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Sublingual immunotherapy: certainties, unmet needs and future directions

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Introduction

The subcutaneous modality of immunotherapy injections (SCIT) remained for several decades the only available administration route. SCIT is effective and safe, when properly prescribed and administered, but a remote risk of severe side effects is present (1), and the occurrence of technical errors is still not negligible (2). The problem of the risk/benefit ratio prompted the search for safer administration routes (nasal, bronchial, oral)(3), including the sublingual one (SLIT) that was described in 1986 (4). In less than 20 years, due to the large amount of clinical data, SLIT achieved credibility, and was introduced in the official documents as a viable alternative to the classic injection route (5, 6) for both adults and children (Fig. 1). To date SLIT is commercialized and routinely used in many European countries.

Despite the increasing optimism, it must be acknowledged that some aspects still need to be clarified, and that there is room for improvement. The unmet needs represent the basis for future research, whereas the clinical hypotheses would open the search for new indications and modalities.

SLIT: where do we stand?

Efficacy

To date, there are 60 randomized double blind placebo controlled trials performed with SLIT. Due to the number of the trials available, meta-analyses could be carried out (Tab. 1), with various inclusion criteria such as rhinitis only (7), asthma only (8), asthma and rhinitis in children (9, 10) (no pediatric meta analysis is available for SCIT). All the meta-analyses concluded for a significant effect of SLIT versus placebo (11). The reliability of the meta-analyses has recently been questioned by Nieto et al (12) expecially on the basis of possible publication biases and incorrect reporting of the data. Nonetheless, due to the

Figure 1 - Cronology of sublingual immunotherapy

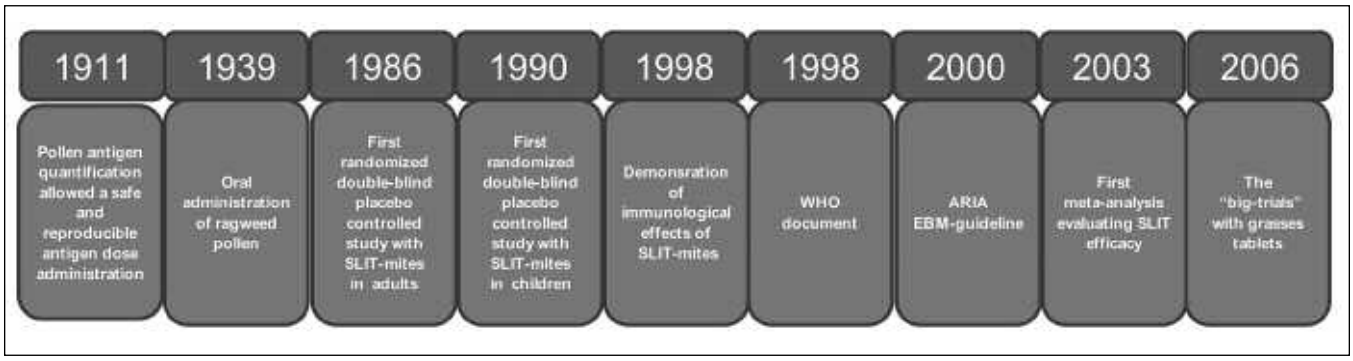

Table 1 - Meta-analyses on SLIT

Author	Patients	Disease	Trials	Effect size on symptoms	Comment
Calamita, 2006	303 adults + children	Asthma	5 pollens 4 mite	-0.38 (p= 0.07)	No change in symptoms score Significant reduction medication score
Olaguibel 2005	256 children	Asthma/ Rhinitis	3 pollens 4 mite	-1.42 (p=0.01)	Decreased symptoms and medications for asthma, rhinitis and conj
Wilson 2005	959 adults + children	Rhinitis	16 pollens 6 mite	-0.42 (p=0.002)	Decreased symptoms and medications for rhinitis. Asthma not evaluable
Penagos 2006	484 children	Rhinitis	5 pollens 4 mite	-0.56 (p=0.02)	Decreased symptoms and medications for rhinitis. No sub analysis feasible
Penagos 2008	441 children	Asthma	3 pollen 3 mite	-1.42 (p=0.02)	Decreased symptoms and medications for asthma

