Oral tolerance attenuates changes in in vitro lung tissue mechanics and extracellular matrix remodeling induced by chronic allergic inflammation in guinea pigs


Departments of 1Medicine and 2Pathology, School of Medicine, University of São Paulo, São Paulo, Brazil; and 3Department of Pathology, The University of Vermont, Burlington, Vermont

Submitted 2 August 2007; accepted in final form 2 April 2008


Recent studies emphasize the presence of alveolar tissue inflammation in asthma. Immunotherapy has been considered a possible therapeutic strategy for asthma, and its effect on lung tissue had not been previously investigated. Measurements of lung tissue resistance and elastance were obtained before and after both ovalbumin and acetylcholine challenges. Using morphometry, we assessed eosinophil and smooth muscle cell density, as well as collagen and elastic fiber content, in lung tissue from guinea pigs with chronic pulmonary allergic inflammation.

Results

Baseline and postchallenge resistance and elastance were attenuated with oral tolerance (OT2 group) compared with the OVA group. Our results show that sublingual immunotherapy is beneficial for asthma, and its effect on lung tissue had not been previously investigated.

Methods

The costs of publication of this article were defrayed in part by the payment of page charges. The article must therefore be hereby marked “advertisement” in accordance with 18 U.S.C. Section 1734 solely to indicate this fact.

The animals weighed 300–350 g initially and were ∼3 wk old. All guinea pigs received humane care in compliance with the Guide for the Care and Use of Laboratory Animals (National Institutes of Health publication 85-23, Revised 1985), and all experiments de-
scribed in this study were approved by the institutional review board of the University of São Paulo (São Paulo, Brazil).

**Experimental model of pulmonary allergic inflammation.** Chronic airway inflammation was induced as previously described (29, 41). The guinea pigs were individually placed in a Plexiglas box (30 × 15 × 20 cm) coupled to an ultrasonic nebulizer (Soniclear, São Paulo, Brazil), and an aerosol of ovalbumin solution (grade V, Sigma Chemical, St. Louis, MO) diluted in 0.9% NaCl sterile solution (saline) was generated for 15 min or until respiratory distress occurred. The time that guinea pigs were in contact with aerosol was termed inhalation time. Respiratory distress was defined as the onset of sneezing, corrya, cough, and/or indrawing of the thoracic wall. The observer who made the decision to withdraw the animals from the inhalation box was not aware of the treatment status of the animal. This protocol consists of seven inhalations performed twice a week for 4 wk with increasing concentrations of ovalbumin (1–5 mg/ml) to avoid tolerance (Fig. 1). In inhalations 1–4 (first 2 wk), the dose of ovalbumin used was 1.0 mg/ml. In the 5th and 6th inhalation, animals received a solution with 2.5 mg/ml of ovalbumin, and in the 7th inhalation the dose was increased to 5.0 mg/ml of ovalbumin (20, 28, 29, 41).

Control animals received aerosolized saline. **Induction of oral tolerance.** Oral tolerance (OT) was induced by offering to the animals 2% of ovalbumin diluted in sterile drinking water (final dilution: 0.02 g/ml) ad libitum for 4 wk beginning concomitantly with the 1st inhalation (OT1 group) or beginning 24 h after the 4th inhalation (OT2 group). Every day we measured the water volume that was drunk by the animals, and the median value was 80 ml of ovalbumin solution during a period of 24 h. The animals of OVA and SAL groups (see Experimental groups) consumed the same sterile water volume. The tolerization process was maintained until the end of the experimental protocol (Fig. 1).

**Experimental groups.** Four groups of guinea pigs were studied: 1) animals that received inhalations with saline and drank sterile water for 4 wk (SAL, n = 11); 2) animals that received inhalations with ovalbumin and drank sterile water for 4 wk (OVA, n = 11); 3) animals that received inhalations with ovalbumin and drank ovalbumin 2% in sterile water for 4 wk beginning with the 1st inhalation (OT1 group, n = 11); and 4) animalsting that received inhalation with ovalbumin and drank ovalbumin 2% in sterile water for 2 wk beginning after the 4th inhalation (OT2 group, n = 11).

