

Herpes Simplex Type I Infection and Atopy Association in Turkish Children with Asthma and Allergic Rhinitis

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ABSTRACT

This study investigated the association between HSV-1 infection and atopy by comparing seropositivity to HSV-1 in atopic children with asthma and allergic rhinitis and in non-atopic children.

Totally 249 children randomly selected from the university outpatient pediatric clinics were prospectively enrolled in the study between September 1 and November 30, 2007. Serum samples were examined using the virus neutralization test (VNT) for HSV-1 Immunoglobulin G(IgG) seropositivity. Skin prick tests (SPTs) were performed to determine atopic status.

The results showed that HSV-1 IgG seropositivity was significantly higher in atopic children (56.8%) with asthma and allergic rhinitis than in the age-matched non-atopic children group (30.4%) ($p < 0.001$). Although the occurrence of atopy was higher in seropositive girls (57%) than in seropositive boys (47%), the difference was not significant ($p = 0.329$).

These results support a possible relationship between the atopic status of children with asthma and allergic rhinitis and HSV-1 infection.

Key words: Atopy; Asthma; Allergic Rhinitis; Children; Herpesvirus-1

INTRODUCTION

Atopy is the abnormal production of IgE specific to common environmental allergens and is immunologically characterized by dominant T helper (Th) 2 mechanisms.¹

The prevalence of atopic disorders, such as asthma, hay fever and atopic dermatitis, has increased dramatically over the past few decades. The prevalence of allergic diseases is increased especially among children.²⁻⁴ It is generally recognized that atopic diseases are caused by the exposure of genetically predisposed individuals to environmental risk factors. Changes in exposure to environmental factors have been proposed as a tentative explanation for the rise in atopic diseases.^{5,6}

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Although the important inherent component in these diseases is well established, the role of environmental factors in relation to allergic disorders has not been well studied. One of the most important environmental factors is infection. A high load of infections during early childhood may help to prevent allergies.⁷ Thus changes in spectrum of infections might be an important factor contributing to the increased prevalence of allergy.

As most infections during childhood are caused by viruses, interest has mainly focused on viral infection. Immunological data showed that while some infections such as respiratory syncytial virus infections^{8,9} can promote atopy, some others such as measles,¹⁰ hepatitis A¹¹ and tuberculosis¹² inhibit it.

Although HSV-1 is such a viral disease with the potential to affect every cell in the body by different mechanisms¹³ including the immune system cells, there have been few studies assessing the association between HSV-1 and atopy. Herpes viruses may affect the immune system by increasing the Th1 or Th2 immune response, or atopic patients may be more susceptible to herpes viruses. In a previous study, we demonstrated that HSV-1 positivity was high in atopic children.¹⁴

This study aimed to estimate the specific seroprevalence of HSV-1 in atopic children with asthma and allergic rhinitis and in non-atopic children, in order to assess the association between HSV-1 infection and atopy. For that purpose, 249 randomly selected serum samples from atopic and non-atopic children were examined using the virus neutralization test (VNT), the effectiveness of which was confirmed in our previous study.¹⁴

PATIENTS AND METHODS

Patient Population

Children aged 4-17 were randomly selected in a prospective manner from the university-based pediatric allergy and pediatric outpatient clinics of Ondokuz Mayıs University in Turkey between September 1 and November 30, 2007. These departments see approximately 450 patients monthly.

Children who were diagnosed with moderate asthma and allergic rhinitis and at least one IgE positivity against allergens was determined by the prick test panel (routinely used in our allergy clinic) were regarded as atopic. Other children applying to outpatient clinics due to temporary diseases such as upper respiratory tract infections or diarrhea and

without positivity against any of the allergens as determined by the skin prick test were regarded as non-atopic. Initially, socio-economic status was evaluated using the 'Socio-economic Status Index for Turkey',¹⁵ and children of middle socio-economic status were selected. Children with different socioeconomic characteristics were excluded from the study.

