

# Immediate hypersensitivity and serum IgE antibody responses in patients with dermatophytosis

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## Summary

**Background:** The association of dermatophytes with atopic patients and improvement in allergic signs with antifungal treatment suggest a possible link between chronic infection and atopy.

**Objective:** The purpose of this study was to determine skin reactivity and serum IgE antibody responses in patients with chronic and acute dermatophytosis.

**Methods:** One hundred and sixty-three patients with chronic dermatophytosis, 35 patients with acute dermatophytosis, 41 atopic patients and 49 healthy subjects were enrolled in this study. Sensitization to *Trichophyton mentagrophytes* (*T. mentagrophytes*), *Candida albicans* and *Aspergillus fumigatus* antigens has been evaluated in patients by skin prick test (SPT) and by the presence of specific IgE antibody in enzyme-linked immuno-sorbent assay (ELISA).

**Results:** Positive immediate hypersensitivity (IH) reactions were obtained in 95.1% of the atopic patients with chronic infection for *T. mentagrophytes*, representing a significant difference from other patient groups ( $P < 0.05$ ). Specific anti-*T. mentagrophytes* IgE antibodies were detected in atopic patients with chronic (65.9%) and acute (50%) dermatophytosis, while none of the atopic subjects had positive IgE reactions to *T. mentagrophytes*.

**Conclusion:** The results showed significant higher positive IH and specific anti-*T. mentagrophytes* IgE responses in atopic patients with chronic dermatophytosis than the other groups. (*Asian Pac J Allergy Immunol* 2012;30:40-7)

**Key words:** dermatophytosis, *Trichophyton mentagrophytes*, atopy, skin prick test, specific IgE antibody.

## Introduction

Infections caused by the dermatophytes, dermatophytosis, are common in many parts of the world. These organisms can invade the keratinized tissues of the stratum corneum, hair and nails and produce the clinical pattern known as tinea or ringworm. The etiologic agents of dermatophytosis are classified into three genera based primarily on differences in microscopic morphology and modes of sporulation as *Epidermophyton*, *Microsporum* and *Trichophyton*.<sup>1</sup> Dermatophytes that cause infections can vary from country to country and from region to region, creating a specific disease spectrum for that region. Different factors have to be taken into consideration when the distribution and importance of dermatophytes are studied at each location. These include life style, population, climate, presence of animals infected with zoophilic dermatophytes and human migration.<sup>2,3</sup>

Infections by dermatophytes induce a specific immune response, with humoral and cellular components. Studies suggest that the nature of the underlying immune response to dermatophyte antigens is related to the severity of dermatophytosis; immediate hypersensitivity (IH) skin tests are associated with chronic recurrent infections characterized by low-grade inflammatory lesions and the presence of IgE antibodies.<sup>4</sup> By contrast, delayed hypersensitivity (DH) reactions are associated with highly inflamed lesions that resolve spontaneously and a resistance to re-infection.<sup>5</sup> However, the immune response that is raised, and especially the degree of inflammation, varies according to the dermatophyte species, to the host species and to the

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pathophysiological status of the host.<sup>6</sup> The different ways in which dermatophytes may counter the immune system, or induce damage via immune defenses are as follows: lymphocyte inhibition by cell wall mannan, macrophage function alteration, differential activation of keratinocytes and differential secretion of proteases.<sup>7,8</sup>

The complex relationship existing between dermatophytosis and allergic diseases has to be discussed here. The predisposing role of atopy in chronic dermatophytosis is not clearly established; on the other hand, it is now suggested that chronic dermatophytosis, associated with IH in skin test reactions and Th<sub>2</sub> cytokines, can contribute to the pathogenesis of allergic diseases.<sup>9</sup> The association of *Trichophyton* with atopic patients and improvement in allergic signs with antifungal treatment suggest a possible link between chronic infection and atopy.<sup>10</sup> In 1980, Jones described the association of allergic rhinitis or asthma with chronic tinea infections, increased total IgE, *Trichophyton*-specific IgE levels and IH as the “atopic-chronic dermatophytosis syndrome”.<sup>11</sup> The identification of the mechanism which induces the control of disease is an interesting point to study in the immunology of dermatophytosis. The purpose of the present study was to determine the allergic immunological responses to *Trichophyton mentagrophytes* (*T. mentagrophytes*), *Candida albicans* (*C. albicans*) and *Aspergillus fumigatus* (*A. fumigatus*) antigens in patients with and without dermatophytosis.

