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Sublingual immunotherapy with a standardised grass pollen extract; a double-blind placebo-controlled study

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SUMMARY

Background: to asses the efficacy and safety of sublingual immunotherapy in patients with allergic rhinitis/conjunctivitis, a double blind placebo controlled study was performed.

Methods and results: fifty-seven (57) patients with a well-documented history of seasonal grass pollen allergy were evaluated in a DBPC trial over a period of 10 months (January to November 1995) with a view to investigating the efficacy and safety of sublingual immunotherapy with a grass pollen extract, 9,500 BU/ml. The course of treatment consisted of an incremental phase of approximately 3 weeks followed by a twice weekly maintenance dosage of 9,500 BU. Compared with the placebo group (30 patients), the group treated with grass pollen extract (27 patients) showed a significant ($p < 0.03$) lower mean severity of allergic complaints, i. e. sneezing, and itchy nose, a watery runny nose and itching of the eyes during the maximum pollen counts of the season. The use of anti-allergic medication was similar in the two groups.

Both groups showed a significant increase in grass-pollen-specific IgG serum levels. However, the increase shown in the patients treated with grass pollen extract occurred earlier in the season and was significantly ($p < 0.002$) higher than the increase detected in the placebo group. Side effects were limited to a small number of generally mild local reactions.

Conclusions: the treatment with sublingually applied grass pollen extract in patients suffering from allergic rhinoconjunctivitis caused by grass pollen was well tolerated and served to reduce the severity of allergic complaints, without reducing the consumption of anti-allergic medication.

Key words: Sublingual. Immunotherapy. Grass pollen. Allergic rhinoconjunctivitis. Specific IgG.

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INTRODUCTION

The efficacy of conventional immunotherapy via the subcutaneous route has been demonstrated in the treatment of both seasonal and perennial allergic disorders (1, 2). Until recently, sublingual immunotherapy was considered controversial, and therefore not recommended as a routine treatment (1, 3, 4). Although the safety of sublingual immunotherapy is well accepted, this standpoint is largely due to the limited number of studies demonstrating its clinical efficacy and the lack of knowledge of the mechanism by means of which allergens applied under the tongue interact with the immune system. However, the local immune system of the oral cavity resembles that of the gut and the lungs (5-7) and preclinical studies have shown that the oral mucosa is a potentially useful site for therapeutic modulation of allergic reactivity (8, 9).

An increasing number of double-blind, placebo-controlled studies with allergen extracts from weed pollen, grass pollen and house dust mite have shown sublingual immunotherapy to be effective and well tolerated (10-17). Certain studies have been criticised (3, 4) for their limited duration (12), the small number of patients included (10), or the criteria adopted to conclude clinical efficacy (13, 16). Negative results have been reported in the case of sublingual immunotherapy with cat dander (18). The present study was a randomised double-blind, placebo-controlled multi-centre study, with a standardised grass pollen extract devised to investigate the efficacy and safety of sublingual immunotherapy in patients with allergic rhinitis and/or conjunctivitis during their first year of treatment. The primary parameter for efficacy was the subjective assessment of symptoms by the patient and the patient''s physician.

Objective grass-pollen-specific IgG and IgE serum levels and the use of anti-allergic medication were considered to be secondary parameters for efficacy.

MATERIAL AND METHODS

Patients

Fifty-seven patients, aged 18-45 years, with a well-documented history of grass pollen allergy, who tested positive in response to a skin prick test for grass pollen allergens were evaluated. A skin test was considered positive if the diameter of the resulting wheal was more than half the diameter of the positive control solution. Patients who tested positive in response to a skin test for the epithelial extract of domestic animals were excluded from the study if their pet was kept indoors. The patients were stratified in terms of a positive or negative response to a skin test for house dust mite and randomised to receive one of two treatments. Patients showing symptoms of house dust mite allergy were excluded from the study, as were patients continuously using anti-asthmatic medication, as shown in their medical history.