Atopic children associated with diagnosed asthma and allergic rhinitis alone were included in the study, atopic children associated with chronic illnesses other than asthma and allergic rhinitis were excluded.

Non-atopic children associated with any diagnosed chronic illnesses, including asthma and allergic rhinitis, were also excluded from the study.

Serum Specimens:

Blood samples were taken and centrifuged at 3000 rpm for 20 minutes at 4°C and a serum specimen obtained. After centrifugation, each serum specimen was poured into another tube (Eppendorf, Germany), heat inactivated at 56 °C for 30 minutes and stored at -20° C until tested.

Virus Neutralization Test

All samples were analyzed using VNT, the effectiveness of which was confirmed in our previous study.¹⁴

Skin Prick Test

Skin Prick Tests (SPTs) were performed in duplicate using a lancet with a 1-mm tip (Mizollen) on the volar surface of the forearm or to the patients' back. Skin testing was performed by the same individual using standardized allergen extracts including house dust mite (*Dermato-phagoides pteronyssinus* and *Dermatophagoides farinae*), mold, grass, tree, weed, feather, dander mix and food allergens (milk, egg and wheat) (Allergo pharma).

Histamine hydrochloride and phenolated glycerol saline were used as positive and negative controls, respectively. Evaluations were performed according to the standards of the European Academy position paper. Skin response was measured 15 minutes later; a positive SPT result was defined as a weal greater than or equal to one half of the diameter of the histamine control and at least 3 mm larger than the diameter of the negative control.¹⁶ Patients took no type of antihistamines for at least 7 days before the prick tests were performed.

A Possible Relationship Between Atopic Status of Children with HSV1 Infection

Table 1. Baseline characteristics of children

	Atopic children(n=88) <i>Number(%) or Mean ± SD</i>	Non atopic children(n=161) <i>Number(%) or Mean ± SD</i>
Age	9.07± 3.98*	8.65±3.26*
Boys	56(63.63)*	98(60.86)*
Middle socia-economical status	88(100)*	161(100)*

*Difference not significant, P >0.05

Statistical Analysis

All statistic analysis were performed using SPSS/Windows Version 16.0 (SPSS Inc, Chicago, IL, USA). A P value of less than 0.05 was considered statistically significant.

Ethical Considerations:

The study was approved by the Ondokuz Mayıs University School of Medicine Committee for Ethics in Medical Research.

RESULTS

Four hundred thirty children were randomly enrolled in the study (150 from the pediatric allergy polyclinic, 280 from the pediatric outpatient polyclinic) 181 children were excluded from the study (62 from the atopic group, 119 from non-atopic group). The

remaining 249 children were duly analyzed (88 from the atopic group in which at least one allergen positivity was shown by skin prick test and diagnosed as allergic rhinitis or asthma or both, 161 from the non-atopic group). Children in non-atopic group were not associated with any chronic illnesses including allergic diseases. Children in atopic group were not associated with any chronic illnesses other than asthma and allergic rhinitis.

The groups were well balanced at the baseline (Table 1). When we compared the age distribution of the groups in terms of HSV IgG positivity percentages we determined a normal distribution (One-Sample Kolmogorov-Smirnov Test, P=0.809). Similarly, when we compared the age distribution of the two groups in respect of atopy positivity percentages distribution was also normal (One-Sample Kolmogorov-Smirnov Test, P=0.950).

Table 2. Seroprevalences of HSV-1, between atopic and non-atopic children groups

Study Group (n=249)						
Group 1:		Group 2:		Odds ratio	95% confidence interval	P value
Atopic Children (n=88)		Non-atopic Children (n=161)				
HSV-1 IgG		HSV-1 IgG				
(+)	(-)	(+)	(-)	3.008	1.754-5.156	<0.001
(n=49)	(n=39)	(n=50)	(n=111)			
56.8%	33.2 %	30.4%	69.6%			

Table 3. Occurance of atopy between HSV-1 IgG seropositive boys and girls

Study group, HSV-1 IgG seropositive children (n=99)						
Girls (n=35)		Boys (n=64)		Odds ratio	95% confidence interval	P value
Atopic Children n=20 57%	Non-atopic Children n=15 43%	Atopic Children n=30 47%	Non-atopic Children n=34 57%			
				1.5111	0.659-3.466	P=0.329

In the atopic children group, seropositivity was significantly higher (3.008 times) than non-atopic group. The HSV-1 seroprevalence of the two groups is presented in Table 2, ($P < 0.05$).