## Methods

### *Patients and healthy subjects*

A total of 239 subjects, 163 patients with chronic dermatophytosis (infection lasting more than 6 months); 35 patients with acute dermatophytosis (infection lasting less than 6 months) and 41 atopic patients without dermatophytosis, were enrolled between 2010 and 2011 in this study. A structured questionnaire was administered to clinically suspected dermatophyte infected individuals by interview. The diagnostic criteria included annular lesions with activity at the edges; scaling patches along with erythema; onycholysis (nail plate separation from nail bed), and thickened, discolored (white, yellow, brown and black) broken and dystrophic nails. The diagnosis of atopy was based on the criteria of Hanifin and Rajka<sup>12</sup> and accurate assessment of the severity of atopy was performed using the SCORAD index according to Stalder et al.,<sup>13</sup> which was determined by a dermatologist from the Department

of Dermatology, Imam Khomeini Hospital, Tehran, Iran. Inclusion criteria for atopic patients were diagnosis according to the U.K. working party criteria and skin lesions not only restricted to the hands.<sup>14</sup> Exclusion criteria for the participants in the study were skin diseases other than those being investigated, autoimmune diseases, immune deficiencies, malignant diseases, pregnancy or lactation, immunosuppressive treatment and age below 15 or above 80 years. The controls comprised 49 healthy volunteers without clinical symptoms or history of allergy. The use of systemic glucocorticoids, systemic antifungal treatment or ultraviolet therapy was not allowed for 2 months before the investigation. Topical antifungal treatment was not permitted for 1 month before and topical corticosteroids were not allowed on the test sites for 1 week before the study. Antihistamines were withdrawn 5 days before the investigation. The study was approved by the Regional Ethics Committee of Dermatology Center of Tehran, Iran and all the subjects were examined after their informed consent.

### *Sampling and fungal cultivation*

The affected areas were swabbed with 70% alcohol. Skin lesions were sampled from the erythematous, peripheral, actively growing margins of the lesions, while nail samples were taken from deeper part of the discolored or dystrophic parts of the nails. Microscopic examination was done using 10% KOH solution to see arthroconidia and hyphal segments in the skin and nail specimens. Another part of sample was inoculated onto sabouraud dextrose agar (SDA, Merck Co., Darmstadt, Germany) containing chloramphenicol (50 mg/l) and cycloheximide (500 mg/l). The plates were incubated at 28°C for four weeks and examined at intervals for evidence of fungal growth. The media not showing growth for four weeks were discarded. Any visible growth from SDA was examined for colony morphology, texture, surface pigmentation and pigmentation on the reverse. The isolated dermatophytes were identified on the basis of their colony morphology and microscopic examination with lactophenol cotton blue preparation. Pigment production on corn meal agar, urease activity and the hair perforation test were performed as well.

### *Cell fractionation and crude antigen preparation*

In this study, *T. mentagrophytes* with *A. fumigatus* and *C. albicans* were selected for evaluating the immunologic responses in different patient groups.



