

Does hyperimmunoglobulinemia-E protect tropical populations from allergic disease?

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The Waorani Indians of eastern Ecuador have the highest blood concentration of IgE reported in a human population. Evidence obtained by medical history, physical examination, and immediate hypersensitivity skin tests suggests that pollen allergy and other atopic diseases are rare among the Waorani. A similar association between parasite induced hyperimmunoglobulinemia-E and a low prevalence of conventional atopic disease has been reported in numerous other tropical populations. Saturation of mast cell IgE receptors with antibodies directed to the parasite and/or other antigens and competitive inhibition of passive binding of pollen allergen-specific IgE is one hypothetical cause of this association. We have tested this interesting conjecture by passively sensitizing the skin of Waorani Indians with serum containing pollen allergen-specific IgE antibodies. Waorani Indians with hyperimmunoglobulinemia-E can be adoptively sensitized with human ragweed or rye grass hyperimmune IgE antisera. This suggests that the cutaneous mast cells of healthy Waorani have active IgE receptors. The high circulating plasma concentrations of IgE in the Waorani do not prevent adoptive cutaneous sensitization with pollen-specific IgE antibodies. (J ALLERGY CLIN IMMUNOL 71:184, 1983.)

IgE is present in increased concentration in sera from patients with allergic rhinitis, bronchial asthma, atopic dermatitis, and parasitic infections.¹ This last association is of special interest because of the possible role of IgE in protection against parasitic disease.² Several investigators have commented on the low prevalence of allergy in primitive populations and suggested that the high serum concentrations of IgE induced by parasitism may prevent allergic disease.³⁻⁵ Three hypothetical mechanisms could contribute to the low prevalence of atopic disease in primitive tropical populations. First, the generation of large quantities of anti-parasite IgE could preclude production of IgE antibodies to the pollen allergens

responsible for atopic disease. Second, the high serum IgE concentrations could be associated with (1) a decreased number of tissue mast cells, or (2) down regulation of the activity and/or number of mast cell IgE receptors. Finally, the high serum IgE levels could competitively displace pollen allergen IgE antibodies from tissue mast cell IgE receptors. An opportunity to test the possible importance of this last mechanism arose during studies of an isolated South American Indian population in which parasitic infection is common and atopic disease is rare.⁶⁻⁸

METHODS

The Waorani Indians inhabit the rain forest at the head waters of the Amazon river in Ecuador. A general description of the Waorani and their health has been presented in previous reports.⁶⁻⁸ The Waorani have the highest plasma concentrations of IgE reported in a normal human population.⁹ By means of a radial immunodiffusion method for IgE measurements, the mean plasma IgE concentration in 227 subjects was 11,975 IU/ml (normal 5 to 500 IU/ml). The reason for the exceptional plasma IgE concentrations in this population is not known. Among 293 individuals examined, four (1.3%) had symptoms suggestive of allergic disease.⁶⁻⁸ Two subjects complained of wheezing respiration, one had a history of contact dermatitis, and the fourth reported symptoms suggestive of allergic rhinitis and conjunctivitis.

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Examination of these subjects revealed no evidence of bronchial constriction or dermatitis.

Intestinal parasites were prevalent in the Waorani. Examination of single stool specimens in 65 of the subjects studied⁹ revealed a 51% frequency of *Entamoeba coli* cysts, a 46% frequency of hook worm eggs, a 28% frequency of *Giardia lamblia* cysts, and a 20% frequency of *E. histolytica* cysts. The frequency of *Ascaris*, *Enterobius*, and *Trichuris* eggs was less than 3%. Multiple parasites were detected in over half the subjects sampled; 23% had no evidence of parasites on a single stool examination.

