

Fire ants represent an important risk for anaphylaxis among residents of an endemic region

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Background: Imported fire ants (IFA) represent a potential anaphylactic risk to IFA-sensitized individuals.

Objective: We examined the prevalence of allergic sensitization to IFA, yellow jacket venom (YJV), and peanut in an adult population from an IFA-infested region, Augusta, Georgia.

Methods: Specific IgE to IFA, YJV, and peanut were determined by using the Pharmacia UniCAP assay in 200 random blood donors from an Augusta blood bank. These results were compared with specific IgE to identical allergens in a random sample of blood donors from Oklahoma City, Oklahoma (OKC), a nonendemic region for IFA.

Results: Prevalence of IFA-specific IgE (17%) in the Augusta population was significantly higher than to YJV (10%, $P = .04$) or peanut (7.5%, $P = .004$). The majority of individuals who had significant IgE to IFA (≥ 0.35 kIU/L) did not have IgE to YJV (24/34 = 71%). YJV caused significantly more inhibition of IgE binding to a YJV solid phase than to an IFA solid phase when the 10 dual-positive sera were analyzed (58% vs 11%, $P = .005$). The prevalence of IFA-specific IgE in adults from Augusta was higher than in OKC (17% vs 2%, $P = .0002$). YJV-specific IgE was also more prevalent in Augusta compared with OKC (10% vs 6.0%, $P = .04$), whereas no difference was demonstrated for peanut-specific IgE (7.5% vs 6.5%, $P = .6$).

Conclusions: Allergic specific IgE to IFA is 1.7 times more common in adults living in an endemic area than specific IgE for other allergens associated with potentially fatal anaphylaxis. This suggests that IFA may pose the greatest risk of anaphylaxis for adults residing in IFA-endemic regions. (J Allergy Clin Immunol 2003;111:1274-7.)

Key words: Fire ant hypersensitivity, immunoglobulin E, anaphylaxis

The imported fire ant (IFA), *Solenopsis invicta*, represents an ongoing and escalating problem in endemic regions. Fire ants, first introduced to Mobile, Alabama, in the late 1930s, have rapidly migrated and now occupy approximately 310 million acres in at least 12 states.¹ Their current territory covers much of the South Atlantic seaboard from North Carolina to Florida,

Abbreviations used

IFA: Imported fire ant
OKC: Oklahoma City, Oklahoma
WBE: Whole-body extract
YJV: Yellow jacket venom

extends throughout the southern United States and across Texas, and stretches into portions of New Mexico, Arizona, and California. The US Department of Agriculture (USDA) estimates that they may eventually occupy 25% of the country.¹

Individuals living in IFA-endemic regions have a 30% to 60% chance of being stung each year.^{2,3} Anaphylactic reactions have been reported from fire ant stings in anywhere from 0.6% to 16% of those stung.⁴⁻⁶ More than 80 deaths have been attributed to fire ant anaphylaxis in the United States.^{7,8} The actual number of deaths is most likely higher because of underreporting of fire ant stings.

The purpose of this study was to define the prevalence of allergic sensitivity to IFAs among adults living in an area where IFAs are endemic. The prevalence of sensitization to IFA was compared with both yellow jacket venom (YJV) and peanut, which are associated with fatal anaphylaxis in adults. Given the frequency of IFA stings in endemic areas, it was hypothesized that sensitization to IFAs would be more common than to the other allergens. To validate the specificity of the in vitro assay used to elucidate allergic sensitization to IFA, an adult population from a region of the country where IFAs are not yet endemic was studied.

METHODS

Study design

This study was a blinded survey of 200 random blood donors from a blood bank in Augusta, Georgia, a known endemic region for *S. invicta*. All samples tested negative for infectious agents. Information about individual donors was limited to age and sex. Permission for this study was granted by the Medical College of Georgia Human Assurance Committee.

Serum samples were stored at -20°C until testing. Allergen-specific IgE antibodies were determined by using the Pharmacia UniCAP (Pharmacia Upjohn, Kalamazoo, Mich) per manufacturer's protocol. Allergens studied were IFA whole-body extract (WBE) (Pharmacia identification code i70), YJV (Pharmacia identification code i3), and peanut (Pharmacia identification code f13). Values of ≥ 0.35 kIU/L were considered positive. Age and sex were compared in all groups studied.

