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Psychological stress affects response to sublingual immunotherapy in asthmatic children allergic to house dust mite

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While the clinical and immunologic efficacy of sublingual immunotherapy (SLIT) in allergic diseases has been extensively demonstrated, some patients display a poor clinical response. Psychological stress has been shown to play a role in atopy and also to affect response to immunomodulating therapies such as vaccination with microbial antigens. This study addresses the possibility of response to SLIT being affected by psychological stress. Forty children with mild asthma caused by allergy to *Dermatophagoides pteronyssinus* and *farinae* were subjected to SLIT and then divided after 6 months into two groups based on the results of the stress integrated measure (SIM) test: group 1 (24 stressed patients, mean SIM value of 60.1) and group 2 (16 non-stressed patients, mean SIM value of 7.6). There was also a higher prevalence of psychosocial stressing factors (divorced/absent parents, low income households, non-working parents) among stressed patients. The symptom score, peak expiratory flow (PEF), forced expiratory volume in 1 s (FEV_1) and serum eosinophie cationic protein (ECP) concentration were evaluated at both times. The serum concentration of neuroendocrine parameters [prolactin, cortisol, adrenocorticotropic hormone (ACTH)] was also measured after 6 months of therapy. While all the clinical parameters and ECP concentration improved after SLIT, symptom score, PEF and ECP showed a significantly greater improvement in non-stressed patients. The concentration of neuroendocrine parameters was significantly increased in stressed patients. Our findings show that psychological stress can affect response to SLIT also in allergic subjects and are consistent with data recently reported showing a correlation between stress and poor response to antimicrobial vaccines. Our data also suggest that stress evaluation may become a useful prognostic factor in immunotherapy.

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Atopic disorders are characterized by a T cell (Th)2-driven response that causes the involvement of many cell types, such as eosinophils and mast cells, as well as various soluble mediators, including cytokines, chemokines and neuropeptides. Disease progression can, however, be influenced by various host and environmental factors. The mechanisms linking psychological stress, personality and emotion to disease expression have been studied in recent years and it is now evident that the dysregulation of homeostatic neural, endocrine and immunologic pathways may play a role in the clinical expression of allergic diseases (1–3). It has been demonstrated that stressful events can contribute to the exacerbation of asthma in subjects with existing

illnesses (4, 5). Sandberg et al. have also shown that children subjected to stressful events are at significantly greater risk of acute exacerbation during the subsequent 6-wk period (6).

In addition, it has been recently observed that the impact of stress on the immune system can also affect vaccine response (7). Chronic stress may inhibit the stability of the IgG antibody responses to a bacterial vaccine or perhaps affect the number of IgG-producing B lymphocytes. The differences found in responses to Hep-B vaccine, influenza virus vaccine, and *Streptococcus pneumoniae* vaccine provide reliable evidence of how stress can alter both the cellular and humoral immune responses to novel pathogens and vaccines (8).

Stress-induced activation of the hypothalamic–pituitary–adrenal (HPA) axis induces increased cortisol levels. Cortisol has been shown to shift T cells toward a Th2 phenotype *in vitro* (9) and *in vivo* (10), and a relationship has been demonstrated between stress-induced cortisol and increased IgE production (11). Attention has also been focused on the role of nervous system in increasing inflammation (neurophlogosis or neurogenic inflammation). Receptors for hormones and neuropeptides are expressed on the surface of lymphocytes, and these cells expresses mRNA for adrenocarticotropic hormone (ACTH), prolactin, thyroid stimulating hormone (TSH) and growth hormone (GH) (12).

The clinical efficacy of sublingual immunotherapy (SLIT) for allergic diseases has been demonstrated. In addition, we previously showed that SLIT has an immunomodulatory effect on the Th1–Th2 balance (13). It is, however, not clear why some people respond poorly to specific immunotherapy. The aim of this study is to address the possibility of psychological stress disturbing the response to SLIT and thus affecting both the clinical outcome and the immunologic and neuroendocrine parameters.