Study drug

The investigational drug was a standardised (9,500 BU/ml) glycerinated (50% w/v) grass pollen extract (Oralgen®; ARTU Biologicals Europe B.V., Lelystad, The Netherlands) containing allergens of five grasses: Anthoxanthum odoratum (Sweet vernal grass), Cynodon dactylon (Bermuda grass), Dactylis glomerata (Orchard grass), Holcus lanatus (Velvet grass) and Phleum pratense (Timothy grass). The grass pollen extract was biologically standardised in accordance with modified Nordic Guidelines (19), using Histamine Phosphate as a reference. The placebo

consisted of the non-active ingredients. Both, the treatment with grass pollen extract and the placebo treatment started with an incremental phase of approximately 3 weeks. The coseasonal maintenance phase consisted of 1 ml (9,500 BU) administered twice weekly. If necessary, the course of the treatment was adjusted to the individual. The study medication was taken in the morning before breakfast; the solution was kept under the tongue for 1-2 minutes after which it was swallowed.

Study design

The study was conducted in accordance with the current guidelines of Good Clinical Practice. Patients were stratified and randomised after approval of the Review Board of the participating hospitals and written informed consent was obtained. Patients were selected on the basis of a clinical evaluation of their allergic history and skin test results. Treatment with the grass pollen extract and the placebo treatment started approximately 3 months prior to the start of the grass pollen season. Throughout the study period, both patients and investigators evaluated the therapy at six-week intervals. Each of the patients kept a diary during the months of April-October 1995, covering the grass pollen season from early beginning to the end. Adverse side effects, including local reactions, were recorded in detail. Compliance with study medication was assessed by recording the number of vials returned and measuring their content.

Symptom scores

The patients were asked to document all allergic complaints in their diaries on a daily basis by grading a list of 21 possible symptoms, using the following grading system: 0 = no complaints, 1 = mild complaints, 2 = moderate complaints and 3 = severe complaints. Prior to statistical evaluation, the symptoms specifically related to allergic rhinitis and conjunctivitis were categorised as sneezing, itchy nose, a watery runny nose and itching of the eyes. The symptom scores were used as the primary parameter for efficacy and evaluated as follows: total score per visit (assessment by the investigator at six-week intervals) and mean total daily score per week (patient diary). Patient scores were evaluated in a particular week if the patient completed the diary for at least four days during the week in question.

Anti-allergic medication

The use of anti-allergic (symptomatic) medication was strongly discouraged at the onset of the study. During the pollen season the following anti-allergic medication was allowed as a means of relieving allergic complaints:

nasal disodium cromoglycate 2% w/v (Fisons Pharmaceuticals) for minor symptoms of allergic rhinitis.

ophthalmic disodium cromoglycate 2% w/v (Fisons Pharmaceuticals) for minor symptoms of allergic conjunctivitis.

loratidine tablets 10 mg (Schering Plough) for moderate symptoms of allergic rhinitis and/or conjunctivitis.

budesonide nasal spray 50 µg/dose (Astra Pharmaceutica) for moderate to severe symptoms of allergic rhinitis.

betamethason tablets 0.5 mg (Schering Plough) for severe symptoms of allergic rhinitis and/or conjunctivitis.

The anti-allergic medication was distributed using code numbers for each patient. The use of anti-allergic medication was recorded daily in the patient"s diary. Returned supplies of anti-allergic medication were assessed, recording the code number and measuring the content or counting the tablets.

Serum parameters

Blood samples were taken at the onset of the study, before the start of the pollen season (after approx. 3 months of treatment) and again at the end of the trial, and analysed for the following parameters:

* Lol p I specific total IgG and Lol p I specific IgG_4.

* Grass pollen IgG_4 RAST.

* Grass-pollen-specific IgE -RAST.

Analyses were carried out by the Central Laboratory of Blood Transfusion of the Red Cross, Amsterdam, The Netherlands according to the method described by Aalberse et al. (20).

Pollen count

Pollen counts were obtained from the aerobiology laboratory of Leiden University Hospital (Leiden, The Netherlands) and the Department of Immunology of the Elkerliek hospital (Helmond, The Netherlands) and collected using the Burkard spore trap. In both cases the spore trap was situated on the roof of the hospital.