The studies presented in this article were accomplished in conjunction with medical and dental clinics carried out among the Waorani during July 1981. Skin tests were accomplished in five Waorani subjects and three expedition member controls. Each expedition member and Waorani volunteer was questioned about allergic and vasomotor nasal symptoms and about asthma and dermatitis. All subjects were tested by an epicutaneous bibarb prick needle method¹⁰ with 1/20 w/v allergenic extracts from short ragweed (*Ambrosia elatior*) and Italian rye grass (*Lolium multiflorum*) pollens prepared by Greer Laboratories, Lenoir, N. C. Control skin tests were accomplished with 60 mM/L histamine and saline. The longest and the bisecting perpendicular diameters of the wheal and flare reactions were measured after 30 min. The measured reactions were averaged to the nearest millimeter and recorded. On the opposite forearm, two 0.1 ml aliquots of neat hepatitis B antigen-negative human serum containing high titers of IgE antibodies to either ragweed or rye grass allergens were administered intracutaneously. Although quantitatively comparable measures of specific antibody concentration, Prausnitz-Küstner titers, and IgE content were not available for these sera, each serum had been previously demonstrated to have high titers of allergen-specific IgE antibodies. For example, the donor of the antiserum to rye grass allergen had a direct intradermal skin test endpoint titer of 10^{-12} and this measure of specific antibody activity exhibited a high degree of correlation ($r > 0.90$) with two different in vitro measures of specific serum antibody activity.¹¹ Twenty-four hours after passive sensitization, the sites were challenged by epicutaneous tests with the allergens. Crossed site testing with each allergen was accomplished as a specificity control.

RESULTS

Each subject was initially prick tested with ragweed, rye grass, and appropriate controls prior to passive cutaneous sensitization with sera containing IgE antibodies to the test allergens. Table I summarizes the skin-test responses detected in expedition members and in the Waorani Indian volunteers. The saline control responses in all subjects were ≤ 2 mm. Expedition member controls and Waorani exhibited the expected wheal and flare response to tests with 60 mM/L of histamine. Differences in histamine reactivity between the controls and volunteers were small and not significant. Although the Waorani exhibited a

TABLE I. Immediate hypersensitivity reactions before passive cutaneous sensitization

	Flare (F) or wheal (W) response (mm)*					
	Ragweed antigen		Rye grass antigen		Histamine 60 mM/L	
	F	W	F	W	F	W
Controls						
C. M.	0	0	3	0	7	5
H. T.	0	0	1	0	21	7
J. L.	0	0	2	0	20	5
Mean	0	0	2.9	0	16.0	5.6
±SEM			0.6		4.5	0.6
Waorani						
2204	2	0	3	0	10	5
2203	2	0	2	0	10	5
2226	0	0	2	0	12	5
2207	3	0	3	0	12	8
2227	0	0	3	0	20	9
Mean	1.5	0	2.5	0	12.8	6.2
±SEM	0.5		0.2		1.8	0.7
Controls vs Waorani (p value)	<0.01	ns	ns	ns	ns	ns

ns = not significant.

*Saline control flare responses were ≤ 2 mm in all subjects.

small but statistically significant (Table I) increase in flare reactivity to ragweed allergen, the magnitude of this difference was small in comparison to the reactivity to the histamine control. These initial tests provided evidence that the skin of the Waorani and expedition member controls could react to histamine and that pollen allergens did not provoke large skin-test reactions in either subject group.

Table II summarizes the serum IgE concentrations and the results of skin tests with ragweed and rye grass allergens at passively sensitized skin-test sites in the same subjects. The geometric mean serum IgE level of the Waorani was 62-fold greater ($p < 0.0006$) than the serum IgE level of the expedition member controls. Although the average magnitude of cutaneous reactivity differed among the compared groups of subjects, all passively sensitized sites exhibited large reactions to prick tests with each homologous allergen. Sites tested with the nonhomologous allergen exhibited reactivity equivalent to or slightly greater than the saline control and/or pollen allergen reactions previously detected at unsensitized sites. These observations suggest that specific passive cutaneous sensitization to pollen allergens could be accomplished in the hyperimmunoglobulinemic-E Waorani Indians.

TABLE II. Immediate hypersensitivity reactions after passive cutaneous sensitization

	Serum IgE (IU/ml)	IgE antiserum							
		Ragweed				Rye grass			
		Test antigen				Test antigen			
		Ragweed		Rye grass		Ragweed		Rye grass	
		F*	W*	F	W	F	W	F	W
Controls									
C. M.	22	12	6	3	0	0	0	21	10
H. T.	96	23	12	0	0	0	0	25	12
J. L.	140	25	10	3	0	0	0	24	10
Geometric mean	65								
Mean	4.1993	20.0	9.3	2.0	0	0	0	23.3	10.7
±SEM	0.6651	4.1	1.7	1.0				1.2	0.6
Waorani									
2204	290	8	5	3	0	2	0	22	10
2203	4100	10	5	0	0	0	0	15	7
2226	6000	5	2	4	0	3	2	12	7
2207	14020	4	2	3	0	4	2	16	5
2227	15480	8	5	3	0	3	2	10	7
Geometric mean	4344								
Mean	8.3767	7.0	3.8	2.6	0	2.4	1.2	13.8	7.6
±SEM	0.7218	1.1	0.7	0.7		0.7	0.2	2.2	0.6
Controls vs Waorani (p value)	≤0.0006	≤0.001	≤0.001	ns	ns	≤0.02	≤0.02	≤0.02	≤0.02

ns = not significant.