poor performance of the funnel-plot analysis, these negative aspects must be interpreted with great caution and without inappropriate generalizations. Moreover, the mentioned meta-analyses pooled together the studies with all allergenic extracts, whereas differences may exist among allergens. In this regard, there is so far one single meta-analysis restricted to house dust mite SLIT, showing a significant effect on symptom and medication scores in allergy due to mite (13). The problem of heterogeneity has been repeatedly highlighted as a drawback, but it is also true that meta-analyses are intended to summarize the results of studies when they are not directly comparable each other. Of particular interest are the recent so called "big trials"(14-19), all conducted with grass pollen extracts (Tab. 2). Those trials enrolled more than 200 patients each, reaching in some cases up to 600 patients. Of note, only one study with a similar number of patients exists for SCIT (20). The big trials invariably showed an effect of SLIT versus placebo ranging from 25% to more than 50%. The cut-off of 20% is unanimously considered the threshold for a clinically relevant effect (21), since antihistamines and nasal steroids hardly reach a 15% improvement versus placebo (22). In addition, those big trials with a dose-ranging design, clearly showed that the clinical effect is dose-dependent, and this is a robust proof of the efficacy according to the GRADE rules (23). On the other hand, the effects on QoL were always statistically significant in the big trials, but a "clinically relevant" difference (0.5 points in the RQLQ was not always achieved (14, 16).
The effect in asthma is still a matter of debate, since some studies (24-25) reported marginal or no effect on asthma symptoms. Nevertheless, in those studies, all the patients (active and controls) had no symptom of asthma at baseline and during the trial, therefore no effect could be seen. When patients have measurable asthma symptoms, the effect of SLIT is apparent, as recently shown in a pediatric trial (26). In addition, it has been shown that SLIT is capable of reducing the grade of bronchial hyperresponsiveness in adults and children (27, 28). Finally, the comparison of the efficacy of SLIT versus medications is still an open problem, because the effects of immunotherapy can be appreciated only in the long term (months). One head-to-head open randomized trial of SLIT versus inhaled budesonide in asthmatic patients, showed in the long term an overall superiority of immunotherapy (29). Another trial (30) in asthmatic children demonstrated that the clinical efficacy of SLIT plus fluticasone is equal to that of fluticasone alone, but the addition of SLIT improves also on non-bronchial symptoms.

Safety

The safety of SLIT is unanimously recognized to be superior to that of SCIT (31). An apparent datum is that no fatality has been ever reported with SLIT in 23 years of trials and clinical use. In addition, the reports of anaphylaxis with SLIT so far available in the literature are only four (32-35), being one of them questionable (32). On the othe hand, the report of an anaphylactic reaction at the first grass tablet SLIT (35), would suggest the opportunity to give the first dose under medical supervi-

Table 2 - The "big trials" with grass extracts

Author, year	Age range	Patients A/P *	Allergen	Durat.	Dose Preparation	Main positive results over placebo
Durham 2006	18-66	569/286	Grass 3 doses	6 m	15 μg (136 pts) 150 μg (139 pts) 450 μg (294 pts) Phl p 5/month Tablets	Drug score -28% (0.012) Symptoms -21% (0.002) only with the highest dose QoL improved No clinical change with the 2 low doses
Dahl, 2006	23-35	316/318	Grass	6 m	450 μg Phl p 5/month. Cumulat. 2.7 mg Tablets	RC symptoms -30% (.001); RC drugs -38% (.001); Well days -52% (.004)
Didier 2007	25-47	472/156	Grass 3 doses	6 m	240 μg (157 pt) 750 μg (155 pt) 1.2 mg (160 pt) /month Tablets	For 300 and 500IR Total and individual symptom and drug scores (<.001); RQLQ improved
Wahn, 2009	4-17	139/139	Grass	8 m	600 μg major allergen/month. Tablets	Rhinitis score -28% (.01) Medications -24% (.006) Medication free days (.01)
Ott, 2009	20-50	142/67	Grass	5 y 4 seas	Cumulative 1.5 mg major allerg/season	Combined score and symptom score significantly reduced since 1st season. Symptoms decrease from -33% to 47% (3rd seas) No change med.scores
Bufe, 2009	5-16	126/127	Grass	6 m	450 μg Phl p 5/month	Significant reduction in RC sympt score (-24%), asthma score (-64%), RC medications (-34%), well days (+28%). All p<.03

sion. A great attention has been paid to the safety in children (36). In fact, the age of 5 years is considered as a relative contraindication for SCIT, mainly because in young children any reaction may be more severe and more difficult to treat than in adults. Some of the post marketing surveys involved also children aged between 3 and 5 years (37-39), and confirmed that the safety is not impaired in the youger ages. A controlled dose finding study of safety (40) involved 48 grass-allergic patients outside pollen season. They received SLIT for 28-day periods at progressively increasing doses, up to 200 mcg Phl p 5 allergen that is about 40 times the amount given with one injection. The overall incidence of side effects was 74%, all of mild or moderate intensity. The most frequently reported events were irritation of the throat and oral itching. According to the recent data, the number of side effects seems to be dose-dependent, as happens with SCIT.