Materials and methods

Patients

A group of 40 children and young adolescents allergic to *Dermatophagoides pteronyssinus* and *farinae* with a history of mild/moderate asthma, a positive skin prick test with a wheal diameter > 5 mm to house-dust mites (HDM) (Lofarma, Italy), and specific IgE to HDM at least of class 3 (RAST method, Pharmacia, Uppsala, Sweden) were selected. Only patients with a FEV_1 > 70% of predicted value was included. Methacoline inhalation tests were performed to determine airway responsiveness in accordance with the method described by Cockcroft and Hargreave (14). Patients with a positive skin test to other inhalant allergens, a clinical history of other allergies, such as seasonal asthma caused by pollens, a history of specific immunotherapy (SIT) in previous years, or severe asthma were excluded. The inclusion and exclusion criteria are detailed in Table 1.

Sublingual swallow therapy (SLIT) was administered to all patients subsequent to a 6-month period of observation in order to assess the symptom score before therapy according to the scale described by Tari et al. (15). The FEV_1 (% of predicted value), PEF and metacholine responsiveness of all patients were measured before therapy. During the study, patients were allowed to take other drugs for the relief of symptoms, if needed, for not more than seven consecutive days: inhaled steroids (200 μg/puff, 2–4 puffs) and inhaled salbutamol (250 μg/puff, 1–3 puffs) on demand. Patients were asked to record daily symptom scores in diary cards on the same scale used before beginning immunotherapy.

After 6 months of treatment all patients were subjected to clinical re-evaluation, all diary cards were evaluated, and FEV_1 and PEF were measured. At this time, all patients were also subjected to evaluation of psychological stress. The physicians performing the clinical evaluation were not aware of the results of the stress evaluation.

Clinical protocol

The immunotherapy consisted of drops of monomeric allergoid *D. pteronyssinus* and *D. farinae*

Table 1. Inclusion and exclusion criteria

Inclusion criteria	Exclusion criteria
Sensitivity to HDM	Sensitivity to allergens other than HDM
Perennial symptom	Seasonal symptom
Positive skin prick test for HDM wheal ≥ 5 mm	Negative skin prick test
RAST for HDM at least of class 3	RAST for HDM lower than class 3
FEV_1 > 70%	FEV_1 < 70%
No SIT in previous years	SIT in previous years
	Fever
	Presence of autoimmune, endocrine or metabolic diseases
	Presence of chronic diseases
	Presence of cancer
	Use of oral contraception in female subjects of childbearing potential
	Smoking
	Known use of voluptuary drugs
	Need for asthma treatment in the previous 2 wk

HDM, house-dust mites; SIT, specific immunotherapy

(LAIS, Laboratorio Farmaceutico Lofarma, Milan, Italy). The allergen was modified through reaction with potassium cyanate at neutral pH to substitute ε-amino groups of lysine residues and thus substantially lower its capacity to react with IgE, as measured by radio-allergosorbent-test inhibition (16). While about 84% of the ε-amino groups were substituted, the molecular dimensions of the carboxylated allergen assessed by means of sodium dodecylsulphate–polyacrylamide gel electrophoresis remained unchanged. The immunotherapy preparation was titrated in allergenic units (AU) and standardized by radio-allergosorbent test inhibition compared with an in-house reference titration. The content of major allergens could not be detected because of the loss of activity because of the chemical treatment (16).

The drops were placed in the patient's oral cavity, under the tongue, and retained for 1–2 min before swallowing. The build-up phase involved the administration of increasing doses of the allergen (25, 50, 75, 100, 150, 300, 450, 600 and 900 AU). Each dose was taken for three consecutive days. Patients received 900 AU twice a week in the mainstream phase, and the maintenance dose was administered until the end of the trial.