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The results from donors in Augusta were compared with a random sample of 1000 Oklahoma City, Oklahoma (OKC), donors who were also analyzed for specific IgE to YJV and peanut by use of the Pharmacia UniCAP assay. A random subset of 100 OKC samples was further tested for specific IgE to IFA. Although IFAs have migrated significantly since their arrival, according to the USDA and entomologists at Oklahoma State University, the OKC region is not considered to be endemic. These samples served as negative controls for the IFA assay.

Ten sera positive for IgE to both IFA and YJV were selected for inhibition studies. The concentration of IgE to IFA ranged from 0.36 kIU/L to 519 kIU/L, whereas the concentration of IgE to YJV ranged from 0.35 kIU/L to 17.9 kIU/L. Three hundred microliters of each dual-positive serum was combined with 15 μ L of YJV at 550 μ g venom protein/mL. As a control, 15 μ L of albumin-saline was added to a second 300 μ L of each serum. Each diluted serum sample was tested for IFA-specific and YJV-specific IgE with the respective solid-phase UniCAPS. Because of the limited amount of blood donor serum, a serial dilution assay was done with pooled serum positive to IFA and YJV from the Augusta donors. An inhibition assay was performed with serial dilutions of YJV in the fluid phase, and an acceptable inhibition curve was generated.

The specificity of YJV as an inhibitor was further analyzed by using existing serum from 3 donors who did not make substantial IgE (<0.35 kIU/L) to IFA or YJV. One donor had positive IgE ($\geq$ 0.35 kIU/L) to common ragweed (Pharmacia identification code w1), the second donor had positive IgE to Timothy grass (Pharmacia identification code g6), and the third donor had positive IgE to dust mite (*D pteronyssius*, Pharmacia identification code d1). Serum from each donor was mixed with albumin saline and with YJV and reanalyzed with their respective solid-phase allergen by means of the Pharmacia UniCAP system. The addition of YJV did not significantly alter the concentration of IgE to their respective allergens. Finally, sera from 10 Augusta donors who had negative reactions to both IFA and YJV were pooled and retested for IFA and YJV after the addition of albumin saline and concentrated YJV. The pooled negative sera remained negative to both IFA and YJV (IgE <0.35 kIU/L) after the addition of YJV and equal amounts of albumin saline to act as a control. For quality control purposes, we retested 10 IFA-positive samples from the Augusta population to IFA to determine a coefficient of variation for the UniCAP-IFA assay.

Data analysis

The prevalence of IgE antibodies to each of the specific allergens was compared with the others by using the χ^2 test.

Comparisons of the percentage of inhibition of specific IgE to IFA and to YJV among the dual-positive sera were analyzed for significance by using the paired Student *t* test.

RESULTS

Table I provides the demographics of the three different study groups. Average age and the sex of the donors were similar across all population groups. No statistical difference was found between the number of men and women among the three groups (51% vs 49%, $P = .54$). When comparing the mean ages among the three groups, a difference was noted between the Augusta donors and the OKC donors (47 years vs 43 years, $P = .0002$) but not between the Augusta donors and the OKC subset (47 years vs 44 years, $P = .07$). Furthermore, no difference was found between the subset of OKC donors and the entire group of OKC donors (44 years vs 43 years, $P = .55$).

Fig 1 shows that 34 (17%) of the 200 Augusta samples had detectable IFA-specific IgE, 20 (10%) had detectable YJV-specific IgE, and 15 (7.5%) had detectable IgE to peanut. In the OKC sample, 60 (6.0%) of 1000 individuals had detectable IgE to YJV, and 65 (6.5%) of 1000 individuals had detectable IgE to peanut. Analysis of the OKC subset showed that 2 (2%) of 100 individuals had detectable IgE to IFA.

There was a significant difference between the prevalence of specific IgE to IFA compared with the prevalence of specific IgE to YJV ($P = .04$) and compared with peanut ($P = .004$) in donors from Augusta. No difference was shown between prevalence of IgE to YJV compared with prevalence of IgE to peanut ($P = .38$) among the same donors.