Statistical analysis of the results

Data was analysed on the basis of the intention to treat principle, using the validated SPSS/PC+ statistical package. Symptom scores, anti-allergic medication and grass-pollen-specific IgG and IgE changes from baseline between both treatment groups were compared using the distribution free Mann Whitney test. A (two sided) level of $p < 0.05$ was used to indicate statistical significance.

RESULTS

Study population

The characteristics of the study population are summarised in table I. The number of patients who tested positive for house dust mite and the sum score of allergic complaints during the pollen seasons prior to randomisation were equal in both groups. There was no difference

between the placebo group and the group treated with the grass pollen extract with regard to the scores of the remaining non-specific allergic sympoms as obtained from the investigations at six-weekly intervals and from the patient diaries. The sum score of these seventeen characteristics, including lung function disorders, remained low throughout the season, indicating non-relevance for the severity of the complaints and the absence of asthma complaints in the study population.

Table I
Summary of the baseline characteristics (demographic data)

	Placebo	Grass pollen extract	P-value#
Age	27.5	27.6	p = 0.860
Gender (M/F)	13/17	14/13	p = 0.600
House dust mite	8 positive 22 negative	11 positive 16 negative	p = 0.278
Sum score I"93	7.4 (sd: 2.6)	7.5 (sd: 2.8)	p = 0.987
Sum score I"94	7.9 (sd: 2.4)	7.4 (sd: 3.1)	p = 0.589

Sum score I: retrospective sum score in the pollen season of 1993 res. 1994: mean sum of scores for the relevant-symptoms of allergic rhinitis: itching of the nose, sneezing, a watery runny nose and itching of the eyes.
#: Mann-Whitney test (Fisher"s exact test for man/woman and house dust mite positive/negative ratio).

Clinical efficacy-symptom scores

The severity of the allergic complaints in the group treated with grass pollen extract and the placebo group was evaluated by the investigators at six-week intervals (Fig. 1). The symptom scores were low in both groups both before and after the pollen season, but the severity of the symptoms increased between May and August and peaked in June and July, during maximum pollen exposure. In the latter period the mean allergic symptom score of the patients treated with grass pollen extract was significantly lower ($p < 0.03$) than the score of the placebo group. Table II summarises the global symptom scores per visit.