Evaluation of psychological stress

Evaluation of psychological stress was performed after 6 months of SLIT using the Italian version (17) of the Mesure du Stress Psychologique (MSP) test for perceived stress (18). This test, designed by Lemyre and Tessier, involves a 49-item questionnaire based on different perceived aspects regarding the patient's state (cognitive, physiological and behavioural aspects). The patient indicates intensity for each item on a scale of 1–4 and the values are then added to give his or her total score. The total scores thus obtained are set out in tables showing the key data in the form of centiles and points specifically registered for males and females. Convergence validity was established with classical depressive and anxiety scales. In addition, concomitant validity with immunity was shown in a double before-after design of school stress and holidays by means of salivary immunoglobulin concentration (18). The test was designed for use with normal populations, not psychiatric patients, and proves useful in evaluating perceived stress in clinical trials as well as the persistence of stress over time.

The stress integrated measure (SIM) values are obtained by means of mathematical algorithms that combine the MSP scores with other parameters such as patient's posture, movements and voice, physiological variables (respiration, skin conductance, neuromuscular excitability) and Spielberger's state-trait anxiety inventory (STAI) scale (19). This is divided into two 20-item sections, with each section used respectively to evaluate anxiety as a transitory state or as a comparatively habitual trait of personality. The SIM values obtained express a functional configuration for each patient on a scale of 0–100, with 100 indicating maximum stress.

The frequency of psychosocial stressing factors (divorced/absent parents, low-income households and non-working parents) among the two groups was also evaluated. Stress evaluation at baseline was not performed so as to avoid the possible influence of the results on the perception of symptoms by the patients or their parents in the diary cards, thus affecting the symptom scores. The evaluation was performed in a blind fashion, as well as the clinical evaluation. The physicians were not aware of the clinical status of the patients.

Blood samples

At baseline and after 6 months of SLIT, blood samples were taken from each fasting patient at the same time (8.30 AM) so as to avoid physiological circadian variations. The serum concentration of ECP was evaluated at both times, while concentration of PRL, cortisol and ACTH was determined after 6 months of immunotherapy.

Serum concentrations were measured by means of chemiluminescence for PRL and cortisol, immunoradiometric assay (IRMA) for ACTH, and enzyme-linked immunosorbent assay (ELISA) for ECP. Each serum was aliquoted in three samples. The ECP assay was carried out on fresh sera while the other sera were stored at −80°C. The standardized assays used were purchased from different sources: Pharmacia (Uppsala, Sweden) for ECP (measuring range 2–200 ng/ml), Roche for cortisol (sensitivity 0.50 μg/dl), Boehringer Mannheim (Germany) for PRL (sensitivity 0.47 ng/ml), and IDS Ltd. (UK) for ACTH (sensitivity 0.55 pg/ml). All the assays were carried out in accordance with the manufacturer's instructions.

Statistical analysis

Due to the small sample size, all the data were evaluated for statistical significance by means of nonparametric tests. The Wilcoxon test was used

to compare symptom score, PEF and FEV_1 within each group and the Mann–Whitney test to compare all the measured parameters (symptom score, PEF, FEV_1 and concentrations of PRL, cortisol, ACTH and ECP) between the two groups. Bonferroni's correction was used for multiple analyses.

The correlation between SIM values and the percentage reduction of symptom score were also examined, both for all patients and for each group, as was the correlation between SIM values and each evaluated laboratory parameter.

Finally, the frequency of patients with a symptom score 0 between the two groups was compared by means of the chi-squared test. The chi-squared test was also used in the evaluation of the frequency of psychosocial stressing factors. Statistical significance was attributed to p-values < 0.05.

All statistical analysis were performed using the StatView software, version 5.0 (SAS Institute Inc.). All laboratory values, the symptom score and FEV_1 and PEF (percentage of predicted) are expressed as mean ± SD.

Results

After evaluation of psychological stress, the patients were divided in two groups on the basis of SIM values: 24 subjects with values indicating the presence of psychological stress and 16 with normal values indicating absence of stress. All the stressed patients showed the presence of anxiety as a habitual trait of personality, determined by means of the STAI scale (data not shown). The demographic data of the two groups are shown in Table 2. The only significant difference between the groups was in the prevalence of patients with associated rhinitis, which was higher among non-stressed group.