**Passive cutaneous anaphylaxis.** To evaluate if oral-induced tolerance interfered with sensitization to ovalbumin, we measured IgE and IgG1 antibodies by passive cutaneous anaphylaxis (PCA), as previously described (26, 42). Three guinea pigs from each experimental group were anesthetized with pentobarbital sodium (50 mg/kg ip) on day 28, and 5 ml of blood was obtained by cardiac puncture. Blood was allowed to clot at a temperature of 4°C and centrifuged at 1,000 g for 20 min. For the estimation of anaphylactic IgG1 antibodies, aliquots of serum were inactivated at 56°C in a water bath for 1 h to neutralize the activity of IgE antibodies. For estimation of IgE antibodies, serum was not heated, and a longer period of sensitization was used. The dorsal region of a normal guinea pig was shaved with an electric hair clipper, and care was taken to avoid irritation of the skin. Several subcutaneous injections (100 μl) of serum dilutions (from 1:5 to 1:1,280) were then made on each side of the dorsal skin with a hypodermic needle. After a sensitization period of 24 h for IgG1 antibodies and 10 days for IgE antibodies, the animals were challenged intravenously with 1 mg of ovalbumin in 0.25% Evans blue solution in saline. Thirty minutes after antigen injection, the animals were euthanized, the skin was inverted, the diameter was measured on the inner surface of the skin with a transparent ruler. The PCA titer was taken to be the highest dilution that presented a blue spot at least 10 mm in diameter.

**Oscillatory mechanics evaluation.** Seventy-two hours after the 7th inhalation, eight guinea pigs from each group were anesthetized with pentobarbital sodium (50 mg/kg ip) and tracheostomized, and after thoracotomy they were exsanguinated. The heart and lungs were excised en bloc, and the lungs were intratracheally infused with Krebs solution (in mM: 118 NaCl, 4.5 KCl, 25.5 NaHCO3, 2.5 CaCl2, 1.2 MgSO4, 1.2 KH2PO4, 10 glucose) (Sigma Chemical). Subpleural tissue strips of the lower lobes (10 mm × 2 mm × 2 mm) were cut. Although guinea pig pleural membrane has contractile cells, it is very difficult to remove it because it is very thin. Thus we decided to maintain the pleura in all animals to avoid a bias in case these membranes were partially removed. The resting length (Lr) and wet weight (Ww) of one strip of each animal were measured. Metal clips were glued to either end of the tissue strip with cyanoacrylate steel wire (diameter, 0.5 mm) were attached to the clips; one end was connected to a force transducer (model 404A; Cambridge Technologies), and the other end was connected to a servo-controlled lever arm (model 300B; Cambridge Technologies). The lever arm was capable of peak-to-peak length excursions of 8 mm and length resolutions of 1 μm and was in turn connected to a function generator (model 3030; BK Precision, Chicago, IL), which controlled the frequency, amplitude, and waveform of the oscillation. The resting tension (T) was set by movement of a screw thumb-wheel system, which effected slow vertical displacements of the force transducer. Length and force signals were low-pass filtered and converted from analog to digital with an analog-to-digital converter (model SCB-68/16 bits of resolution; National Instruments) and recorded on a compatible computer. The strips were preconditioned by slow cyclic tension from 0 to 2 g three times, and after the third cycle, they were fixed at 1 g and maintained in the organ bath containing Krebs solution aerated with 95% O2-5% CO2 for 50 min to allow stress relaxation. In this period of 50 min called stress relaxation, the Krebs solution was changed every 15 min. The frequency of oscillation and amplitude were adjusted, respectively, to 1 Hz and 2.5% of measured Lr. After these 50 min, the final resting tension was ~0.8 g.

The strips of all animals were challenged with ovalbumin (0.1%) (final dose 0.18 mg/ml) in the bath to obtain a maximal response (17, 27). After 10 min, Krebs solution was changed before adding Ach at 10−3 M (final dose: 0.1931 mg/ml) (Sigma Chemical) to the bath. Measurements of tissue resistance and elastance were obtained at baseline for 5 min, and after ovalbumin and Ach challenges in the bath in all experimental groups.

Resistance (R) and elastance (E) were estimated by the recursive least-squares algorithm to the equation of motion (18).