A large difference existed between donors with specific IgE to IFA living in Augusta compared with those from OKC (17% vs 2%, $P = .0002$). The data further demonstrated a significant difference between the prevalence of specific IgE to YJV between donors living in Augusta and those living in OKC (10% vs 6%, $P = .04$). No difference was noted in the prevalence of peanut-specific IgE by city (7.5% vs 6.5%, respectively, $P = .60$).

The 10 sera with positive IgE to both IFA and YJV were evaluated by inhibition. Addition of concentrated YJV produced an average inhibition of $50.5\% \pm 25.6\%$ to YJV, compared with $10.9\% \pm 17.6\%$ ($P = .001$) for IFA. Repeat analysis of 10 randomly selected sera that had positive specific IgE to IFA showed a coefficient of variation of 2.6%.

DISCUSSION

These results show that nearly 1 of every 5 blood donors from Augusta has detectable IFA-specific IgE. Furthermore, adults living in this IFA-endemic region are significantly more likely to be sensitized to IFAs than they are to two other allergens associated with fatal anaphylaxis, YJV and peanut. The high prevalence of sensitization to IFAs suggests that stings from *S invicta* may represent the greatest risk of anaphylaxis for adults living in IFA-endemic areas. To our knowledge, this is the first study documenting the prevalence of sensitization to IFA in a large sample of relatively unselected adults living in an endemic region of the United States.

After moving to an endemic region, adults are rapidly exposed and sensitized to IFAs. Tracy et al⁹ found that 51% of individuals newly relocated to an IFA endemic region reported being stung within the first 3 weeks of arrival. Of those individuals, 15% became sensitized and demonstrated IFA-specific IgE.⁹ The 15% sensitization rate from the study of Tracy et al is similar to the 17% sensitization rate demonstrated by our study, suggesting that sensitization may rapidly reach a plateau or equilibrium in IFA-endemic areas. Considering estimates that 77 million individuals will be living in an IFA-endemic region in the near future, a 17% sensitization rate means that 13 million people may be at risk for allergic reactions from IFA stings. Currently, it is difficult to precisely estimate the percentage of the sensitized population at

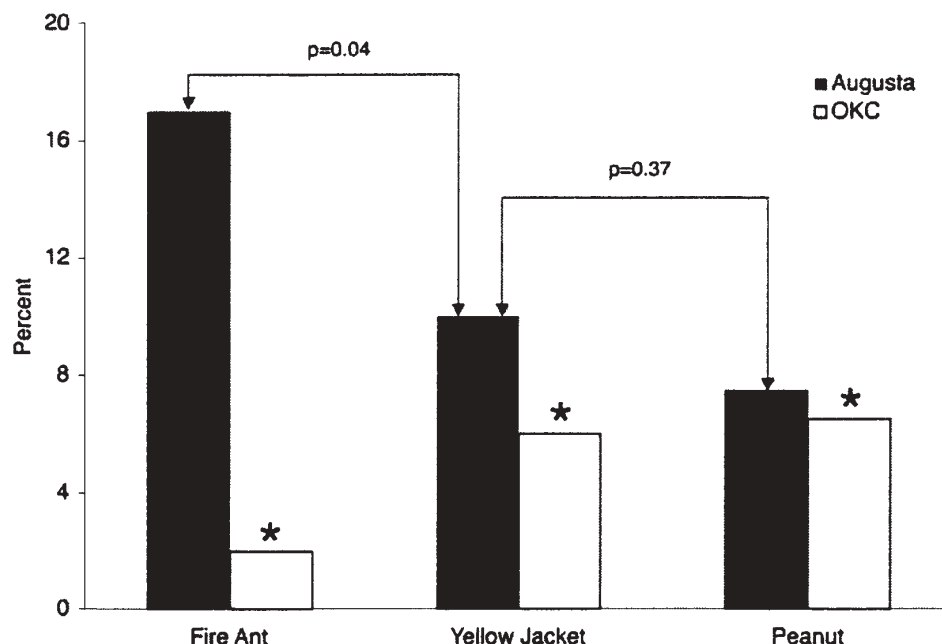


FIG 1. Percentage of adults with specific IgE to listed allergens at a concentration of >0.35 kIU/L. * $P > .07$, comparing prevalence of detectable IgE with listed allergens in OKC samples.