The stressed group displayed a significantly higher percentage of family stressing psychosocial factors, such as divorced/absent parents, low-income households or non-working parents

Table 2. Demographic data, RAST values and stress integrated measure (SIM) values (mean ± SD) of the studied patients

	Group 1 n = 24	Group 2 n = 16	p-value
Median age (range) in years	13 (9–19)	13 (8–16)	0.063
Sex (M/F)	12/12	8/8	1.000
Patients with associated rhinitis (%)	2 (8.3)	10 (62.5)	0.003
RAST (mean ± SD) kU/l	15.4 ± 9.1	17.6 ± 9	0.663
SIM (±SD)	60.1 ± 18.3	7.6 ± 4.2	<0.0001

Table 3. Symptom score, pulmonary function test results and ECP concentration before sublingual immunotherapy and after 6 months of immunotherapy. Both the complete analysis and the restricted analysis are shown for each parameter (see the text for details)

	Group 1	Group 2	p-value*
(a)			
Symptom score T_0	2.83 ± 0.37	2.87 ± 0.33	0.8252
Symptom score T_1	2.17 ± 0.19	0.12 ± 0.33	<0.0001
p†	0.001	<0.0001	
Symptom score T_0‡	2.91 ± 0.29	3 ± 0	0.7369
Symptom score T_1‡	2.36 ± 0.79	0	0.0002
p†	0.0117	0.0277	
Score reduction (%)	25 ± 34.4	95.8 ± 11.4	<0.0001
Score reduction (%)‡	18.2 ± 26.7	100	0.0002
(b)			
PEF§ T_0	73.8 ± 19	81.1 ± 10	0.2141
PEF§ T_1	80.2 ± 16.3	88.4 ± 10.7	0.0292
p†	0.0009	<0.0001	
PEF§ T_0‡	76.1 ± 18.7	86.7 ± 13.4	0.117
PEF§ T_1‡	80.5 ± 17.4	95 ± 13.1	0.0187
p†	0.0015	0.0277	
FEV_1§ T_0	85.5 ± 18.1	90.9 ± 3.8	0.6587
FEV_1§ T_1	91.9 ± 14.7	96.1 ± 4.1	0.8252
p†	<0.0001	<0.0001	
FEV_1§ T_0‡	85.4 ± 19.4	94.6 ± 3.4	0.3135
FEV_1§ T_1‡	91.6 ± 15.7	99.7 ± 3.4	0.093
p†	<0.0001	0.0277	
(c)			
ECP T_0 ng/ml	68.3 ± 72.4	51.6 ± 27.6	0.1591
ECP T_1 ng/ml	66.4 ± 66.8	9.9 ± 3.1	<0.0001
p†	0.449	0.0005	
ECP T_0‡ ng/ml	73.1 ± 75.5	50.9 ± 26.1	0.218
ECP T_1‡ ng/ml	70.3 ± 70.6	8.9 ± 3.3	<0.0001
p†	0.2425	0.0277	

*p groups 1 vs. 2.
†p T_0 vs. T_1.
‡Asthmatic patients only.
§Percentage of predicted.

(17 subjects, 70.8%, in group 1 vs. 3 subjects, 18.7%, in group 2, $p = 0.022$).

The clinically relevant data for both groups before and after 6 months of SLIT (symptom score, PEF and FEV_1), together with ECP concentration at both times, are summarized in Table 3. There were no significant differences between the two groups at baseline.

While the improvement of all clinical parameters was statistically significant in both groups after 6 months of SLIT, comparison of the groups shows a significant difference in the symptom score (0.12 ± 0.33 in non-stressed group vs. 2.17 ± 0.99, $p < 0.0001$). The percentage of reduction in clinical score was also significantly higher among non-stressed patients (Table 3a). As regards the pulmonary function tests, PEF values were significantly higher in non-stressed patients (88.4 ± 10.7% vs 80.2 ± 16.3%, $p = 0.029$), whereas no statistically significant difference is registered in FEV

values (Table 3b). The serum concentration of ECP, which displayed an increase in both groups at baseline, was also significantly lower in non-stressed patients after 6 months of immunotherapy (Table 3c).

