergic rhinitis and asthma was made according to ARIA and GINA guidelines,¹³ respectively. Skin prick tests (SPTs) were performed with 123 allergen extracts. The extracts included 19 pollens, 5 dusts, 2 dust mites, 10 fungi, 10 insects, 3 epithelia and 74 food allergens. Allergen extracts for SPTs were obtained from Creative Drug Industries, Navi Mumbai. Pollen antigens were selected based on pollen calendar.

The SPTs were performed on the flexor side of upper arm and forearm after cleaning with isopropyl alcohol. A single drop of each allergen extract of 1:10 v/v concentration was placed along with positive control (Histamine) di-phosphate 5mg/mL and negative control 50% glycerinated phosphate buffer (saline).

Sterile lancet was used to prick by making a shallow lift (at an acute angle) for a second to allow entry of antigen beneath the stratum corneum epidermis. This was repeated for each allergen after wiping the lancet with dry cotton. The allergen was left in place for 20 minutes then the reading of wheal size of allergen, saline and histamine was taken with the help of a measuring scale.^{14,15} Grading of skin reactions was done according to criteria of Agarwal and Co-workers¹⁶ and skin

reactions with wheal size more than 2+ (grade 2 and above) were considered as marked positive reactions.

Twenty-five patients were administered swallow SLIT and rush SCIT using randomisation technique and followed for one year. Patients with severe asthma, high blood pressure, ischemic heart disease, pregnancy, immune disorders, history of anaphylaxis and those receiving immunosuppressive medications or beta-blockers were excluded from the study. The protocol was approved by the Institutional Ethics Committee. The selected therapy was administered to patients by explaining the details of the therapy and after obtaining an informed written consent from the patients.

Procedure for Sublingual Swallow Immunotherapy^{17,18}

Customised vaccine kits for SLIT were procured from Creative Drug Industries, Navi Mumbai based on the allergen profile of the patients. Vaccines were in 1:99 concentration (vial 3), from which dilutions of 1:999 (vial 2) and 1:9999 (vial 1) were prepared. Therapy was started with two drops of vial 1 diluted in 10 mL of water administered sublingually. Patient was instructed to keep the vaccine below the tongue for two minutes before swallowing. The dose was increased by two drops once in three days and the patient was followed-up every 4 weeks. Similar schedule was followed for vial 2 and vial 3 and maintenance dose was fixed according to patient's tolerance.

Procedure for Subcutaneous (rush) immunotherapy^{11,19}

Patients were examined one week prior to rush immunotherapy to obtain baseline peak expiratory flow rate (PEFR), general physical assessment and to provide education. All received pre-medications (Prednisolone-20 mg, Loratadine-10 mg once daily and Ranitidine -150 mg, the previous day and during two days of the procedure. These patients were admitted on the first day in the morning, to permit time for physical examination, PEFR and placement of intravenous line. Procedure was done in the presence of a crash cart equipped with all emergency medicines like epinephrine, oxygen, intubation tube, ambu bag etc. Before each injection, blood pressure, pulse rate, respiratory rate and PEFR were measured for all the patients and these were observed for two hours after the last injection on both the days.

Table: 1 Vaccines (antigens) used for Sublingual Immunotherapy in patients with respiratory allergy