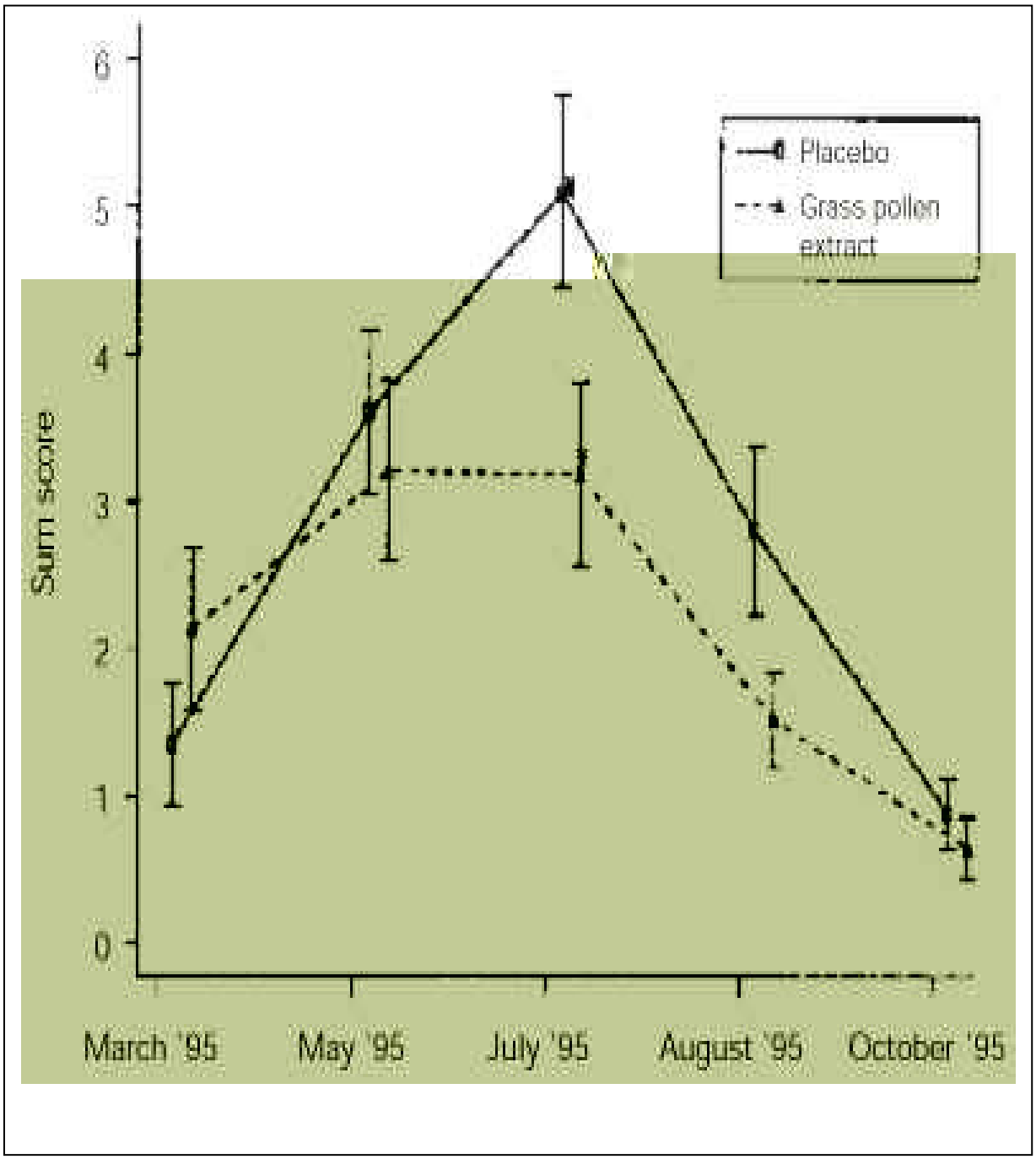


Figure 1.--Sumscore (± SD) of allergic symptoms per visit; investigators assessment of allergic complaints (sneezing, itching of the nose, a watery runny nose and itching of the eyes) at six week intervals. (*$p < 0.03$).

Table II
Mean total score per visit

		March"95		May"95		July"95		August"95		October"95	
Group		Placebo	GP*	Placebo	GP	Placebo	GP	Placebo	GP	Placebo	GP
	n	30	25	30	26	30	24	29	24	29	24
Complaints	mean	1.37	2.16	3.63	3.23	5.13	3.21	2.83	1.54	0.90	0.67
	median	0	2	3	3	4	2.5	2	1	0	0
	sd	2.27	2.75	3.03	3.15	3.60	3.05	3.11	1.61	1.29	1.05
	p-value	NS		NS		0.03		NS		NS	

*GP = Grass pollen extract.
NS: non significant.

Figure 2 shows the symptom scores obtained from the patient diaries. A clear relationship was observed between the severity of the symptoms scores and the amount of pollen in the air. During the peak of the pollen season, in the months of June and July, the symptom scores of the patients treated with grass pollen extract tended towards significance ($p < 0.08$).