Ninety-nine of the 249 (40%) children were HSV-1 Ig G-seropositive (Table 3). Atopy was determined in 20 (57%) seropositive girls ($n=35$) and in 30 (47%) seropositive boys ($n=64$). Although the difference was higher in females (1.511 times) in seropositive atopic children, gender distribution was not significantly different ($p=0.329$, CI: 0.659) (Table 3).

DISCUSSION

We determined a relationship between HSV and allergy. Hence how might HSV infection influence the prevalence of allergy? To answer this question, we must first elucidate the pathogenic mechanisms that are believed to underlie the allergic diseases.

The main pathology in allergic diseases is atopy development. Atopy is characteristically associated with an imbalance between various types of T cells and an increase synthesis of IgE. Early studies revealed a T-cell imbalance that was found to be present in all the important types of childhood atopic diseases, such as bronchial asthma and hay fever, as well as atopic dermatitis.¹⁷

Infections, which may cause a change in balance of cytokines, may lead to atopy development. Only few studies have investigated the correlation between HSV and allergic diseases, and the results of those studies are inconsistent. Some reported a protective effect of HSV against allergic diseases, while others suggested the opposite.

Multicentre Allergy Study (MAS) Group¹⁸ members have shown that in German children repeated upper respiratory tract infection and non-respiratory viruses' infection like herpes simplex in infancy may have protective effect against development of asthma until the school age period.

Prabripataloong et al.¹⁹ suggested that atopy is a risk factor for herpes simplex ocular disease and that this risk is increased if a patient has severe atopy.

Brandt et al.²⁰ organised another multicenter case control study for examining the association between atopy and HSV keratitis, and they could not find a significant statistical difference between the two groups. Rezende et al.²¹ compared the characteristics of ocular HSV in patients with and without atopy and

showed that atopic patients had considerably more infections and fewer inflammatory episodes compared with atopics.

In our previous study⁽¹⁴⁾ we determined high HSV-1 IgG seropositivity among atopic Turkish children without, however, comparing them with non-atopic children.

Although various studies^{14, 18, 19, 22-25} seem to differ in their accounts of the relationship between HSV-1 and atopy, one common point on which they all agree, and with which our results are also compatible, is that there is an association between HSV-1 infection and atopy.

This raises the question of the kind of relationship between HSV type 1 infection and atopy. Two simple suggestions can be made.

The first is the long-standing clinical view that atopic disease increases the risk of HSV infection.²²⁻²⁴ Because atopic individuals react to antigens primarily through Th2 responses and because an effective Th1 response is needed to provide an effective immune response to HSV infection,^{22,26,27} atopic patients, especially those with severe atopy, may be more susceptible to HSV infection.

The second is that HSV-1 infection itself may be a causative factor in atopy development. Dendritic cells (DC), as antigen-presenting cells located at the border zones of the body with the environment, have been shown to play a crucial role as one of the first cells, beside epithelial cells, interacting with HSV on the one hand, and as important controllers of viral spread on the other. HSV-infected DC secretes lower levels of IL-12 in consequence of stimulation by lipopolysaccharides (LPS) *in vitro*.²⁸ IL-12 is required for the polarization of naive T cells into Th1 helper cells, which are characterized by high interferon (IFN)- γ secretion. Furthermore, IL-12 is produced during the maturation of DC together with a weaker stimulatory capacity toward T cells,²⁹ the amount of IL-12 production shows the maturation stage of HSV-infected DC and the capacity to induce substantial quantities of IFN producing Th1 T cells. A reduced ability to produce IFN-gamma by dendritic cells during HSV infection may therefore be closely correlated with an increased tendency to develop allergic sensitization and atopic diseases later in life. The second explanation makes it possible to theorize that HSV-1 may cause atopy development, and that the prevalence of this virus may be higher in atopic children.