*A. fumigatus* (af1023) and *C. albicans* (ca512) isolates were obtained from the Collection of Mycology Research Center, University of Tehran and cultured on SDA (Merck Co., Darmstadt, Germany) containing chloramphenicol (50 mg/l) at 37°C for 2-7 days. *T. mentagrophytes* (Tm11) was grown on SDA (Merck Co., Darmstadt, Germany) containing chloramphenicol (50 mg/l) and cycloheximide (500 mg/l) at 28°C for four weeks. A suspension of wet fungal colonies was prepared in lysing buffer (62.5 mM Tris, 1 mM Dithiothritol, 0.2 mg/ml of Phenyl sulfonyl fluoride and 15% Glycerol, pH 6.8) and then subjected to cell disruption in two consecutive steps using glass beads (diameter; 1 mm) on a vortex mixer for 15 minutes with 5-minute intervals and then using the sonication method (*Sonicator set, UP 200 s, dr. hielscher sonicator set, Germany*). Cell disruption was carried out using 60% amplitude for 48 min (2:4 min pulse on:off basis) to achieve maximum disruption of cells. After cell disruption, the crude antigens were separated from other cell components by centrifugation at 3000 g for 20 min and subsequently ultracentrifuged at 35000 g for 1 h. The antigenic extracts were sterilized using a filter (0.2 µ, *Sartorius, USA*), lyophilized by means of freeze-dryer (*Labconco, USA*) and stored at -20°C until used.

#### Determination of protein

The protein concentration of each sample was determined after reconstituting it in 50 and 100 µl of distilled water by Bradford method.<sup>15</sup>

#### Skin prick test (SPT)

Fungal antigens were diluted with 0.05 M PBS buffer, pH 7.4, containing 50% glycerin and then sterilized by means of a filter (0.2 µ, *Sartorius, USA*). Skin tests were performed in both patients and healthy subjects. It was confirmed that neither the patient group nor the healthy group had received antihistamines prior to undergoing the skin tests. Skin prick testing was performed with 3×10<sup>-6</sup> ml of the standardized antigens (protein concentration, 20 µg/ml) along with negative controls (0.05 M PBS buffer, pH 7.4, containing 50% glycerin) and positive controls (0.1% histamine) on the volar aspect of the forearm and pricked with a 27 gauge needle. The tests were read after 15 min and any reaction with a mean wheal diameter at least 3 mm greater than negative control was taken as positive.

#### ELISA assay

The total IgE level was determined by indirect-ELISA test using commercial kits (*Immuno-Biological. Laboratories, Hamburg, Germany*), according to manufacturer's protocol. The specific serum IgE antibodies for fungal antigens in patients' sera were detected as previously described.<sup>16</sup> Briefly, fungal antigens were diluted in 0.05 M carbonate-bicarbonate buffer, pH 9.5, and a suitable titre was chosen using checker board titration. One hundred microliters of the antigen (30 µg/ml) was added to the wells and stored overnight at 4°C. The wells were rinsed extensively with washing buffer (PBS-Tween 20, 0.05%) and then 150 µl of blocking buffer was added to the wells and the microplate was stored at 37°C for 2.5 h. Then the wells were rinsed again. Serum samples were diluted 1:4 in PBS-Tween 20-BSA 1% and 100 µl were added to each well and incubated with shaking at 37°C for 2 h, followed by washing with PBS-Tween 20, 0.05%. Anti-human IgE conjugated with ALP (*Sigma, St. Louis, USA*) for IgE assay was diluted 1/2000 and 1/4000, respectively, in PBS-Tween 20-BSA 1% and 100 µl was added to each well and incubated at 37°C for 1.5 h, followed by washing with PBS-Tween 20-BSA 1%. Each well received 100 µl of a solution of 0.1 M diethanolamine (DEA), pH 9.8, 1 mM magnesium chloride and paranitrophenol phosphate (PNPD). After 30 min, the reaction was stopped with 50 µl of 3 M NaOH and the OD was read at 405 nm. Each sample was tested in duplicate. Sera from the control group were used as negative controls. OD405 values more than the OD value of the mean obtained with controls ± 3 standard deviations (SD<sub>s</sub>) were considered positive.

#### Statistical analysis

The prevalence of positive SPT and ELISA tests for each individual antigen was computed using simple percentages. Proportions were compared using Fisher's exact test. Correlation between SPT and specific serum IgE was calculated using a 2-tailed Spearman's rho with statistical significance determined using the *t* test. Statistical significance was considered a *P* value less than 0.05.