S. No.	Patients	Assigned No.	Vaccines (antigen mix)
1	PJ	45	<i>D. pteronyssinus</i> , <i>D. ferinae</i>
2	AG	47	<i>Xanthium strumarium</i> , <i>Parthenium hysterophorus</i>
3	SBG	2	<i>D. pteronyssinus</i> , <i>D. ferinae</i>
4	VR	6	<i>Prosopis juliflora</i> , <i>D. pteronyssinus</i> , <i>D. ferinae</i>
5	US	8	<i>Prosopis juliflora</i> , <i>D. pteronyssinus</i> , <i>D. ferinae</i>
6	VA	10	House dust, <i>D. pteronyssinus</i> , <i>D. ferinae</i>
7	SBN	13	<i>Prosopis juliflora</i> , <i>Holopteflea integrifolia</i> , <i>Ailanthus excelsa</i> , <i>Casuarina equisetifolia</i>
8	SH	14	<i>D. pteronyssinus</i> , <i>D. ferinae</i>
9	UM	16	<i>D. pteronyssinus</i> , <i>D. ferinae</i>
10	AN	17	<i>Prosopis juliflora</i> , <i>Parthenium hysterophorus</i> , <i>Cynodon doctylon</i> , <i>Holopteflea integrifolia</i>
11	AN	27	<i>D. pteronyssinus</i> , <i>D. ferinae</i>
12	NK	26	<i>D. pteronyssinus</i> , <i>D. ferinae</i>
13	AR	22	<i>D. pteronyssinus</i> , <i>D. ferinae</i>
14	TA	21	<i>D. ferinae</i> , <i>D. pteronyssinus</i> , <i>Acacia arabica</i> , <i>Prosopis juliflora</i>
15	MM	19	<i>D. pteronyssinus</i> , <i>D. ferinae</i>
16	MM	28	<i>D. pteronyssinus</i> , <i>D. ferinae</i>
17	RO	30	<i>Prosopis juliflora</i> , <i>Ricinus communis</i> , <i>Parthenium hysterophorus</i> , <i>D. pteronyssinus</i> , <i>D. ferinae</i>
18	MD	33	<i>D. pteronyssinus</i> , <i>D. ferinae</i>
19	LK	35	<i>D. pteronyssinus</i> , <i>D. ferinae</i>
20	AS	37	<i>D. pteronyssinus</i> , <i>D. ferinae</i>
21	SBV	38	<i>D. pteronyssinus</i> , <i>D. ferinae</i>
22	CH	39	<i>Holopteflea integrifolia</i> , <i>Prosopis juliflora</i>
23	VA	44	<i>D. ferinae</i> , <i>D. pteronyssinus</i>

Table 2. Vaccines (antigens) used for Subcutaneous Immunotherapy in patients with respiratory allergy

S. No.	Patient	Assigned No.	Vaccines (antigen mix)
1	MS	12	<i>Parthenium hysterophorus</i> , <i>Cynodon dactylon</i> , <i>Xanthium strumarium</i> , <i>Cocos nucifera</i>
2	RJ	11	Cotton dust, <i>D. pteronyssinus</i>
3	SS	9	<i>D. pteronyssinus</i> , <i>D. ferinae</i>
4	SG	7	<i>Parthenium hysterophorus</i>
5	PY	4	<i>Cassia siamea</i> , <i>Amaranthus spinosus</i> , <i>D. ferinae</i> , <i>D. pteronyssinus</i>
6	LK	3	<i>D. pteronyssinus</i> , <i>D. ferinae</i> , <i>Proposis juliflora</i> , <i>Chenopodium album</i> , <i>Amaranthus spinosus</i>
7	CB	1	<i>Prosopis juliflora</i> , <i>Parthenium hysterophorus</i> , <i>D. pteronyssinus</i> , <i>D. ferinae</i>
8	US	43	<i>D. pteronyssinus</i> , <i>D. ferinae</i>
9	YN	46	<i>D. pteronyssinus</i> , <i>D. ferinae</i>
10	CG	48	<i>Typha angustata</i> , <i>Prosopis juliflora</i> , <i>Ailanthus excelsa</i> , <i>Dodonea viscosa</i> , <i>Parthenium hysterophorus</i>
11	LD	36	<i>Prosopis juliflora</i> , <i>Parthenium hysterophorus</i> , <i>D. pteronyssinus</i> , <i>D. ferinae</i>
12	NM	40	<i>D. pteronyssinus</i> , <i>D. ferinae</i>
13	ML	42	<i>D. pteronyssinus</i> , <i>D. ferinae</i>
14	ST	24	<i>Prosopis juliflora</i> , <i>Acacia arabica</i> , <i>Chenopodium album</i>
15	PT	25	House Dust, <i>D. pteronyssinus</i> , <i>D. ferinae</i>
16	HK	29	<i>Prosopis juliflora</i> , <i>Xanthium strumarium</i>

17	PL	31	<i>Cocos nucifera</i> , <i>Parthenium hysterophorus</i> , <i>D. pteronyssinus</i> , <i>D. ferinae</i>
18	AV	32	<i>Prosopis juliflora</i> , <i>Cynodon dactylon</i> , <i>Chenopodium album</i>
19	SK	34	<i>D. pteronyssinus</i> , <i>D. ferinae</i>
20	LM	23	<i>D. pteronyssinus</i> , <i>D. ferinae</i>
21	BC	20	<i>D. ferinae</i> , <i>D. pteronyssinus</i> , <i>Casuarina equisetifolia</i> , <i>Azadirachta indica</i> , <i>Amaranthus spinosus</i> .
22	ID	15	<i>Ageratum conyzoides</i> , <i>Parthenium hysterophorus</i> , <i>Amaranthus spinosus</i> , <i>Peltophorum pterocarpum</i>
23	RM	5	<i>D. pteronyssinus</i> , <i>D. ferinae</i>