Fig. 1. Timeline of the experimental protocol. Guinea pigs received 7 inhalations (2 per wk during 4 wk) with aerosols of saline (SAL group) or ovalbumin solution (OVA group) with increasing doses of antigen. From the 1st to the 4th inhalations, animals received 1 mg/ml of ovalbumin (2 wk). In the 5th and 6th inhalations (3rd wk), animals inhaled with 2.5 mg/ml of ovalbumin, and in the 7th inhalation (beginning of the 4th wk) the dose of 5 mg/ml of antigen was used. The solution of ovalbumin or saline was continuously aerosolized either concomitantly with the 1st inhalation (OT1 group) or beginning 24 h after the 4th inhalation (OT2 group), and the other end was connected to a servo-controlled lever arm. The oral tolerance was induced by a solution of ovalbumin 2% diluted in drinking water for 2 wk and offered to guinea pigs ad libitum until the experimental day. Seventy-two hours after the 7th inhalation, all guinea pigs were anesthetized and exsanguinated, and the lungs were removed and submitted to oscillatory mechanics analysis.
where $T$ is tension, $l$ is length, $\Delta l/\Delta t$ is the length change per unit time, and $K$ is a constant reflecting resting tension. Results were standardized for strip size. The unstressed cross-sectional area ($A_0$) of the strip was obtained from the formula:

$$A_0 (\text{cm}^2) = W_0/p \times L_0$$

where $p$ is the mass density of the tissue, taken as 1.06 g/cm$^3$; $W_0$ is the wet weight in grams; and $L_0$ is the resting length in centimeters. Values of $R$ and $E$ were multiplied by $L_0/A_0$. Hysteresis, a dimensionless variable coupling the dissipative and elastic behavior, was calculated by the equation:

$$\eta = (R/E)2\pi f$$

where $f$ is frequency.

Immediately after mechanical oscillation, the strips were fixed in 10% formalin for 48 h, processed, and embedded in paraffin for histological analysis.

**Morphometric analysis.** A 5-μm-thick slide of lung strips was stained with hematoxylin and eosin for routine histological analysis and with Luna staining for eosinophils (23, 43). Lung tissue strips were also submitted to immunohistochemistry with anti-smooth muscle actin and stained with Sirius red for collagen and resorcin-fuchsin staining for elastic fibers (8). For immunohistochemistry, sections were deparaffinized, and a 0.5% peroxidase in methanol solution was applied for 10 min to inhibit endogenous peroxidase activity. Antigen retrieval was performed with citrate solution for 30 min. Sections were incubated with anti-human smooth muscle actin (1A 4, Dako) overnight at 4°C. LSAB Plus-HRP kit (K-0690 DAKO, Carpinteria, CA) was used as secondary antibody, and 3,3′-diaminobenzidine (DAB) (Sigma Chemical) was used as chromagen. The sections were counterstained with Harris hematoxylin.

By conventional morphometry, we analyzed the density of eosinophils within alveolar septa of lung strips. Using a 100-point grid with a known area (62,500 μm$^2$ at a ×400 magnification) attached to the ocular of the microscope, we counted the number of points hitting alveolar tissue in each field and the number of eosinophils within the alveolar septa. Eosinophil density was determined as the number of eosinophils in each field divided by tissue area. Measurements are expressed as cells per square micrometer. The results were then transformed to cells per square millimeter by adjusting the units (17). Counting was performed in 10 fields of lung strip in each animal.

The volume proportion of actin-positive cells in the lung strips was determined by dividing the number of points hitting actin-positive cells by the total number of points hitting the tissue. The volume proportion of collagen and elastic fibers in the alveolar tissue of lung strips was determined by dividing the number of points hitting collagen or elastin by the total number of points hitting alveolar septa. Measurements were performed in 10 fields of lung strip in each animal at a magnification of ×400. Results were expressed as a percentage of positive area per total area of tissue.

To ensure the homogeneity of the strips samples used, we quantified the volume proportion of small airways, vessels, and alveolar walls, using the same method of point counting described above, at a magnification of ×400. All morphometric measurements were performed by people blinded to protocol design. The results were expressed as a percentage.

**Data analysis.** Statistical analysis was performed using SigmaStat software (SPSS, Chicago, IL). The data were examined using ANOVA, the distribution was normal, the variances were homogeneous, and the multiple comparisons were made using Student-Newman-Keuls method (48). Values were expressed as mean and SE and with vertical bar graphics. $P < 0.05$ values were considered significant.

![Results](http://jap.physiology.org/)
groups studied. Considering the differences between baseline and maximal responses in each experimental group, we observed in all groups a significant increase in the values of lung tissue resistance and elastance ($P < 0.05$) only after ACh challenge compared with the baseline values. Neither saline group nor OT1 group had a significant increase in lung tissue resistance and elastance after ovalbumin challenge compared with their baseline values.