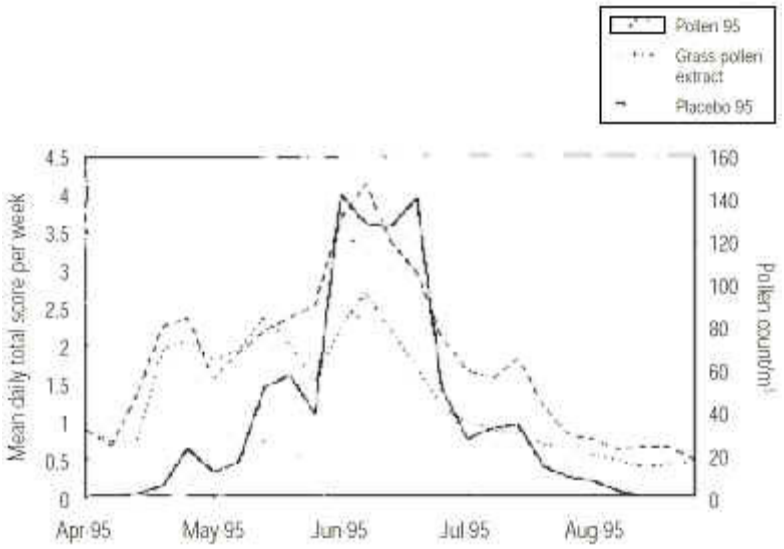


Figure 2.--Mean daily score per week and pollen count of 1995.

Clinical efficacy-anti-allergic medication

The use of anti-allergic medication, expressed as the percentage of days on which anti-allergic medication was used, was similar in both groups (Fig. 3). Furthermore the mean number of units used by the group treated with grass pollen extract (0.16 ± 0.37) was equal to the number of units used by the placebo group (0.31 ± 0.45).