#### Results

A total of 163 and 35 cases were positive with chronic and acute dermatophytosis, respectively. The age and sex distributions of the different patient groups and the healthy subjects are presented in Table 1. The male sex (58.9%) was more affected

**Table 1.** Frequency distribution of patients with chronic and acute dermatophytosis, atopic patients and healthy subjects under study with respect to age and sex characteristics (No., %).

Age	Chronic dermatophytosis		Acute dermatophytosis		Atopic patients		Healthy subjects	
	male	female	male	female	male	female	male	female
15-29	17 (17.7)	4 (20.9)	5 (29.5)	4 (22.2)	4 (17.4)	4 (14.8)	6 (22.2)	5 (22.7)
30-44	28 (29.2)	19 (28.4)	4 (23.5)	5 (27.8)	6 (26.1)	4 (14.8)	18 (100)	6 (27.3)
45-59	26 (27.1)	18 (26.9)	4 (23.5)	5 (27.8)	6 (26.1)	5 (18.5)	18 (100)	6 (27.3)
60-80	25 (26)	16 (23.8)	4 (23.5)	4 (22.2)	7 (30.4)	5 (18.5)	5 (18.4)	5 (22.7)
total	96 (100)	67 (100)	17 (100)	18 (100)	23 (100)	18 (100)	27 (100)	22 (100)

with chronic dermatophytosis than their female (41.1%) counterparts clinically. The age ranges were from 15 to 80 years old (mean age: 62.5 years) with a duration of infection from 3 to 19 years (mean duration: 8.7 years) in patients with chronic infection and also from 15 to 41 years old (mean age: 28 years) with duration of infection from 1 to 4 months (mean duration: 2.2 months) in patients with acute infection. The 45-59 year age-group was the most commonly infected age-group with chronic dermatophytosis.

The most common affected area in patients with chronic infection was tinea unguium (48, 29.4%), followed by tinea unguium altogether with tinea pedis (66, 40.5%), tinea unguium altogether with tinea manuum (14, 8.6%), tinea unguium altogether with tinea cruris (11, 6.7%), tinea pedis (15, 9.2%) and tinea corporis (9, 5.5%). The most frequent dermatophyte isolated from this patient group was *T. mentagrophytes* (119), which accounted for 73% of all dermatophytes recovered. It was followed by *T. rubrum* (27, 16.6%), *Epidermophyton floccosum* (*E. floccosum*) (9, 5.5%) and *T. violaceum* (8, 4.9%). The most common clinical diagnosis in patients with acute dermatophytosis was tinea corporis (25, 71.4%), followed by tinea cruris (6, 17.1%) and tinea barbae (4, 11.4%). From these patients, *Microsporum canis* (*M. canis*) (17, 48.6%), *T. verrucosum* (11, 31.4%) and *T. mentagrophytes* (7, 20%) were isolated.

In this study, a total of 239 patients and 49 healthy subjects were screened with SPT. Of patients with dermatophytosis, 41 (17.2%) and 2 (0.8%) cases were atopic patients with chronic and acute infections, respectively. Seventy-two (44.2%) out of 163 patients with chronic infection showed significantly more positive reactions in SPT to one or more fungal strains than other groups ( $P < 0.05$ ). Among them, atopic patients with chronic infection

(39, 95.1%) were significantly more susceptible to *T. mentagrophytes* than *C. albicans* and *A. fumigatus* antigens ( $P < 0.05$ ). No significant differences were observed in atopic patients with acute dermatophytosis and atopic patients according to the fungal isolates. There were no reactions to *A. fumigatus* in both atopic patients with acute infection and other atopic patients. Only one healthy subject (2%) had a positive response on SPT to *C. albicans* antigen (Table 2).