A Possible Relationship Between Atopic Status of Children with HSV1 Infection

There is no doubt that preventing atopic diseases will always be much preferable to curing them, whatever treatment choices exist. Establishing the kind of relationship existing between atopy and herpes simplex virus infection may be an important step in the development of primary preventive measures.

In our study HSV-1 IgG seropositivity was significantly much higher in atopic children with asthma and allergic rhinitis than in non-atopic children. HSV-1 IgG seropositivity was similar in both genders for both groups.

In an evaluation of the HSV status of a population, age and socioeconomic status are particularly important. HSV antibodies are acquired by 20-50% of children by the age of 5 and by 90% of children by age of 10 years, depending on socioeconomic conditions. When we compared the age distribution of the two groups, they were similar and normal; no difference was determined between the groups, and all the children selected were of middle socio-economic status. Any difference in age or socioeconomic status between the atopic and non-atopic groups that might have caused the difference in seropositivity for HSV-1 rather than atopic status was thus excluded.

Our study is the first seroprevalence investigation of HSV-1 IgG antibodies in atopic children with asthma and allergic rhinitis and the first to compare these with seroprevalence in non-atopic children. Previous studies have evaluated the relationship between HSV and allergic diseases without excluding possible bias factors which might lead to inaccurate results, such as patients' socioeconomic status or age group distribution, whereas our study is interesting because such possible influences were excluded. Although our results support the existence of an association between HSV Type I infection and atopy, the present data do not allow us to explain the kind of relationship. HSV is either the cause or the result of atopy development; further detailed, prospective, community-based studies are now needed in order to clarify the link between cause and effect.

REFERENCES

1. Del Prete G. Human Th1 and Th2 lymphocytes: their role in the pathophysiology of atopy. *Allergy* 1992; 47(5):450-5.
2. Mannino DM, Homa DM, Pertowski CA, Ashizawa A, Nixon LL, Johnson CA, et al. Surveillance for asthma--United States, 1960-1995. *MMWR CDC Surveill Summ* 1998; 47(1):1-27.
3. Upton MN, McConnachie A, McSharry C, Hart CL, Smith GD, Gillis CR, et al. Intergenerational 20 year trends in the prevalence of asthma and hay fever surveys of parents and offspring. *BMJ* 2000; 321(7253):88-92.
4. Matricardi PM. Infections preventing atopy: facts and new questions. *Allergy* 1997; 52(9):879-82.
5. Matricardi PM, Rosmini F, Panetta V, Ferrigno L, Bonini S. Hay fever and asthma in relation to markers of infection in the United States. *J Allergy Clin Immunol* 2002; 110(3):381-7.
6. Serafini U. Do infections protect against asthma and atopy? *Allergy* 1997; 52(9):955-7.
7. Schaub B, Lauener R, von Mutius E. The many faces of the hygiene hypothesis. *J Allergy Clin Immunol* 2006 ; 117(5):969-77.
8. Ichinohe S, Chida S, Inoue H. Viral infection and asthma: respiratory syncytial virus and wheezing illness. *Allergology Int* 1999; 48(2):93-101.
9. Sigurs N, Bjarnason R, Sigurbergsson F, Kjellman B. Respiratory syncytial virus bronchiolitis in infancy is an important risk factor for asthma and allergy at age 7. *Am J Care Med* 2000; 161(5):1501-7.
10. Shaheen SO, Aaby P, Hall AJ, Barker DJ, Heyes CB, Shiell AW, et al. Measles and atopy Guinea-Bissau. *Lancet* 1996; 347(9018):1792-96.
11. Matricardi PM, Rosmini F, Ferrigno L, Nisini R, Rapisetta M, Chionne P, et al. Cross sectional retrospective study of prevalence of atopy among Italian military students with antibodies against hepatitis A virus. *BMJ* 1997; 314(7086):999-1003.
12. Shirakawa T, Enomoto T, Shimazu S, Hopkin JM. The inverse association between tuberculin responses and atopic disorder. *Science* 1997; 275(5296):77-9.
13. Favoreel HW, Nauwynck HJ, Pensaert MB. Immunological hiding of herpesvirus-infected cells. *Arch Virol* 2000; 145(7):1269-90.
14. Artiran Igde F, Igde M, Yazici Z, Okur Gumusova S, Birinci A, Sancak R, et al. Distribution of the HSV-1 IgG antibodies by two methods comparing in Turkish Atopic Children. *New Microbiologica* 2007; 30(2):109-12.
15. Kalaycioglu S, Kardam F, Tuzun S, Ulusoy M. Approaches and tries with regard to develop a socioeconomic status index for Turkey. *Commun Physician (Toplum ve Hekim)* 1998; 13:126-37.
16. Position paper: Allergen standardization and skin tests. The European Academy of Allergology and Clinical Immunology. *Allergy* 1993; 48(14 Suppl): 48-82.
17. Strannegard O, Strannegard IL. T lymphocyte numbers and function in human IgE-mediated allergy. *Immunol Rev* 1978; 41:148-70.
18. Illi S, von Mutius E, Lau S, Bergmann R, Niggemann B, Sommerfeld C, et al. Early childhood infectious diseases