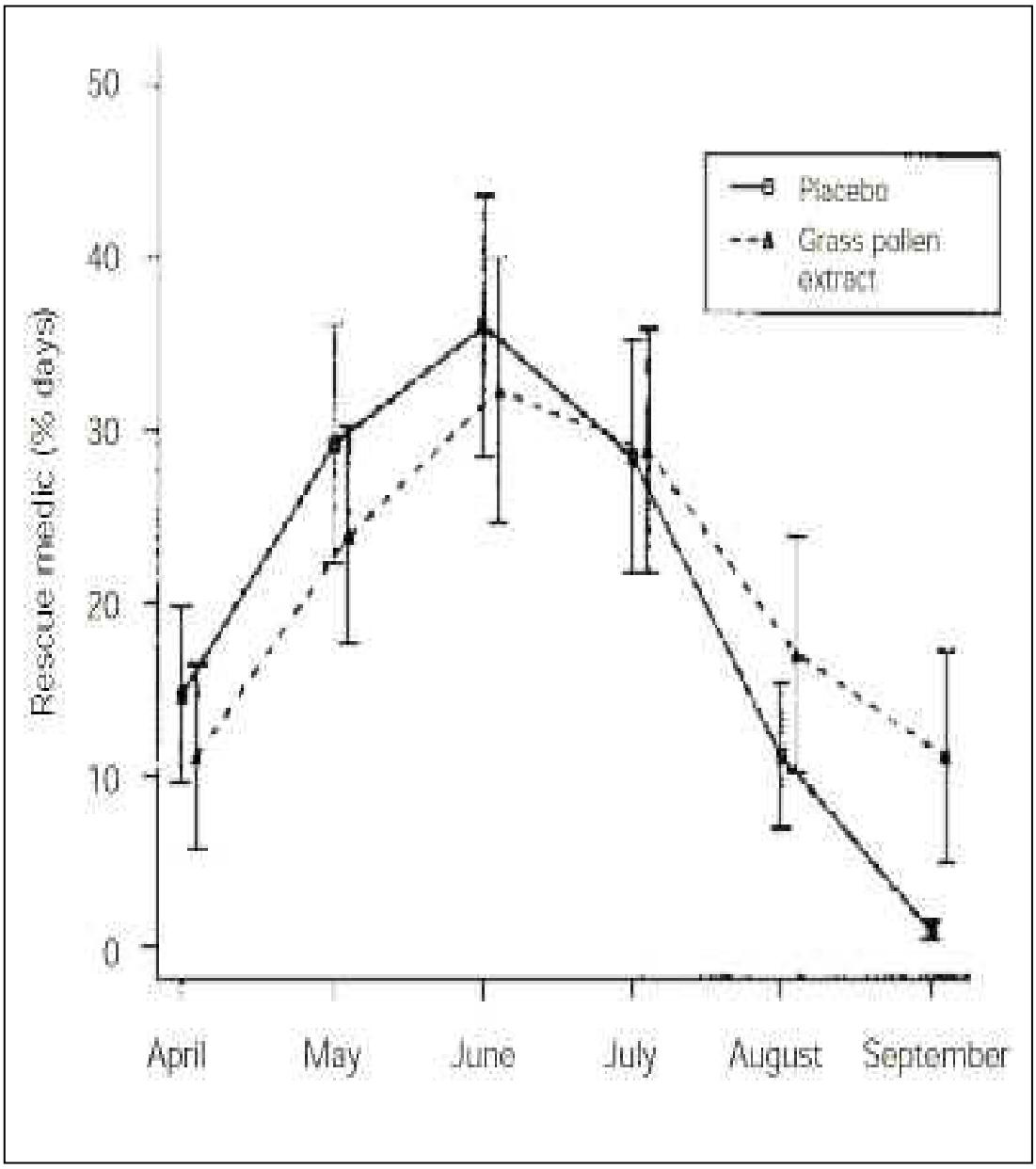
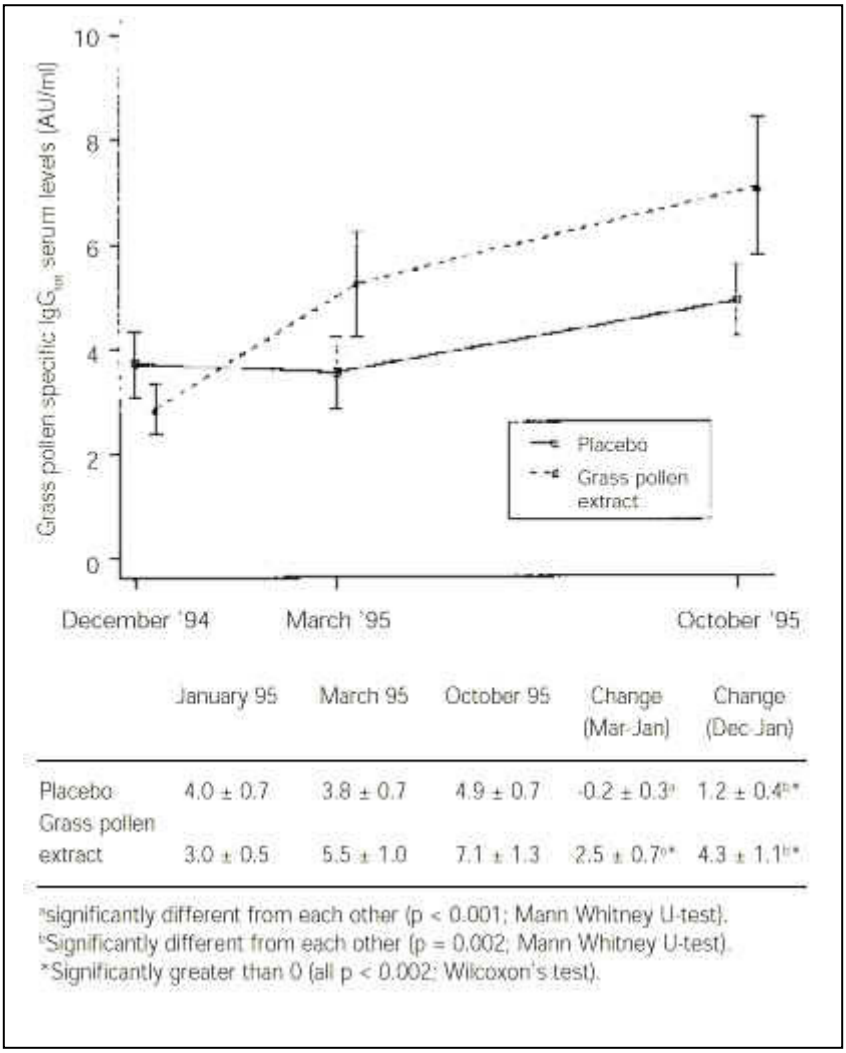


Figure 3.--The overall use of symptomatic medication, expressed as percentage of days (mean ± SEM) on which symptomatic medication was used.