*Flare (F) or wheal (W) response expressed in millimeters; saline control flare responses were ≤2 mm in all subjects.

The grouped mean $\pm$ SEM of the wheal and flare reactivity of expedition member controls and Waorani volunteers are compared in Table II. The wheal and flare reactions detected with the homologous allergens were significantly smaller for ragweed and rye grass in the Waorani (Table II). This suggests that the skin of the Waorani was less reactive when tested with the same quantity of passively transferred IgE antibody and homologous allergens used in expedition member controls. No significant differences were detected in the reactivity of controls and Waorani tested with rye grass at ragweed-sensitized sites. In contrast, the Waorani were again slightly more reactive than controls (Table II) when tested with ragweed at rye grass-sensitized skin-test sites. This finding is similar to the small but significant trend detected previously with ragweed at unsensitized skin-test sites. This trend suggests prior exposure of the Waorani to an allergen similar to ragweed.

DISCUSSION

Much evidence suggests that familial,¹² racial,^{13, 14} and environmental influences contribute to serum IgE concentrations. Increased serum concentrations of

IgE can be produced in experimental animals and in man by infections with helminth parasites.¹⁻² Parasite infestation or infection probably constitutes the most significant environmental stimulus to the increased IgE concentrations detected in the Waorani. Single stool examinations in the Waorani revealed a high prevalence of intestinal parasites and almost a 50% frequency of helminth eggs.⁶⁻⁹ This prevalence of helminths would be expected to be much higher had opportunity existed for examination of more than a single stool specimen. The Waorani are an isolated population, and inbreeding could contribute to the relative invariance of high serum IgE concentrations observed among these people. High serum IgE concentrations have been reported in other tropical populations.^{4, 5, 13, 14} Like the Waorani, many of these populations exhibit a relatively low incidence of atopic diseases.^{4, 5, 13} These considerations and the extreme serum concentrations of IgE in the Waorani provided an ideal opportunity to test the relationship between circulating concentrations of IgE and the capacity to elicit cutaneous reactivity to pollen allergens commonly associated with immediate hypersensitivity.

Helminth parasites provoke increased production of

IgE in two ways. Infection stimulates the production of specific anti-parasite IgE. Helminth infection also causes polyclonal stimulation of IgE-producing B cells and the generation of a large excess of IgE that is not specific for the antigens of the parasite. This last mechanism possibly accounts for a major portion of the IgE in helminth-infected subjects.¹⁶ Specific IgE antibodies mediate allergic reactions by binding to mast cell membrane receptors. The subsequent binding of antigen to two or more cell-fixed IgE antibodies initiates mast cell membrane changes, leading to the release of histamine and other chemical permeases responsible for allergic disease.¹⁷ The binding of IgE to mast cells and basophils is a reversible process in which specific and nonspecific serum IgE is in equilibrium with IgE bound to highly avid mast cell surface receptors. As much as a 10,000-fold excess of IgE antibodies may be in equilibrium with 100,000 receptors on each cell surface.¹⁸⁻²⁰

Mast cells can be passively sensitized *in vivo* and *in vitro* by specific IgE, and passive sensitization can be inhibited by prior saturation of available IgE receptors with "nonspecific" IgE.²¹ Inhibition of passive sensitization with IgE antibodies has been reported in man and animals bearing IgE producing myelomas.²² These observations and the mechanisms by which IgE antibodies initiate allergic reactions predict the possible refractoriness of helminth-infected individuals to passive sensitization with allergen-specific IgE antibodies. This conjecture has stimulated several important interrelated ideas. First, the recruited polyclonal production of large amounts of nonspecific IgE might preclude production of pollen allergen-specific IgE. Second, the possibility exists that (1) the number of tissue mast cells might be decreased, or (2) the number and/or activity of IgE mast cell receptors might be down regulated by passive administration or active stimulation of the production of a large amount of nonspecific IgE. Finally, a high serum concentration of parasite-induced IgE might protect against the occurrence of allergic disease by competitive saturation of available mast cell IgE receptors. The informative observations presented in this article provide a direct test of the relative importance of this last hypothesis.