In order to rule out the possibility of improvement in asthma being due to the effect of SLIT on rhinitis, the statistical analysis was restricted to patients affected exclusively by asthma, excluding all the subjects who also had rhinitis. In this restricted analysis, the results for the non-stressed group once again showed a statistically significant improvement as regards clinical score (with significant higher reduction of the score), serum ECP concentration and PEF values. Though not statistically significant, the difference in FEV_1 values showed a p-value lower than that obtained from the analysis involving all subjects (Table 3).

Correlation analysis was applied to the SIM values and the reduction of symptom score (expressed as percentage of reduction after 6 months of SLIT). Analysis of all the patients revealed a significant inverse correlation between SIM values and the reduction of symptom score, with $r = -0.906$, $p < 0.0001$ (Fig. 1a). The results of correlation analysis within the single groups showed that while score reduction was not correlated to SIM values in non-stressed patients ($r = 0.193$, $p = 0.48$) (Fig. 1c), there was a statistically significant inverse correlation between SIM and score reduction in the stressed group ($r = -0.773$, $p < 0.0001$) (Fig. 1b).

In addition, the number of patients with symptom score 0 after 6 months of therapy was 14 in the non-stressed group (87.5%) and only two (8.33%) in the stressed group ($p < 0.0001$).

After 6 months of SLIT, the serum concentration of PRL, ACTH and cortisol was evaluated in addition to diary cards and PFT assessment. The concentration of all these parameters was higher in the stressed group. Mean PRL concentration (Fig. 2a) was 15.9 ± 8.6 μg/l in group 1 and 9.8 ± 6.2 μg/l in group 2 ($p = 0.0037$) and ACTH concentration (Fig. 2b) was 22.2 ± 9.5 and 12.9 ± 4.9 μg/l respectively ($p = 0.003$). Finally, cortisol values were similarly higher among stressed patients (Fig. 2c), with a mean concentration of 17.8 ± 10.8 μg/dl vs. 10.7 ± 3.3 μg/dl ($p = 0.0022$). Additional analysis of correlation between SIM results and serological parameters revealed a significant direct correlation between SIM values and the serum concentration of prolactin ($p = 0.0002$), ACTH ($p < 0.0001$), cortisol ($p = 0.0003$) and ECP ($p < 0.0001$). The direct correlation was also significant within each group (data not shown).