A positive total IgE result was defined as a value of at least 100 IU/ml. As illustrated in Table 2, positive sera were recorded in 29 (70.7%) of the atopic patients with chronic dermatophytosis and 19 (46.3%) of the atopic patients, representing significant differences with other groups ( $P < 0.05$ ). Concerning the specific IgE antibody assay, the mean OD ( $405 \pm 3$  SD<sub>s</sub>) of healthy subjects was calculated to be 0.25 for *T. mentagrophytes*-specific serum IgE, 0.49 for *C. albicans*-specific serum IgE and 0.45 for *A. fumigatus*-specific serum IgE. Positive sera were defined as those with OD<sub>s</sub> of more than 0.25, 0.49 and 0.45. Of 163 sera obtained from patients with chronic infection, 27 (65.9%) were atopic cases with positive specific IgE responses to at least one fungal isolate. *T. mentagrophytes* (19%) was the most prominent allergen showing a positive reaction, followed by *C. albicans* (7.4%) and *A. fumigatus* (3.7%). The frequencies of specific IgE antibodies to different fungal isolates in other groups were as follows: *T. mentagrophytes* (5.8%) and *C. albicans* (5.8%) in atopic patients with acute infection and *C. albicans* (12.2%) and *A. fumigatus* (4.9%) in atopic patients. None of the healthy subjects had positive IgE reactions to fungal antigens. Only one person (2%) had a positive response for the specific IgE antibody against *C. albicans* antigen.

**Table 2.** Frequency of positive skin prick tests (immediate hypersensitivity), total IgE and specific IgE -ELISA tests to *Trichophyton mentagrophytes*, *Candida albicans* and *Aspergillus fumigatus* antigens in subjects with and without dermatophytosis (No., %)

Group	No.	Skin prick test			Specific IgE			Total IgE
		<i>T. mentagrophytes</i>	<i>C. albicans</i>	<i>A. fumigatus</i>	<i>T. mentagrophytes</i>	<i>C. albicans</i>	<i>A. fumigatus</i>	
<b>Chronic dermatophytosis</b>								
- Atopy	41	39 (95.1)	11 (26.8)	3 (7.3)	27 (65.9)	7 (17.1)	2 (4.9)	29 (70.7)
- SLE *	6	1 (16.7)	2 (33.3)	0 (0)	1 (16.7)	2 (33.3)	0 (0)	1 (16.7)
- Corticosteroids	14	0 (0)	0 (0)	0 (0)	0 (0)	0 (0)	0 (0)	0 (0)
- Pemphigus vulgaris	2	0 (0)	0 (0)	0 (0)	0 (0)	0 (0)	0 (0)	0 (0)
- Organ transplantation	6	0 (0)	1 (16.7)	1 (16.7)	0 (0)	0 (0)	1 (16.7)	0 (0)
- AIDS	5	0 (0)	0 (0)	0 (0)	0 (0)	0 (0)	0 (0)	1 (20)
- Diabetes mellitus	13	2 (15.4)	2 (15.4)	0 (0)	1 (7.7)	2 (15.4)	0 (0)	4 (30.8)
- Non- risk factors	76	3 (3.9)	3 (3.9)	4 (5.3)	2 (2.6)	1 (1.3)	3 (3.9)	3 (3.9)
- Total	163	45 (27.6)	19 (11.7)	8 (4.9)	31 (19)	12 (7.4)	6 (3.7)	38 (23.3)
<b>Acute dermatophytosis</b>								
- Atopy	2	1 (50)	1 (50)	0 (0)	1 (50)	1 (50)	0 (0)	0 (0)
- Diabetes mellitus	2	1 (50)	1 (50)	0 (0)	1 (50)	1 (50)	0 (0)	0 (0)
- Non- risk factors	31	0 (0)	0 (0)	0 (0)	0 (0)	0 (0)	0 (0)	0 (0)
- Total	35	2 (5.7)	2 (5.7)	0 (0)	2 (5.7)	2 (5.7)	0 (0)	0 (0)
<b>Atopic patients</b>	41	2 (4.9)	3 (7.3)	0 (0)	0 (0)	5 (12.2)	2 (4.9)	19 (46.3)
<b>Healthy subjects</b>	49	0 (0)	1 (2)	0 (0)	0 (0)	0 (0)	0 (0)	0 (0)

\* SLE: Systemic lupus erythematosus

OD 405 values greater than the OD value of the mean with healthy controls  $\pm$  3 SD mentioned as positive; *Trichophyton mentagrophytes*: 0.25, *Candida albicans*: 0.49, *Aspergillus fumigatus*: 0.45