Comparing the baseline and maximal responses among the experimental groups, we noticed that at baseline and after ovalbumin and ACh challenges the values of tissue resistance and elastance were significantly higher in ovalbumin-exposed animals compared with saline-exposed animals ($P < 0.05$). Both oral-induced tolerance treatments (OT1 and OT2 groups) attenuated the maximal responses of tissue resistance and elastance after ovalbumin and ACh challenges compared with ovalbumin-exposed animals ($P < 0.05$). In addition, after ovalbumin challenge, the OT1 group also presented lower values of tissue resistance and elastance compared with the OT2 group ($P < 0.05$).

Concerning hysteresivity evaluation, there were no differences in either the baseline values or the data obtained after ovalbumin and ACh challenges among the four experimental groups (data not shown).

**Morphometric analysis.** Ovalbumin-exposed animals had presented significantly increasing eosinophilic inflammation compared with saline-exposed animals ($P < 0.01$). Oral induction of tolerance had decreased recruitment of these inflammatory cells ($P < 0.05$ vs. OVA, Fig. 3), and no difference was observed between OT1 and OT2 groups.

**ECM remodeling.** Figure 4, A and B, shows, respectively, the volume proportion of alveolar collagen and elastic fiber content in the four groups. We observed that ovalbumin-exposed animals presented an increase in both collagen (Fig. 4A) and elastic fiber content (Fig. 4B) ($P \leq 0.01$) compared with saline-exposed animals. Considering the oral tolerance groups, both treatments (OT1 and OT2 groups) attenuated the deposition of collagen and elastic fibers in alveolar septa ($P < 0.05$) compared with animals exposed to ovalbumin that received sterile drinking water (OVA group).

There was no significant difference in the actin content in lung strips among the four experimental groups (SAL group $13.77 \pm 0.69\%$; OVA group $16.06 \pm 1.47\%$; OT1 group $14.78 \pm 1.31\%$; OT2 group $12.81 \pm 1.11\%$). We observed that 60–70% of the fractional area of lung strips was represented by alveolar septa. There were no significant differences in the contents of airways (SAL group $3.97 \pm 0.9\%$, OVA group $2.61 \pm 0.65\%$, OT1 group $1.7 \pm 0.6\%$, OT2 group $1.4 \pm 0.3\%$), vessels (SAL group $1.1 \pm 0.4\%$, OVA group $1.6 \pm 0.4\%$, OT1 group $1.9 \pm 0.0\%$, OT2 group $0.2 \pm 0.1\%$), and alveolar tissue (SAL group $74.4 \pm 2.6\%$, OVA group $52.4 \pm 9.0\%$, OT1 group $68.3 \pm 5.8\%$, OT2 group $74.3 \pm 3.1\%$) among the experimental groups.

Figure 5 shows photomicrographs of lung tissue strips from animals exposed to ovalbumin (OVA group) that show intense eosinophilic infiltration (Fig. 5D) and increase in collagen density (Fig. 5E) and elastic fiber content (Fig. 5F) compared with saline-exposed animals (SAL group; Fig. 5A–C). In both groups of induced oral tolerized animals, there was a decrease in the eosinophilic infiltration (OT1, Fig. 5G; OT2, Fig. 5J). Considering the ECM remodeling we noticed that in tolerance animals the collagen (OT1, Fig. 5H; OT2, Fig. 5K) and elastic fiber (OT1, Fig. 5I; OT2, Fig. 5L) contents were...
attenuated compared with the ovalbumin-exposed nontolerized guinea pigs.

DISCUSSION

The present study shows that inducing oral tolerance attenuates peripheral lung tissue responsiveness, eosinophilic inflammation, and ECM remodeling in an experimental model of chronic allergic pulmonary inflammation, suggesting that this approach could attenuate or prevent distal lung functional and structural changes induced by chronic allergic inflammation.

Although some studies propose the importance of inducing oral tolerance in the control of asthma pathological alterations and this type of treatment has been used in humans, most of the studies evaluated only the effects on airways. Considering the importance of lung periphery in asthma and the relevant results obtained using immunological tolerance in airways (4, 32, 33, 34, 35), studies that evaluate the efficacy of this treatment in lung parenchyma are relevant. There were no previous studies that evaluated the effects of oral tolerance in lung periphery responses in an experimental model of chronic lung inflammation. We know from previous studies that this experimental model presents some important features of asthma pathophysiology, as intense bronchoconstriction, airways eosinophilic and lymphocytic inflammation, and airway and bronchial vessels remodeling (28, 29, 41). Allergic asthmatics have established antigen-responsive CD4+ T cells in the airways and local lymphoid tissue. Children predisposed to develop asthma often have antibodies to common allergens, indicating that their immune system is already primed to the allergen. Based on animal models of asthma, one might speculate that feeding allergens to allergic asthmatics would increase inflammation, which can be dangerous. Thus animal models might convince us that attempts to stimulate...
mucosal tolerance should be used to prevent or treat asthma (5).