Since the majority of allergic patients are polysensitized, it is often necessary to prescribe immunotherapy with multiple allergens and it is crucial to know if the administration of different allergens with SLIT increases the risk of side-effects .Two post-marketing surveys performed in adults and children consistently suggested that the use of multiple allergens for SLIT does not increase the rate of side-effects (41, 42).

Mechanisms

Although the traditional effects on IgE and IgG4 are less pronounced with SLIT than with SCIT, several observation have recently began to clarify the mechanism of ac-

tion (Tab. 3). Some studies reported an increase of production of the regulatory cytokine IL-10 (43-46) and another study showed a reduction of the Th2 cytokine IL-13 (47). Savolainen et al demonstrated in vitro that SLIT reduces the expression of IL-5 and enhances the expression of IL-10 in PBMC stimulated with the allergen (48). Overall, the clinical effects of SLIT resemble those of SCIT, and the data available suggest that the mechanisms of action of the two routes are partially similar.
Additional mechanisms operating at the level of the sublingual mucosa and regional lymph-nodes may also be involved. During SLIT, allergens are captured within the oral mucosa by Langerhans-like dendritic cells expressing high-affinity IgE-receptors, producing IL-10 and TGF-β, and upregulating indoleamine dioxygenase (IDO), this suggesting that such cells are prone to induce tolerance (49). Finally, unique data on biodistribution in humans are available for SLIT, showing a long-lasting persistence of the allergen in the mouth, with an absent or negligible absorption through the mucosa (50, 51).

Table 3 - Modification of the parameters of immune system after SLIT

Local immune responses:
- no differences in CD3+, CD1a+, CD68+ cell counts (Lima et al., 2002)
- significant decrease of sublingual salivary ECP levels (Marcucci et al., 2001)

Systemic immune responses:
- significant decrease of serum ECP levels (Passalacqua et al., 1998; Sanchez Palacios et al., 2001; Vourdas et al.,1998)
- serum ICAM-1, IL-2 receptor, E-selectin, IL-12 levels unchanged (Reich et al., 2003)
- reduction of IL-13 and Th2 related hormone prolactin (Ippoliti et al., 2003)
- increase of IL-10 production (Ciprandi et al., 2005)
- changes in IgE levels ??
- significant increase in serum IgG4 levels (dose-response depending) (Tari et al., 1990; La Rosa et al., 1999; Bufe et al., 2004; Lima et al., 2002)
- increase in serum IgG1 levels (Tari et al., 1990; Bufe et al., 2004)
- no significant changes in CD40+, CD3+, CD4+, CD8+ cell counts (Ippoliti et al., 2003)
- significant increase in peripheral blood CD8+ T cells (Tari et al., 1990)
- induction of a specific T-reg response (Ciprandi et al., 2007)
- urinary leukotrienes: conflicting results (rhinitis: yes; asthma: no) (Yuksel et al., 1999)
- nasal eosinophils: significant decrease (La Grutta et al., 2007)

Additional effects

Recently, it was demonstrated that SLIT, similarly to SCIT can prevent the onset of new sensitizations. In a study involving more than 500 patients, the rate of occurrence of new sensitizations was 5.8% in the active group and 38% in the control group ($p< 0,001$) (52). An open controlled study by Novembre et al (53), perrformed in children, demonstrated that SLIT is capable of reducing the risk of asthma onset. These results were replicated in a larger randomized open study, involving more than 200 children followed up for three years (54). The occurrence of persistent asthma after 3 years was 38% of the controls and 2% of the SLIT. Certainly, the evidence of a preventative effect is still weak and relies on small numbers of patients: 151 for SCIT (55) and 340 for SLIT. In addition, this study confirmed the prevention of the onset of new sensitizations. On the other hand, there are so far only two studies, one non randomized (56) and the other randomized and double blind (57) that demonstrated a long-lasting effect of SLIT after discontinuation.

Adherence

In the case of SLIT, the adherence has been always considered a major concern, since the treatment is self-administered. The problem of the adherence was systematically addressed in three studies with method of the random telephonic interviews.
In the first study (58), involving 126 patients the adherence was 95% for pollen immunotherapy and 97% for mite immunotherapy. In the second study (59), conducted in more than 400 patients, the adherence rate at 3 and 6 months was greater than 90% in about 75% of the patients. The third study was conducted in children (60), and the results on compliance did not differ from those in adults. On the contrary, one retrospective study by Pajno et al. showed that the adherence was slightly greater with SCIT than with SLIT, but no quantitative assessment was provided in this study (61). Finally, a priori subgroup analysis was conducted in an open-label European study where adult patients received once-daily SLIT grass tablets with or without a device to aid compliance. Eighty-two patients reported using the device sometimes or always, and rated it easy to use (62).

Unmet needs and critical aspects

Despite the official positions and the relatively wide clinical use, some aspects of SLIT need urgently to be elucidated, in order to provide clinicians with clear and evidence-based recommendation for the clinical use of the treatment.