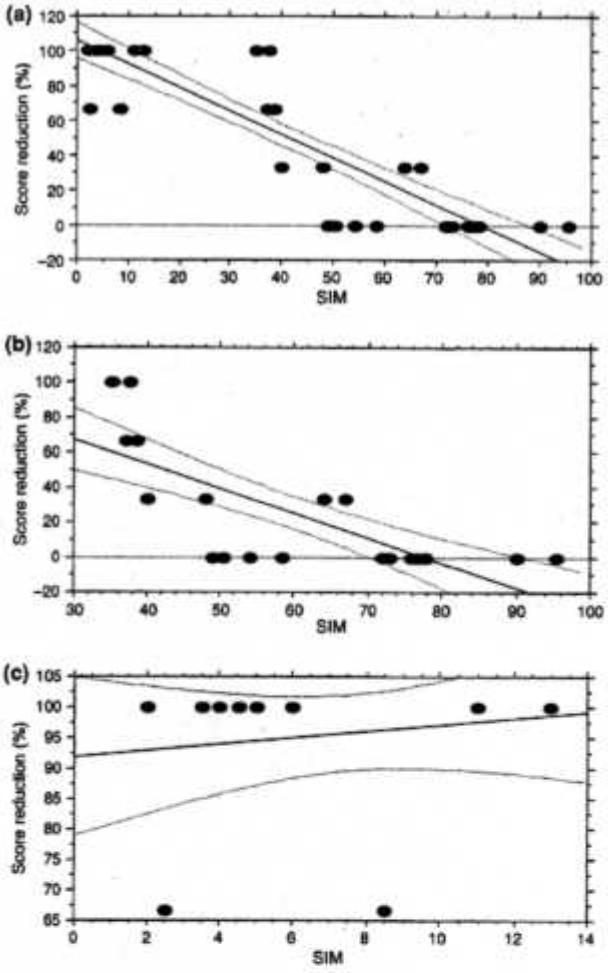

Fig. 1. Regression graphs of stress integrated measure (SIM) values and variation of symptom score. (a) shows the results for all patients, (b) for group 1 (stressed patients), and (c) for group 2 (non-stressed patients). See text for *r* and p-values.

Discussion

The specific immunotherapy of allergic diseases is perhaps the very first therapeutic strategy developed with the objective of modulating the immune response to environmental allergens, and its effectiveness and safety have been largely proven over the decades (20, 21). Specific immunotherapy appears to affect the Th2/Th1 ratio by 'reorienting' the immune system from a Th2-dependent reaction to allergens. Sublingual administration has proved as effective as subcutaneous injection (22), and SLIT has also been shown to modulate the Th2/Th1 balance and lower the serum concentration of IL-13 (13).

Despite all these positive clinical results, there are some patients who do not respond well to specific immunotherapy. Lack of response can depend on a whole range of host and

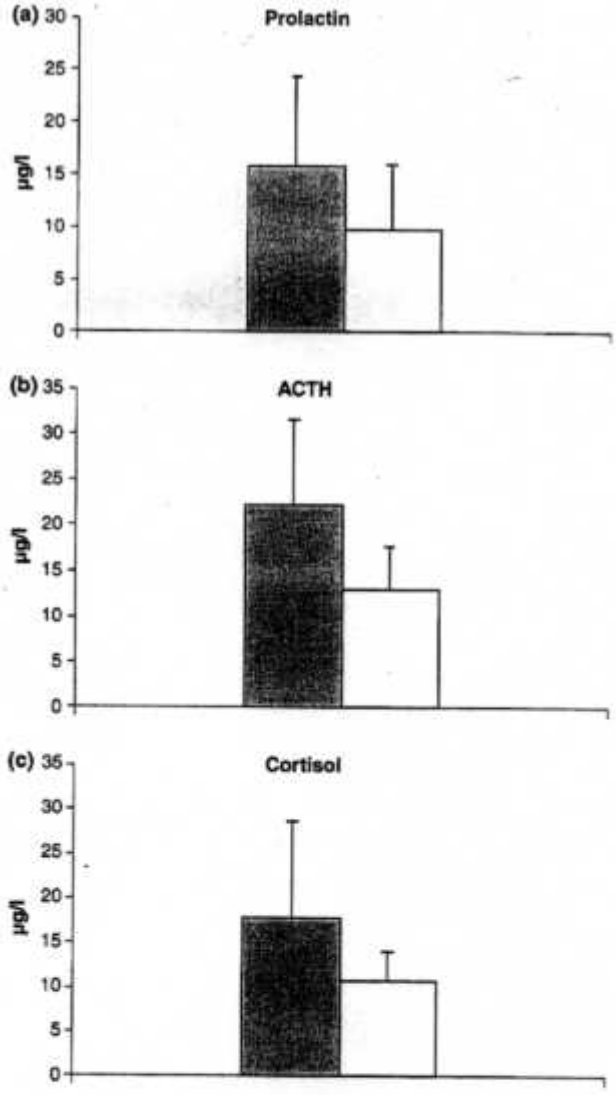

Fig. 2. Comparison between serum concentrations of prolactin (a), adrenocorticotropic hormone (b) and cortisol (c) in stressed (grey bars) and non-stressed (white bars) patients after 6 months of sublingual immunotherapy. See text for p-values.

environmental factors, but it is not yet clear why this should happen. Many studies have shown that the neuroendocrine system can affect immune responses by means of hormones and neuropeptides. Human lymphocyte responses can be modulated by hormones and neuropeptides produced by the nervous system and can synthesize hormones, such as ACTH, and neuropeptides, such as endorphins, from a precursor, pro-opio-melano-cortin, similar to that produced in the pituitary (12, 23). The lymphocyte has indeed been described on the basis of these findings as the 'internal sensitive organ or circulating mini-pituitary' (12). Moreover, the nervous and immune systems speak a common biochemical language and communicate via a circuit of shared ligands and their respective receptors (24).