Preparation of Extract for Subcutaneous (Rush) immunotherapy

Inj. Credisol,^R aqueous allergen extracts were standardised according to w/v ratio following protocol of Allergo Pharma, West Germany in graded concentration of strength: 1-4 supplied by Creative Drug Industries, Navi Mumbai, India. The antigens included in vaccines for SLIT and SCIT are given in tables 1 and 2.

Strength 1 (Orange Label) : 1+24999 (0.004%w/v),

Strength 2 (Green Label) : 1+2499 (0.04%w/v),

Strength 3 (Blue Label) : 1+249 (0.4%w/v),

Strength 4 (Red Label) : 1+49 (2%w/v),

Maintenance treatment set : (1 vial of 4.5mL), this vial contains extract of strength 4.

The dose schedule used for rush SCIT is described in table 3.

Table 3 - Schedule for administration of subcutaneous rush immunotherapy

Day	Vial (Strength)	Time	Volume (in mL)
Day 1	Vial 1 (1:25000)	9.00 am	0.2
		9.30 am	0.4
		10.00am	0.6
	After one hour interval		
	Vial 2 (1:2500)	11.00 am	0.2
		11.30 am	0.4
		12.00 noon	0.6
	+ Two hours of observation		
Day 2	Vial 3 (1:250)	9.00 am	0.2
		9.30 am	0.4
		10.00am	0.6
	After one hour interval		
	Vial 4 (1:50)	11.00 am	0.2
		11.30 am	0.4
		12.00 noon	0.6
	+ Two hours of observation		

Following initial rush immunotherapy, patients received maintenance immunotherapy once in two weeks consisting of 0.6mL of 1:50, w/v.

The validated and standardised translated Kannada version of Juniper and co-workers' (QoL) questionnaire²⁰ was administered once in a month and the patients were followed for a period of one year to assess the (QoL).

RESULTS

Out of 25 SLIT patients, 2 patients dropped out after obtaining the prescription for immunotherapy, because of financial constraints. Of 25 rush SCIT patients, 2 patients dropped out, one was not able to tolerate the maintenance dose and another patient was not able to maintain the cold chain. Among 23 patients in each SLIT and rush SCIT groups, 11 (48%) and 9 (39%) were suffering from allergic rhinitis, whereas 12 (52%) and 14 (61%) were having both allergic rhinitis and bronchial asthma, respectively.

Of 23 patients in SLIT group, all the patients showed sensitisation a positive SPT reaction to dust mites (*Dermatophagoides farinae*, *Dermatophagoides pteronyssinus*); followed by food items (Almond, Banana, Brinjal, Cashew nut, Orange, Mustard, Lemon, Curd and Cucumber), 10 patients, tree pollens (*Prosopis juliflora*, *Cocos nucifera*, *Cassia siamea*, *Typha angustata* and *Carica papaya*, *Eucalyptus spp*, *Ailanthus excelsa*, *Acacia arabica*, *Casuarina equisetifolia* and *Holoptelea intergrifolia*) in 9 patients and weed cultigen pollens (*Brassica nigra*, *Dodona viscosa*, *Ageratum conyzoides*, *Xanthium strumarium*, *Parthenium hysterophorus*, *Amaranthus spinosus* *Chenopodium album*) in eight patients 8. In case of rush SCIT group 18 out of 23 of patients elicited marked positive reaction to dust mites, 15 to one or more tree pollens (*Prosopis juliflora*, *Cocos nucifera*, *Cassia siamea*, *Typha angustata* and *Carica papaya*, *Eucalyptus spp*, *Ailanthus excelsa*, *Acacia arabica*, *Casuarina equisetifolia* and *Holoptelea intergrifolia*), 14 to weed cultigens pollens (*Brassica nigra*, *Dodona viscosa*, *Ageratum conyzoides*, *Xanthium strumarium*, *Parthenium hysterophorus*, *Amaranthus spinosus*, *Chenopodium album*), 9 to foods, 8 to house dust and 7 to grass pollens namely *Cynodon dactylon*, and *Sorghum vulgare*. Only 11 patients demonstrated positive skin reaction to insects (Cockroach, Mosquito and Housefly) in both the SLIT and SCIT groups.