The role of lung periphery in asthma pathophysiology has been intensely investigated in the last several years. Several authors have shown that there is an inflammation and remodeling process in lung periphery as well as in central airways (2, 16, 47). The lung periphery strips have been used to study the mechanical and pharmacological properties of the lung periphery (22, 46). We have recently demonstrated that in this experimental model of chronic allergic inflammation, the lung periphery is involved not only in the early but also in the late inflammatory response (17).

The pharmacology industries are pursuing the new technology to develop drugs that also act in lung periphery, and, in this regard, HFA formulation has been shown as a promising medication (25). The effects of HFA have been shown to reduce the peripheral pulmonary inflammation, as previously discussed in a recent study performed by Bergeron et al. (2). They showed that the HFA administration in asthmatic patients did not reduce total collagen deposition and the collagen type III content in peripheral airways. However, the authors did not evaluate the lung parenchyma alterations.

We used two different types of oral tolerance. In the first protocol we began the tolerance induction together with the sensitization protocol. The aim of this protocol is to evaluate a preventive strategy. Our results suggested that this approach is sufficient to avoid the alterations observed in this experimental model. After the 4th inhalation, guinea pigs had already shown pulmonary eosinophilic infiltration, as well as ECM alterations (unpublished results). We had also previously demonstrated (20) that at this moment (after the fourth ovalbumin inhalation) the animals had an increase in the IgG1 antibody titles (1:320). For this reason, we also evaluated if oral-induced tolerance may be used as a therapeutic approach, reverting the inflammatory and remodeling processes. Then, in the second protocol, the animals received the oral antigen beginning 24 h after the 4th ovalbumin inhalation. Furthermore, we initially evaluated the effects of these protocols of oral-induced tolerance in inflammatory, remodeling, and functional alterations in airways of guinea pigs with chronic allergic inflammation. Ruiz et al. (32) showed that both tolerization protocols attenuated these alterations.

Acute responses to antigen exposure were evaluated by the time that guinea pigs were able to be in contact with the aerosol of ovalbumin (inhalation time). In this context, repeated inhalations of increasing concentrations of ovalbumin progressively reduced these measurements after the 5th inhalation. Both oral-induced tolerance treatments augment the inhalation time in sensitized animals, suggesting that oral tolerance contributes to the control of the acute responses to antigen challenge.

Although in vivo apparatus of oscillatory mechanics permits the evaluation of large and small airways, the oscillatory mechanics in vitro provides a tool for the specific evaluation of lung periphery with the minimum interference of the compartment represented by the small airways. In addition, we argued that this method in vitro permits a specific analysis of the effects of oral tolerance in lung periphery avoiding other compensatory mechanisms that could be activated in in vivo studies.

Considering the differences between baseline and maximal responses in each experimental group, we observed in all groups a significant increase in the values of lung tissue mechanics only after ACh challenge compared with the baseline values. It is interesting to notice that neither saline group nor OT1 group had a significant increase in lung tissue mechanics after ovalbumin challenge compared with their baseline values. The comparison among the experimental groups showed that, at baseline and after either ovalbumin or ACh challenges, the values of lung tissue resistance and elastance were attenuated by both oral-induced tolerance treatments (OT1 and OT2 groups) compared with ovalbumin-exposed animals (P < 0.05).

These results suggested that oral-induced tolerance treatments were able to attenuate the mechanical changes in lung periphery induced by chronic inflammation, particularly by specific antigen challenge. In these experimental models the tolerization process was antigen specific, and the mechanical responses confirmed this statement.

The animals that received the first protocol presented a lower response after ovalbumin challenge compared with animals that received the second protocol. The differences could be related to the fact that in the first protocol, animals received the oral ovalbumin for 4 wk, although in the second protocol animals received this treatment only for 2 wk. It is interesting to point out that there is no difference between the titles of anaphylactic antibody (IgG1) between the different protocols used. Both OT1 and OT2 presented the same title of IgG1 (1:40). Unfortunately, the PCA was a semiquantitative method for antibody title quantification that may not have detected minor differences in antibody titles between tolerized groups. In addition, neither the eosinophilic inflammation nor the collagen and elastin content could explain these differences, since they were similar in both OT1 and OT2 groups.