TABLE I. Demographic data of blood donors

Category	Augusta, Ga	OKC	OKC subset
Sample size	200	1000	100
Female, n (%)	93 (46)	489 (49)	53 (53)
Male, n (%)	107 (54)	511 (51)	47 (47)
Age range (y)	17-80	17-80	17-80
Mean age (y)	47	43	44
SD (y)	13.5	14.0	16.1

risk of anaphylaxis. Freeman et al¹⁰ reported 6 individuals sensitized to IFA who all had anaphylactic reactions after being stung. Other surveys report that from 0.6% to 16% of sensitized individuals will have anaphylactic reactions after stings.⁴⁻⁶ If the true risk of anaphylaxis lies somewhere between these extremes, a large number of people are at risk for IFA-induced anaphylaxis.

Our study analyzed sera from three groups of donors: Augusta, OKC, and a subset of OKC donors. Although a statistical difference was found between the mean age of Augusta donors and OKC donors, it is unlikely that this small difference accounts for the differences in the prevalence of allergic sensitization.

The anticipated difference between adults with specific IgE to IFA from an endemic region and adults with IgE to IFA from a nonendemic region was demonstrated; a difference was also found between blood donors from Augusta and OKC with YJV-specific IgE. We believe the higher prevalence in Augusta probably is due to a higher average annual temperature and adults spending more time outdoors, increasing the exposure risk to stinging insects. The insignificant difference in the prevalence of peanut-

specific IgE between Augusta and OKC suggests that neither donor group had a higher overall risk of allergy.

Although IFA immunotherapy has been practiced for approximately 30 years, no established guidelines or indications for its use exist.¹ No purified extract of fire ant venom is commercially available in the United States. IFA venom has been shown to be more potent than WBE in skin testing.¹¹ However, WBE is being routinely used for skin testing, immunotherapy, and in vitro analysis because it has been shown to contain allergens to which individuals are allergic.^{11,12}

The sensitivity and specificity of the Pharmacia UniCAP system through the use of WBE has been reported at 82% and 90%, respectively.¹³⁻¹⁵ Our inhibition assays on the dual-positive sera confirm the high level of assay specificity for IFA-specific IgE. If 1 donor among the 10 who had similar inhibition to IFA and YJV (presumably because much of the IgE was specific for Sol i 1, an allergen known to cross-react with YJV¹⁶) was excluded from analysis, the average inhibition to IFA in the remaining 9 serum samples was 5.6%, compared with 51.4% inhibition of YJV ($P < .0001$). These results strongly suggest that cross-reactivity did not contribute to the prevalence of IFA-specific IgE compared with YJV-specific IgE in the endemic region. Furthermore, of the 34 individuals who were positive for specific IgE to IFA, only 10 were also positive for specific IgE to YJV. The low prevalence of cross-reactive IgE between IFA and YJV is further supported by the low prevalence of IFA-specific IgE in the OKC subset.

Asymptomatic sensitization to other hymenoptera, including honeybee and YJV, has been reported in previous studies at approximately 15%.^{17,18} Our results show

the prevalence of adults with IgE to YJV to be 10% in the Augusta population and 6% in the OKC population. The differences between the studies may be attributed to different geographical regions and the fact that our study tested for IgE to YJV independently, whereas the previous studies included prevalence of IgE to both YJV and honey bee in their results.

Our study design had several strengths. We were able to survey a relatively large number of unselected individuals from an IFA-endemic region. The UniCAP analysis delivers high levels of sensitivity and specificity when testing for serum-specific IgE to solid-phase allergens.¹³ A control population (OKC) was also tested to support the validity of our results. Limitations of the study included the exclusively adult age group, the lack of a truly random sample, and the lack of additional demographic information and medical history needed for more thorough clinical correlation.

In summary, the prevalence of specific IgE to IFA among adults residing in an endemic region was significantly greater than the prevalence of IgE to either YJV or peanut. Our study suggests that stings from IFAs may pose the greatest risk among these allergens for anaphylaxis in adults living in Augusta, Georgia. Studies in other IFA-endemic regions would likely show similar results suggesting that all physicians practicing in IFA-endemic regions need to be vigilant in recognizing IFA allergy.

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