Patients with positive SPTs to pollens, mites and dusts were administered IT with antigen mix (1-5) vaccines, where as patients allergic to foods (SPT+history+ve) were advised to avoid that food in their diet.

All the patients were observed for both local and systemic adverse reactions. Among patients receiving SLIT, two patients developed taste disturbance and one reported itching of ears which subsided on its own whereas in case of SCIT (rush), three patients developed erythema and induration at the site of injection which also subsided on its own. None of the patients receiving SLIT developed systemic reactions. In SCIT (rush) group, one patient developed generalised purities which subsided on treatment and another patient developed breathlessness which also subsided on treatment with bronchodilators but was dropped from the study because he was not able to reach the maintenance dose.

Medicines used during SLIT and rush SCIT were antihistamines (Cetirizine 10 mg, Levocetirizine 5-10 mg), decongestants (Pseudoephedrine), bronchodilators (Salbutamol 100-200 µg, Salmeterol 25 µg, Formeterol 12-14 µg, Theophylline 100-300 mg, Doxophylline 400 mg), inhaled steroids (Beclomethasone 50-200 µg, Fluticasone 100-250 µg).

Assessment of Quality of Life

To assess the QoL self administered QoL questionnaire was given once in a month to all the patients asking them to answer questions by circling the number that best describes how troubled they have been during that period as a result of symptoms SLIT and SCIT was given and patients were for 12 months. The symptoms and drug requirement were decreased in both SLIT and rush SCIT groups after a period of one year treatment compared to the baseline.

There was an improvement in QoL after a period of one year of treatment with a change in mean rank from 13 to 1.43 in SLIT group and 13 to 1.07 in rush SCIT group, and this was statistically significant (Table 3). From the baseline to 12 months of follow-up, there was no statistically significant difference in QoL between SLIT and rush SCIT group patients. (Table 4)

The improvement in overall QoL between SLIT and rush SCIT groups was not statistically significant ($P=0.522$) [Mann-Whitney U test, $Z = 0.641$], suggesting that both SLIT and rush SCIT are equally efficacious in

improving QoL among patients with respiratory allergy. The QoL was assessed within the group, between the groups from the baseline to 12 month of treatment and also between overall QoL scores between the groups.

DISCUSSION

Sublingual immunotherapy involves putting large doses of allergen extracts under the tongue, where the allergen is absorbed and subsequently induces immune response that may promote tolerance to the allergen. Subcutaneous (rush) immunotherapy is a technique for advancing an allergic patient^{11,19,21} (through a series of injections) to reach "maintenance dose" of allergen extract in a short period of time resulting in more rapid build up phase for faster clinical efficacy.²¹

In the present study, QoL has been assessed within the group, between the groups from the baseline to 12 months of treatment and also between overall QoL scores between the groups. It was observed that there was significant improvement in QoL from the baseline to 12 month of treatment in each group of patients. However, the difference in QoL score between the two groups was not statistically significant from the baseline to 12 months of treatment. Thus, the above findings suggest that both the SLIT and rush SCIT are equally efficacious in improving the QoL of patients with respiratory allergy.

Bousquet *et al*¹⁷ compared QoL in patients with asthma receiving SLIT and placebo and the changes from baseline to end point showed significant differences in favour of the SLIT group compared to the placebo group. In a study by Samantha *et al*²², there was a significant improvement in QoL among patient receiving SCIT compared to the patient in the placebo group. However, Khinchi *et al*²⁴ observed no statistical significant difference in QoL scores among three groups, i.e. SLIT, SCIT and placebo, using a 36-item short-form health survey (SF-36) questionnaire.

In conclusion, both SLIT and rush SCIT are equally efficacious in improving the QoL among patients suffering from respiratory allergy. There is a need for similar studies involving larger sample size and with a follow-up period of longer duration.

The limitation of the present study is that the control groups (pharmacotherapy and placebo injections immunotherapy) could not be taken to compare with SLIT and rush SCIT groups.