- and the development of asthma up to school age: a birth cohort study. *BMJ* 2001; 322(7283):390-5.
19. Prabritputaloong T, Margolis TP, Lietman TM, Wong IG, Mather R, Gritz DC. Atopic disease and herpes simplex eye disease: A population-based case-control study. *Am J Ophthalmol* 2006; 142(5):745-9.
 20. Brandt BM, Mandleblatt J, Asbell PA. Risk factors for herpes simplex-induced keratitis: a case-control study. *Ann Ophthalmol* 1994; 26:12-6.
 21. Rezende RA, Hammersmith K, Bisol T, Lima AL, Webster GF, Freitas JF, et al. Comparative study of ocular herpes simplex virus in patients with and without self-reported atopy. *Am J Ophthalmol* 2006; 141(6):1120-5.
 22. Wollenberg A, Wetzel S, Burgdorf WH, Haas J. Viral infections in atopic dermatitis: pathogenic aspects and clinical management. *J Allergy Clin Immunol* 2003; 112(4):667-74.
 23. Bruijnzeel-Koomen C, Madde G, Kapp A. Atopic dermatitis. In Kay A, editor. *Allergy and allergic disease*. Oxford: Blackwell science 1997:1573-85.
 24. David TJ, Longson M. Herpes simplex infections in atopic eczema. *Arch Dis Child* 1985; 60(4): 338-43.
 25. Wahn U, Bos JD, Goodfield M, Caputo R, Papp K, Manjra A, et al. Efficacy and safety of pimecrolimus cream in the long-term management of atopic dermatitis in children. *Pediatrics* 2002; 110(1 Pt 1):e2.
 26. Carr DJ, Harle P, Gebhardt BM. The immune response to ocular herpes simplex virus type 1 infection. *Exp Biol Med* 2001; 226(5):353-66.
 27. Pepose J, Leib DA, Stuart M. Herpes Simplex virus diseases: anterior segment of the eye. In: Pepose JS, Holland GN, Wilhelmus KR, editors. *Ocular infection and immunity*. St Louis, Missouri: Mosby-Year Book, 1996:905-932.
 28. Mikloska Z, Bosnjak L, Cunningham AL. Immature monocytederived dendritic cells are productively infected with herpes simplex virus type 1. *J Virol* 2001; 75(13):5958-64.
 29. Pollara G, Speidel K, Samady L, Rajpopat M, McGrath Y, Ledermann J, et al. Herpes simplex virus infection of dendritic cells: balance among activation, inhibition, and immunity. *J Infect Dis* 2003; 187(2):165-78.