Grass pollen specific serum parameters

Figure 4 shows a significant increase in grass-pollen-specific Lol p I, IgG serum levels in both the placebo group and the group treated with grass pollen. In the placebo group the grass-pollen-specific Lol p I, IgG serum levels increased only during the grass pollen season. The increase shown in the patients treated with grass-pollen-extract occurred prior to the pollen season and was significantly (p ¾ 0.002) higher than the increase seen in the placebo group.



	January 95	March 95	October 95	Change (Mar-Jan)	Change (Dec-Jan)
Placebo	4.0 ± 0.7	3.8 ± 0.7	4.9 ± 0.7	-0.2 ± 0.3[a]	1.2 ± 0.4[b]*
Grass pollen extract	3.0 ± 0.5	5.5 ± 1.0	7.1 ± 1.3	2.5 ± 0.7[a]*	4.3 ± 1.1[b]*

[a]significantly different from each other (p < 0.001; Mann Whitney U-test).
[b]Significantly different from each other (p = 0.002; Mann Whitney U-test).
*Significantly greater than 0 (all p < 0.002; Wilcoxon's test).

Figure 4.--Grass pollen specific IgG_{tot} serum levels (mean ± SEM) in arbitrary Units per **ml.**

Grass-pollen-specific IgG_4 and IgE serum levels did not differ from baseline values.

Drop outs

In general, patients dropped out for reasons not related to the clinical procedure of the study protocol. Two patients refused to continue the study prior to the onset of the grass pollen season due to an adverse event, i.e. an asthma attack (patient on placebo treatment; skin test for house dust mite positive) and paresthesia (patient on active treatment; skin test for house dust mite negative). In January, 69 patients were entered for the study. A total of 14 patients withdrew.

Nine patients dropped out before the pollen season (grass pollen extract group: n = 5; placebo group n = 4). There were 5 additional drop outs during the pollen season (grass pollen extract group: n = 3; placebo group: n = 2). Fifty-seven patients were evaluated and fifty-two patients completed the study per protocol.

Safety

The adverse events that occurred during the trial are presented in table III. Systemic reactions or serious drug-related adverse events were not observed. Local reactions consisted of itching of the palate and tongue. These reactions occurred in both groups and did not require special treatment or a reduction of the dose.

Table III
Summary of adverse events reported by 27 patients

	Grass pollen extract	Placebo	Total
Type of adverse event			
Skin	1	2	3
Psyche	2	0	2
Resistance	6	6	12
Cardiovascular	1	1	2
Mouth and tongue mucosa	3	1	4
Nervous system	0	1	1
Musculo skeletal	0	1	1
Gastro intestinal	1	1	2
Respiratory	1	3	4
Ear, nose and throat	3	5	8
Bleeding & clotting (sepc. nose bleedings)	0	2	2
Increasing hayfever complaints	2	2	4
Total	20	25	45
Classification of severity			
mild	45%	40%	
moderate	50%	36%	
severe	5%	20%	

DISCUSSION

The present study was completed following the pollen season of 1995. The number of patients evaluated was sufficiently large and the study population was well defined, with the baseline characteristics of both treatment groups being similar at the onset of the study and again on completion. Remarkably, the allergic symptom scores in both groups during the pollen season of 1993 and 1994, as obtained at the onset of the study (> 7, see table I) were higher than the scores obtained during the study period (patient score: 4-5 [Fig. 2); investigator score: 5 [Fig. 1]). This indicated that, in retrospect, patients experienced their allergies as more severe. The possibility of low exposure to grass pollens could be excluded because the summer of 1995 was characterised by the highest average grass pollen counts since 1988. The influence of sensitisation to mites was precluded, due to the clear distinction between the grass pollen season and the occurrence of house dust mite allergic complaints, and also as a result of the stratification, which resulted in an equal distribution of patients who tested positive in response to a skin test for house dust mite extract between both groups.