REFERENCES

1. Pawankar. Editorial Allergy Clin Immun Int. - JWAO. 2005 (Sept. Oct 2005)
2. Bousquet J, Fayol C. Assessment of quality of life in patients with perennial allergic rhinitis. *J Allergy Clin Immunol* 1994;74:182-8.
3. World Health Organization Prevention and Control of Chronic Respiratory Diseases at Country Level, Towards a Global Alliance against Chronic Respiratory Diseases (GARD) based on the WHO Meeting on Prevention and Control of Chronic Respiratory Diseases, Geneva, Switzerland. WHO/NMH/CHP/CPM/CRA/05. 17-19 June, 2004.
4. Lanny J. Treatment of allergic rhinitis. *Am J Med.* 2002; 113(9A): 17-24.
5. Ronald Efficacy and safety of sublingual immunotherapy with gross pollen allergen tablets for seasonal rhinoconjunctivitis. *J Allergy Clin Immunol* 2006 ; 118(2): 434-40.
6. Srivastava D, Singh BP, Sudha VT, Arora N, Gaur SN. Immunotherapy with mosquito (*Culex quinquefasciatus*) extract: a double-blind, placebo-controlled study. *Ann Allergy Asthma Immunol* 2007; 99: 273-80.
7. Srivastava D, Singh BP, Arora N, Gaur SN. Clinico-immunologic study on immunotherapy with mixed and single insect allergens. *J Clin Immunol* 2009; 29: 665-73.
8. Srivastava D, Singh BP, Arora N. Current immunological approaches for management of allergic rhinitis and bronchial asthma. *Inflamm.* 2009; 58(9): 523-36.
9. Technical Evaluation Centre, Assessment programme, June 2003: 18(4): 1-42.
10. Frankum B, Katelaris C, Eckerman S, et al. A comparative study of cost-effectiveness and tolerability of rush versus conventional bee venom immunotherapy. *Ann Allergy Asthma Immunol* 1995; 74: 61.
11. Portnoy J, King K, Kanarck H. Premedication reduces the incidence systemic reactions during rush immunotherapy. *Ann Allergy* 1994;73:409-18.
12. Bousquet J, Cauwenberg PV, Khaltev N. Allergic rhinitis and its impact on asthma. *J Allergy Clin Immunol* 2001; 108: S147-S334.
13. American Thoracic Society Lung function testing: selection of reference values and interpretative strategies. *Am Rev Respir Dis* 1991; 144: 1202-18.
14. Tripathi DM. Role of allergen testing and immunotherapy in the management of respiratory allergic diseases. *Bombay Hospital J.* 2002; 44(3): 419-25.
15. Tripathi DM. Status of skin testing and specific immunotherapy in management of asthma. *Bombay Hospital Journal* 1994; 36(1): 164-71.

16. Agarwal MK, Gupta S, Bansal SK, Vijayan VK. Cross reacting and unique allergenic components in insect extracts used for the diagnosis and immunotherapy of patients suffering with respiratory allergy. In: Sarma PU, Singh BP, Rao DN *et al*, editors. Trends in Clinical Biochemistry and Laboratory Medicine, Association of Clinical Biochemists; 2003: pp 314-24.
17. Bousquet J, Scheinmann P, Guinneeain MT, et al. Sublingual swallow immunotherapy (SLIT) in patients with asthma due to house dust mites; a double blind placebo controlled study. *Allergy* 1999; 54: 249-60.
18. Shyam AC, Parasuramalu BG, Chandregowda BV. Clinical efficacy of sublingual immunotherapy in respiratory allergy patients; a preliminary study. *Indian J Allergy Asthma Immunol* 2008;22(1):1-5.
19. Sharkey P, Portnoy J. Rush immunotherapy: experience with a one-day schedule. *Ann Allergy Asthma Immunol* 1996; 76: 175-80.
20. Juniper EF, Rohrbaugh T, Meltzer EO. A questionnaire to measure quality of life in adults with nocturnal allergic rhinoconjunctivitis. *J Allergy Clin Immunol* 2003; 111(3): 484-90.
21. Bousquet J, Lockey R, Malling HJ, et al. Allergen immunotherapy: therapeutic vaccines for allergic diseases: A WHO position paper. *J Allergy Clin Immunol* 1998; 102: 558-62.
23. Walker S M, et al. Gross pollen immunotherapy for seasonal rhinitis and asthma: a randomized, controlled trial. *J Allergy Clin Immunol* 2001; 107: 87-93.
24. Khinchi MS, *et al*. Clinical efficacy of sublingual and subcutaneous birch pollen allergen-specific immunotherapy: a randomized, placebo-controlled, double-blind, double-dummy study. *Allergy* 2004; 59: 45-53.