The skin-test reactivity detected with histamine (Table I) provides direct evidence that the expedition member controls and Waorani volunteers were able to exhibit typical wheal and flare responses to a chemical permease. This suggests that naturally occurring exposure to antihistamine-like compounds does not account for the lack of allergic disease in the Waorani. Neither the Waorani nor the control subjects exhibited innate pollen allergen reactivity that differed greatly from the saline control. The magnitude of naturally

occurring cutaneous reactivity usually associated with ragweed- and rye grass-related allergic disease was not detected in the Waorani or in the controls. The possibility of a low level of increased reactivity to ragweed in the Waorani could not be excluded.

Passive sensitization of skin-test sites in the Waorani and control subjects resulted in a typical wheal and flare response after skin tests with the homologous allergen. This suggests that specific passive sensitization is possible in the Waorani despite an average 62-fold higher concentration of circulating IgE. This observation permits several important deductions. First, the skin of the Waorani contain mast cells capable of releasing sufficient permease to cause wheal and flare reactivity. Second, the mast cell receptors of the Waorani are accessible to the quantity of allergen-specific IgE in small aliquots of passively transferred serum from individuals with symptomatic pollen allergy. Competitive inhibition of binding to mast cell IgE receptors by the high circulating concentrations of IgE did not prevent the cutaneous reaction to the pollen allergens. This observation in man differs from the observations of Jarret et al.,²³ who observed that the "nonspecific" IgE induced by infecting rats with *Nippostrongylus brasiliensis* could block passive sensitization but not reactivity produced by actively synthesized IgE antibodies. The high concentration of "nonspecific" IgE antibody produced by the Waorani failed to inhibit specific passive cutaneous sensitization. This suggests that competitive inhibition of binding to mast cell IgE receptors by circulating nonspecific IgE is not a satisfactory explanation for the low prevalence of allergic disease in the Waorani. Several studies of the degranulation of sensitized leukocytes from allergic humans suggest that only a few allergen-induced bridges between cell-bound IgE antibodies are needed to cause histamine release.^{20, 24} The passively transferred serum provided sufficient antibody to initiate histamine release. These observations suggest that the failure to produce quantities of allergen-specific antibodies equivalent to those in patients with pollen allergy is a more likely cause of the lack of pollen allergen reactivity in the Waorani.

It is important to note that the magnitude of the passively induced allergen reactivity of the Waorani was much less than the reactivity in controls. There are three possible explanations for this observation. The Waorani could have fewer tissue mast cells at the site of allergen challenge. This seems unlikely in view of reports of increased tissue mast cells in inflamed tissues²⁵ and in patients with parasitic infestation.^{26, 27} Alternately, permease released by each mast cell could be much less than the permease released by the cells of the expedition member controls. This obser-

vation is consistent with recent evidence of a lack of cutaneous responsiveness to a cholinergic agonist in the Waorani.⁸ Finally, this effect could be a consequence of competitive inhibition of the amount of specific IgE binding by circulating nonspecific IgE. These studies with neat aliquots of high-titered serum were designed to evaluate responsiveness as opposed to an endpoint titration of sensitivity¹⁰ to passively transferred specific antibodies. Competitive inhibition by the high circulating concentrations of IgE could contribute to the 62% and 35% average reduction in responsiveness to the ragweed and rye grass allergens, respectively. This same mechanism is also suggested by the subtle inverse relationship between the magnitude of the passively transferred skin-test reactions and the circulating IgE concentrations in the five Waorani subjects (Table II). This last mechanism is also consistent with the observations of Sydbom et al.,²⁸ who observed that high levels of nonspecific IgE attenuated both specific IgE-mediated and nonspecific compound 48/80-induced histamine reactivity.

In summary, we have shown that a primitive tropical population with extremely high serum IgE concentrations and a low prevalence of allergic disease can be passively sensitized with specific IgE antibodies to pollen allergens. Even though the magnitude of the responsiveness in passively sensitized experimental subjects was less than that in appropriate controls, large skin-test reactions could be produced. These findings suggest that the decreased prevalence of allergic disease in parasite-infested primitive populations with high serum IgE concentrations is caused by the failure to produce allergen-specific IgE antibodies.

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