The modulation of immune response by the neuroendocrine system also plays a role in allergic disease and the exacerbation of asthma (5, 25) and this interaction may also affect the response to immunomodulating therapies, such as vaccination with bacterial antigens (7). While mechanisms linking stress and mast cell function have been studied, the relationship between stress response and specific immunotherapy in allergy is in need of further investigation. There are in fact no studies into the effect of the neuroendocrine system on immunomodulating therapies for allergic diseases. In a previous study we observed a reduction of prolactin concentration after SLIT in allergic children (13), which may prove a key to better understanding of the influence of psychological stress and stressful conditions on the clinical outcome of specific immunotherapy.

The MSP test was used to evaluate this possible influence. The test, which was not designed for use with psychiatric patients, is a useful tool to evaluate psychological stress in 'normal' populations, particularly in research settings. Concomitant validity with immunity was also demonstrated by means of salivary immunoglobulin concentration (18). The patients' anxiety status was evaluated by means of the STAI scale, and the presence of anxiety as a habitual trait emerged in all cases. These results appear to rule out an acute state of anxiety. The difference between the two groups in the clinical outcome after 6 months of immunotherapy was remarkable. While the improvement in symptom score and PFT results was statistically significant in both groups, the clinical score was significantly lower in the non-stressed group at 6 months by comparison with the stressed group. The percentage of patients with a symptom score 0 was also significantly lower in the non-stressed group. The evaluation of diary cards showed a consistent perception of symptoms throughout the whole study period. In addition, the frequency of family psychosocial factors was significantly higher among stressed patients. This significant difference seems to reinforce the data from diary cards and the STAI scale results, indicating the persistence of stress conditions over time. Taken together, these facts seem to rule out the possibility that a momentary state of stress could account for a different perception of symptoms.

In addition, the two objective parameters of PEF and ECP, which show no statistical difference at baseline, are significantly improved among non-stressed subjects after SLIT. These results are consistent with recent studies

demonstrating the role of psychological stress and stressful conditions in exacerbating airway inflammation and the expression of atopic disorders (4, 5).

The only significant difference in the baseline characteristics of the two groups was the prevalence of patients also affected by rhinitis, which was higher among the non-stressed group. It has been largely demonstrated that SLIT is really effective on rhinitis subjects and that improvement in rhinitis may also lead to improvements in associated asthma (15, 20, 22). This possible bias on asthma results seems to be ruled out by the results of the restricted statistical analysis excluding rhinitis patients, the results of which proved similar to those of the analysis including all the patients studied. On the contrary, the lower p-value of the difference in FEV_1 values between the two groups after 6 months of SLIT in this restricted analysis seems to suggest a possible major role of stressing conditions on asthma. This possibility needs to be studied in further studies with larger groups of subjects.

It is also interesting that an inverse correlation was found in the stressed patients between SIM values and the percentage of symptom score reduction, whereas the correlation in the non-stressed patients was not significant. This may indicate that the presence of psychological stress can directly influence clinical outcome, while other factors play a role when no stress is present. Given that the SIM results were directly correlated with all the parameters measured in all our patients, it might be that psychological stress influences clinical outcome through the immunologic and neuroendocrine factors evaluated (prolactin, cortisol and ACTH) as well as others.