Clinical improvement was assessed by means of symptom scores evaluated by two independent methods; i.e., at six-week intervals by the investigators and daily by the patients in their diaries. Both assessment methods showed a similar result: clinical improvement of hay fever complaints was demonstrated ($p < 0.03$) by the investigator and daily symptoms scores in the patients on active treatment tended towards significance ($p = 0.08$). Clinical improvement was not accompanied by a decreased consumption of anti-allergic medication. Therefore, the differences detected in the symptom scores could not be attributed to the type or amount of anti-allergic medication. A few mild local adverse reactions were reported. These were possibly related to the dehydrating effect of glycerol, one of the constituents of the study medication.

Published DBPC studies on the efficacy of pollen-sublingual immunotherapy include symptom scores as well as the consumption of anti-allergic medication as efficacy parameters for clinical improvement. Mixed results were observed in terms of clinical improvement. Both an exclusive reduction of the consumption of anti-allergic medication (15) and a combined decrease of symptom scores and consumption of anti-allergic medication (11, 12, 17) were reported. The majority of clinical trials also include serum levels as an objective parameter, although no relationship has been established between specific IgG/IgE levels and clinical improvement. However, specific IgG can show that relatively high doses have been administered (1). In the present study a significant increase in Lol p I specific serum IgG was observed in both the group of patients treated with grass pollen extract and the placebo group. The increase in the placebo group was ascribed to the result of exposure to high pollen levels. In the patients treated with grass pollen extract the increase of serum levels was observed before the start of the pollen season, the result of the application of relatively high pollen doses. Objective verification by means of a nasal provocation test was not included because in many study-centres the test was not routinely performed.

The results of the present study as well as those of others, demonstrated that sublingual immunotherapy with pollen induced clinical improvement in patients suffering from rhinoconjunctivitis. The side effects of sublingual immunotherapy were negligible. Besides a positive risk/benefit ratio, advantages of sublingual immunotherapy were ease of administration and the short incremental phase during which intensive medical supervision was not necessary.

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RESUMEN

Antecedentes: para estimar la eficacia y seguridad de la inmunoterapia sublingual en pacientes con rinitis/conjuntivitis alérgica, se realizó un estudio, con un placebo, en el que ni el analizador ni el sujeto conocían las características.

Métodos y resultados: en dicho estudio se evaluaron cincuenta y siete (57) pacientes con un bien documentado historial de alergia estacional al polen de gramíneas, en un ensayo de DBPC, durante un período de diez meses (de enero a noviembre de 1995), con el propósito de investigar la eficacia y seguridad de la inmunoterapia sublingual con un extracto de polen de hierba, 9.500 BU/ml. El curso del tratamiento consistió en una fase de tres semanas, aproximadamente, de dosificación en aumento, seguida de una dosificación de mantenimiento de 9.500 BU, de dos veces por semana. Comparado con el grupo que utilizó el placebo (30 pacientes), el grupo tratado con el extracto de polen de gramíneas (27 pacientes) mostró una intensidad media de molestias alérgicas muy inferior ($p < 0,03$), a saber, estornudos, picor de nariz, mucosidades acuosas y picor en los ojos durante la máxima concentración de polen registrada en la estación. El uso de medicación antialérgica fue similar en los dos grupos. Ambos grupos mostraron un aumento importante en niveles de suero de polen de gramíneas específico IgG. Sin embargo, el aumento mostrado en los pacientes tratados con extracto de polen de gramíneas, se produjo en un momento anterior en la estación y fue significativamente más alto ($p < 0,002$) que el aumento detectado en el grupo tratado con el placebo. Los efectos secundarios se limitaron a algunas reacciones generalmente locales y leves.

Conclusiones: el tratamiento con el extracto de polen de gramíneas aplicado sublingualmente en pacientes que sufren de rinoconjuntivitis alérgica causada por el polen de gramíneas fue bien tolerado y sirvió para reducir la intensidad de las molestias alérgicas, sin reducir el consumo de la medicación antialérgica.

Palabras clave: Sublingual. Inmunoterapia. Polen de gramíneas. Rinitis/conjuntivitis alérgica. IgG específico.

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