One of the critical aspects is that some studies provided totally negative results, although conducted with a rigorous methodology (63-65). No clear explanation has been provided for those results, but a generic "uncorrect patient's selection". This underlines the need for univocal criteria or parameters that help identifying the best candidates to SLIT. Another relevant problem is the large variability of the doses used in clinical trials. Indeed, both positive and negative results have been obtained with both low and high doses of allergens, and the dose interval for efficacy is reported to range between 2 and 375 times the amount given with SCIT. A clear dose response relationship has been formally demonstrated only for grass extracts, where the optimal dose has been identified in 15 to 25 mcg major allergen per day, that is roughly 50 times the monthly dose of SCIT. Thus, dose-response trials and the identification of the optimal maintenance dose are needed at least for the more relevant allergens. The variability of the study design, patients' selection, duration and regimen among the trials is anoter major problem that importantly affects the interpretation of the meta-analyses. Concerning this latter point, it should be remembered that meta-analyses put together the results obtained with various allergens and conclude for the efficacy of all allergens, that is not true. This underlines the need for a separate analysis of each single allergen, and this has been so far done only for dust mite (14).

From a clinical point of view, there is no consensus on which is the best administration regimen among the preseasonal, coseasonal, pre-coseasonal or continuous. It is true that for pollen allergens, the vast majority of the trials have utilized a pre-coseasonal regimen (66), but this cannot be immediately extrapolated to all extracts and to all patients. Similarly, the usefulness of a build-up phase is still a matter of debate. The no-updosing has been shown to be safe enough (67, 68), and some of the big trials have used a no-updosing regimen, but the applicability of this concept to all allergens and patients is not unanimously accepted. Also the best maintenance dosing (once daily, on alternate days, once weekly) has not yet been defined (66), as well as the optimal duration of a SLIT course. Another critical point is the the allergen and protein content of commercial extracts is highly variable among manufacturers (69). The use of multiple extracts have been shown to be safe (41, 42), but few and sparse data are available on its efficacy (70). A recent RDBPC trial was conducted to assess the efficacy of SLIT with a grass extract alone or in combination (71). This study showed that an immunological response is achieved with the thimothy extract over a period of 10 months, but the same dose combined with 9 other pollen extracts produced only a limited response.

Concerning the safety, it would be crucial to identify risk factors for systemic side effects, if any. In addition, the safety in subjects with previous reactions to injections has not been studied, as well as the risk for adverse events after a temporary suspension of SLIT.

Working hypotheses for the future

The efficacy and safety of SLIT can be, in principle, improved by the chemical modification of allergens, by using adjuvants or by enhancing the contact with the oral mucosa. This latter aspect has been recently addressed with the use of mucoadhesive substances, which have been demonstrated to improve the immunological effects in an animal model (72). More realistically, it can be expected that the good safety profile of SLIT would allow to expand its indications, expecially for conditions different than respiratory allergy.

There are, in fact, two studies in food allergy, one with hazelnuts (73) and one with the Pru p 3 allergen of peach (74), reporting positive results. In both studies a significant reduction of the oral provocation threshold was described, this suggesting the possible use of SLIT in food allergy. The use of SLIT has been also proposed in the past for the treatment of extrinsic atopic dermatitis (75). A randomized double blind placebo controlled trial, conducted in 30 children with mite allergy (76), reported a significant effect of mite-SLIT in reducing the SCORAD in mild to moderate atopic eczema. Those data substantially replicated which obtained in another open non-controlled, non-randomized pilot trial in 86 adult HDM-sensitized patients with mild-moderate atopic dermatitis (77). Surprisingly, a randomized controlled trial showed that SLIT with honeybee venom (maintenance 525 mcg) reduced the diameter of the large local reactions after sting challenge (78). This, although appealing, was only a proof of concept study, and SLIT cannot be presently recommended for the treatment of hymenoptera venom allergy (79). On the

other hand, several studies strongly suggest the applicability of SLIT for treating latex allergy (80-82). Other possible fields of investigation are the desensitization for nickel allergy, but in this case there are only basic studies in animal models (83), and baker's asthma (84).

Conclusion

SLIT represents a significant advance because of the efficacy, safety and convenience, and it appears particularly suitable in paediatric patients, where an optimal safety profile is required. Despite the general optimism, more studies are needed about the mechanisms of action, the pharmacoeconomics, the optimal doses for each allergen and on the ideal patients. The available data and the results of meta-analyses confirm the official positions on SLIT and justify the tangible change in the general opinion, which considers SLIT a more acceptable treatment. It is essential to remember that SLIT prescription must be made only by a specialist, after a detailed diagnosis has been established and the expected benefit/cost ratio has been carefully evaluated (85).

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