Significant correlations were found between the SPT and specific serum IgE results for *T. mentagrophytes* tested in both atopic patients with chronic and acute infections ( $P < 0.05$ ). Significant positive correlations were found between the SPT reactions and *A. fumigatus*-specific serum IgE in atopic patients with chronic infection as well as between the SPT reaction and *C. albicans*-specific serum IgE in atopic patients with acute infection ( $P < 0.05$ ), but not for SPT reactions with *A. fumigatus*- and *C. albicans*-specific serum IgE in other groups. No significant correlations were observed between *T. mentagrophytes*-specific serum IgE and *C. albicans*-specific serum IgE, *T. mentagrophytes*-specific serum IgE and *A. fumigatus*-specific serum IgE, and *A. fumigatus*-specific serum IgE and *C. albicans*-specific serum IgE.

## Discussion

Reaction to a dermatophyte infection may range from mild to severe as a consequence of the host's reactions to the metabolic products of the fungus, the virulence of the infecting strain or species, the anatomic location of the infection and local environmental factors.<sup>17</sup> It is now accepted that a cell-mediated immune response is responsible for the control of infection by dermatophytes. On the other hand, susceptibility to chronic dermatophytosis is associated with atopy and with immediate type hypersensitivity.<sup>9</sup> In this study, we evaluated IH and serum IgE antibody responses in patients with chronic and acute dermatophytosis and healthy subjects.

In our study the highest prevalence (50.9%) of the patients with chronic dermatophytosis was in the

fourth and fifth decades (45-59 years) of life. There are certain factors that influence the distribution of dermatophytes and these include environment, age and sex. Indeed, an increase in infection with increasing age is well established.<sup>18</sup> This observation is consistent with the view that changes in the immune response which occur with advancing age lead to disease susceptibility. In addition, chronic dermatophyte infections and atopic disorders were demonstrated more often in males (58.9% and 56.1%) than in females (41.1% and 43.9%), respectively. Increased dermatophyte infections in men have also been reported in a multicenter survey carried out in North America as well as among atopic and nonatopic subjects in Venezuela.<sup>19,20</sup> Thus, a propensity for men to develop dermatophytosis appears to be a widespread phenomenon.

Tinea unguium (85.3%) was the commonest clinical type in patients with chronic dermatophytosis. *T. mentagrophytes* (73%) caused infection in the five clinical types found in the study. Our results are in agreement with those reported by some other authors, who identified *T. mentagrophytes* as the most frequent dermatophyte.<sup>21-24</sup> The true prevalence of *T. mentagrophytes* infection in Iran may be even higher, because our data were related only to patients who sought medical attention and were referred to a dermatologist. Other anthropophilic dermatophytes (*E. floccosum* and *T. violaceum*) were isolated only rarely, probably because migration of people from other countries to Iran is of minor epidemiological importance. The most common clinical diagnosis in patients with acute infection was tinea corporis (71.4%) caused by *M. canis* (48.6%). The high frequency of tinea corporis observed in our study is mostly due to high prevalence of *M. canis* infection, which shows a predilection for the trunk and extremities. According to our patients' anamnestic data, cats were implicated as the main source of *M. canis* infection. In our previous study<sup>25</sup> *M. canis* was the most frequent dermatophyte isolated from domestic animals, mainly carrier cats, in Iran.

Dermatophyte metabolites are able to penetrate into the skin to induce an immune and eczematous response. *Trichophyton* is the genera most frequently observed and most patients have a self-limited disease that is controlled by cell-mediated immunity.<sup>26</sup> In atopic patients and in those with IgE antibodies to *Trichophyton* antigens, an increased prevalence of chronic dermatophytosis has been suggested.<sup>27</sup> In the present study, 44.2% of patients