It is well known that the tolerization process attenuates B-cell responses. Hasegawa et al. (12) demonstrated that B-cells have been implicated in myofibroblast activation mainly by secreting IL-6, IL-9, and fibroblast growth factor. In this regard, it is important to consider that myofibroblasts are one of the contractile elements that modulate lung parenchyma responses (21, 22, 45).

The morphometric analysis of the fractional area of lung constituents within the strips (alveolar wall, blood vessel wall, and bronchial wall) showed that there is no difference in the percentage of each structure in the experimental groups, corroborating the idea that the differences in the contractile responses observed among the experimental groups were not due to a variation in the structural components of the lung studied. Several authors have discussed the importance of these structures in the mechanical behavior of lung tissue, including the consequences of stiffening of the ECM network, as well as of elastin and collagen digestion in these responses (36, 39, 47). We studied strips obtained from subpleural region. In the lung tissue used there was a scanty number of bronchial and blood vessels (<30%). Dolhnikoff et al. (7) demonstrated that the human strips responded to contractile stimulation with increases in the dynamic mechanical parameters such as tissue elastance and resistance. These increments were found regardless of whether strips contained or were devoid of small airways, arguing that contractile responses occur at the tissue level.
Recent studies suggest that mechanical properties of lung tissue in baseline conditions may be actually dominated by the connective tissue fiber network, whereas interstitial cells play a less significant role. Romero et al. (31) concluded that pneumoconstriction significantly modifies the intrinsic mechanical properties of the connective matrix by means of a mechanism differing from that of passive stretching. In fact it could be accepted that the contractile cells are able to modulate the mechanical properties of the connective matrix.

The actin content is similar among all experimental groups. This result differs from others that evaluated airway walls and observed an increase in actin content after sensitization (45, 46). Lanças et al. (17), using the same animal model, did not find any differences in the total actin content in saline- and ovalbumin-exposed animals. The authors showed an increase in the lung tissue mechanics after either ovalbumin or ACh challenges. It is important to consider that the actin evaluation was used as a marker of smooth muscle protein, which stained the alveolar duct, airway, and vascular walls, as well as myofibroblasts.

Human studies demonstrated that the eosinophilic infiltration is widely distributed within the respiratory tract, from the nasal mucosa to the distal lung (6, 16). In this experimental model, we previously showed that there is an increase in eosinophils density not only in airways but also in lung tissue (17).

The macromolecules that are most important in determining the mechanical properties of the lung are collagen, elastin, and proteoglycans (39). ECM alterations represent one part of the remodeling process that may have important clinical repercussions on chronic asthma responses influencing the decrease in lung function observed in some asthmatic patients (13). However, the intensity of the response of eosinophilic infiltration and ECM remodeling is greater in airway wall than in lung alveolar parenchyma (16, 17, 41). In the present study we observed that animals submitted to chronic inflammatory stimuli have an increase in the lung periphery collagen and elastic fiber content, and the tolerance process attenuated these alterations.

The present data in this article showed that both oral tolerance protocols reduced the eosinophil density as well as the collagen and elastic fiber content in lung parenchyma. Eosinophil-derived cytokines were involved in the modulation of T(H)2 responses that trigger macrophage production of transforming growth factor-β1, which serves as a stimulus for ECM production (10, 13, 23). Our study corroborates this statement since the reduction in eosinophilic inflammation was associated with a decrease in collagen and elastic fiber content.

In conclusion we observed that oral tolerance attenuates lung tissue hyperresponsiveness, which is associated with attenuation in eosinophil inflammation and ECM collagen and elastic fibers deposition. Although the mechanism involved in these responses need to be further investigated, these findings can open a therapeutic strategy to prevent and/or treat the peripheral lung alterations in asthmatic patients.

ACKNOWLEDGMENTS

We thank Dr. Adenir Perini for assistance in passive cutaneous anaphylaxis techniques. This study was presented in part at the International Meeting of the American Thoracic Society in Orlando, 2004, and at the International Meeting of the European Respiratory Society in Glasgow, 2004.

GRANTS

We are grateful to Brazilian Scientific Agencies Conselho Nacional de Desenvolvimento Científico e Tecnológico (CNPq), Fundação de Amparo à Pesquisa do Estado de São Paulo (FAPESP), Programa de Núcleos de Excelência (PRONEX-MCT), and Laboratório de Investigação Médica do Hospital das Clínicas da Faculdade de Medicina da Universidade de São Paulo (LIM 20) for financial support.

REFERENCES

ORAL TOLERANCE AND LUNG INFLAMMATION