In actual fact, evidence has been accumulated suggesting that neuropetides such as PRL, ACTH and cortisol play a major role in modulating immune responses. In particular, it has been demonstrated that glucocorticoids are capable of influencing the Th1/Th2 balance. *In vitro* studies showed that a Th2 cytokine response by activated $CD4^+$ T cells was induced by means of pre-treatment with glucocorticoids, with increased mRNA of IL-4, IL-5 and IL-13 and diminished synthesis of IFNγ and TNFα (9). Glucocorticoids may work through this inhibition of Th1 response to protect the organism from systemic 'overshooting' with Th1/pro-inflammatory cytokines. Conditions associated with changes in glucocorticoids levels (such as acute of chronic stress, cessation of chronic stress, severe exercise, pregnancy and post-partum) may, however, affect the susceptibility to or the course of infections and of autoimmune and allergic diseases (10). In addition, it has been demonstrated that PRL has a stimulating function both in ontogenesis and in modulation of the immune system (26). *In vitro*, PRL induces an over-expression of the γ/δ TCR (27) and γ/δ+ T cells are essential for inducing IL-4 dependent IgE and IgG1 responses and for Th2-mediated airway inflammation (28). It was observed in a previous double-blind study that, in addition to its immunomodulatory effect on the Th1/Th2 balance, SLIT causes a decrease in PRL concentration among treated subjects (13). The higher PRL concentration observed among stressed subjects in the present study may represent a key linking the efficacy of SLIT to the stress status of patients.

Taken together, these findings could indicate the possibility of clinical outcome being influenced by psychological stress. No other clinical differences were registered between the two groups and the environmental factors (exposition to HDM) were also similar (data not shown). Many of the patients were children at an age when the influence of caregivers is of particular importance. Some studies have in fact suggested that parental stress may influence hormonal stress response in early childhood and constitute a predisposing factor of atopy (29). Our data on the higher prevalence of stressing psychosocial family factors among patients with poorer clinical response seems to be consistent with these observations, but they need to be further evaluated in larger populations of paediatric patients.

A chronic clinical condition such as allergy can of course contribute in turn to the development of psychological distress. A persistent antigen capable of causing chronic illness is similar to a persistent stressful psychosocial stimulus. Lymphocytes and monocytes/macrophages release cytokines, which modulate the CNS and therefore behaviour. This process is amplified during the course of illnesses, thus leading to 'sickness behaviour'. In the response to exogenous factors such as allergens, importance attaches to the concept of 'allostasis', defined as 'the capacity of individual to maintain stability through change' (30). Through allostasis, the autonomic nervous system, the hypothalamus–pituitary–adrenal axis (HPA), and the cardiovascular, metabolic and immune systems protect our organism from exogenous (psychosocial) and endogenous (antigenic) stress events. The allostatic load is defined as the wear and tear caused by this adjustment. Acute and chronic stress, stress, resulting respectively from the impeding of genetically derived fight-and-flight behaviour and the accumulation of minor stressful events,

provoke long-term sequelae because of allostatic load.

The two groups of patients displayed no significant differences in clinical features (symptoms, duration of diseases, previous use of SIT etc.), with the exception of a higher prevalence of associated rhinitis among non-stressed patients. As the analysis excluding all subjects with rhinitis also showed similar results, the differences observed in stress levels are therefore not explained by the disease itself and are probably because of the different ways in which individuals respond to exogenous and endogenous stimuli, including parental distress, coping with a chronic disease, distress caused by the impairment of daily activities due to asthma etc. The psychological stress derived from the high allostatic load perceived by some individuals may contribute to exacerbation of the disease as well as poorer response to immunomodulating therapy.

Our findings appear to be consistent with the view of Wright et al. (5) that psychological stress should be considered a social pollutant breathed into the body that increases the risk of atopy. Our finding in this connection was that chronic psychological stress can influence the response to SLIT, which may account for some response failures. Further studies are needed to clarify the possible role of stress in affecting the response to immunomodulating therapies. The evaluation of stress conditions may become useful as a prognostic co-factor of immunotherapy. Given both the fact that caregiver-related stress can play a key role in the case of children (29) and the higher frequency of stressing family conditions observed in our group of stressed patients, a study is now underway to clarify the role of parental distress on disease manifestation and response to therapies.

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