with chronic infection showed significantly more positive reactions in SPT to one or more fungal strains than other groups ( $P < 0.05$ ). Atopic patients with chronic dermatophytosis were significantly more susceptible to *T. mentagrophytes* than *C. albicans* and *A. fumigatus* antigens ( $P < 0.05$ ). By contrast, there were no significant differences between atopic patients with acute dermatophytosis and atopic patients according to the fungal isolates. The association of chronic dermatophyte infection with atopy was 95.1%, in contrast to the results of the studies by Rajka and Barlinn,<sup>28</sup> Ward et al.<sup>29</sup> and Escalante et al.<sup>19</sup> who reported 40%, 44% and 49.2%, respectively. A previous survey of fungal sensitivity among 3248 allergic subjects with respiratory symptoms (rhinitis or asthma) showed a 19.1% prevalence of SPT reactivity to some fungal genera, including *Alternaria*, *Aspergillus*, *Candida*, *Cladosporium*, *Penicillium*, *Saccharomyces* and *Trichophyton*. Furthermore, 10.2% of subjects who were SPT positive to at least one fungus exhibited IH to *Trichophyton*.<sup>30</sup> Surprisingly, a much higher rate of positive SPTs was found in atopic patients with chronic infection, whereas the rates of positive SPTs in atopic patients with acute infection (11.6%) and atopic patients (12.2%) were in close agreement with above-mentioned study.<sup>31</sup> Reasons for this increase are not exactly clear, although it may be due to higher atopic population in the chronic infection group than in other groups, different contents of the allergens in various strains tested and the lack of a single diagnostic criterion with a standardized test.

The ELISA method was used to determine the total IgE and specific serum IgE against tested fungi. As expected, we observed that 70.7% of atopic patients with chronic dermatophytosis and 46.3% of atopic patients had significantly higher total IgE values, representing significant differences with other groups ( $P < 0.05$ ). In addition, 30.1% of patients with chronic infection had a positive specific IgE response to at least one fungal isolate. *T. mentagrophytes* (19%) was the most prominent isolate showing positive reactions, followed by *C. albicans* (7.4%) and *A. fumigatus* (3.7%). The frequencies of specific IgE antibodies to *T. mentagrophytes* in other groups were lower than those of specific IgE antibodies in atopic patients with chronic dermatophytosis. In agreement with our study, previous studies also demonstrated that there is a significant statistical correlation between skin tests and ELISA.<sup>19,31</sup> In a study by Alonso et

al.,<sup>32</sup> a high prevalence of elevated IgE antibody levels (31% of subjects) among podiatrists was demonstrated. More recently, Braunstein and Wilson et al.<sup>33</sup> reported about an atopic patient who had tinea unguium, IH and specific IgE antibody to *Trichophyton* and recalcitrant hand and foot eczema. Interestingly, the eczematous lesions resolved after systemic antifungal therapy. They proposed that atopic eczema in this patient was exacerbated by chronic dermatophyte infection and *Trichophyton* hypersensitivity. The same group observed a patient with severe asthma, tinea pedis and cruris, and IH plus positive radioallergosorbent test (RAST) to *Trichophyton* whose asthma improved with antifungal therapy.<sup>31</sup> Our results show a link between chronic dermatophytosis and atopic status, because of an increased prevalence of IH skin test to *T. mentagrophytes* in patients with chronic infection.<sup>34</sup> In addition, although significantly increased levels of specific serum IgE antibodies were present in atopic patients compared with nonatopic subjects, the total IgE levels were comparable among nonatopic subjects with and without dermatophytosis. Although our findings are consistent with previous reports that *Trichophyton* sensitization is common in patients with allergic disorders, differences observed between our results and other investigations are related to a lack of standardized antigenic extracts, the large number of possible antigenic epitopes, the poor sensitivity of the skin immediate hypersensitivity response, the stability of crude antigens, the length of the culture period, variations in the fungal strains, the allergenic extracts used and the different assay systems employed.<sup>35</sup> In conclusion, our study confirmed the role of atopy in IH and specific anti-*T. mentagrophytes* IgE responses in patients with chronic dermatophytosis. In order to evaluate the other predisposing factors to induce IH and to increase IgE, this study should be continued and focused on the exact factors.

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