

Comparison of the allergenicity and antigenicity of yellow jacket and hornet venoms

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The immunologic properties of yellow jacket and hornet venoms were compared by measuring their reaction with rabbit antisera and human IgE and IgG antibodies. Anti-hornet venom rabbit serum showed precipitin bands unique to hornet venom and several bands crossreacting with yellow jacket venom. Anti-yellow jacket venom rabbit serum reacted with yellow jacket venom but failed to react with the hornet venoms. Most sera from patients who had had allergic reactions after vespid stings reacted with yellow jacket and hornet venoms in RAST analysis. A few sera reacted with only one of the venoms. RAST inhibition studies confirmed the crossreactivity of these IgE antibodies. The IgG antibody response of 14 patients was measured after yellow jacket venom immunotherapy. All had rising titers of yellow jacket venom-specific IgG. There was also an increase in the IgG antibody response measured with hornet venom in the majority of patients. The rise was significant with yellow hornet venom ($p < 0.02$) but failed to reach significance with bald-faced hornet venom ($p > 0.05$). In IgG radioimmunoassay inhibition studies using yellow jacket venom-coupled discs, yellow jacket venom was considerably more potent than hornet venom. These studies indicate major crossreactivity between yellow jacket and hornet venoms. In this group of patients, yellow jacket venom appeared to be the primary allergen. (J ALLERGY CLIN IMMUNOL 69:268, 1982.)

One of the current problems in the field of stinging-insect allergy is the question of crossreactivity or individual sensitivity to the vespid venoms (yellow jacket, hornet, and wasp). Positive skin-test reactions to several vespid venoms are common in the evaluation of patients with insect-sting sensitivity. Present recommendations are to treat patients with each of the venoms to which reactions are found.¹

From a practical viewpoint, most allergic reactions follow a single sting and thus suggest that there is only one specific allergen. From a biologic viewpoint,

the vespids are all in the same family, separate from the apids (honeybee).

Prior studies from our laboratory have examined the relative antigenicity and allergenicity of venoms from different yellow jacket species² and from four hornet species.³ The results of these studies have suggested that with one exception, the venoms of the yellow jacket species examined all had similar antigenic properties. Venoms of the two common North American hornets, *Vespula maculata* and *Vespula arenaria*, had similar antigenic properties.

It is the purpose of this report to describe the comparison of the antigenic and allergenic properties of the yellow jacket and hornet venoms. Results of these studies suggest that there is a major crossreactivity between these venoms and that the majority of individuals showing dual sensitivity are reacting to a common allergen(s).

MATERIALS AND METHODS

Antigens

Yellow jacket and hornet venoms were collected locally by electrical stimulation.^{4, 5} The yellow jacket venoms were

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obtained from *Vespula maculifrons*, *Vespula germanica*, and *Vespula vulgaris* species. A mixture was used for these studies. Hornet venoms were obtained from both common North American species, *V. maculata* (bald-faced hornet) and *V. arenaria* (yellow hornet). After venom collection, all the venoms were dissolved in borate-buffered saline, pH 8, filtered through a Millipore filter (1.2 μ) and were lyophilized after protein assay by biuret reaction.

Yellow jacket venom (*V. maculifrons*), bald-faced hornet venom, and yellow hornet venom solutions, prepared by venom sac extraction,⁶ were obtained through the courtesy of Dr. Daniel Mullally, Head, Collaborative Product Section, IAIDP-NIAID, National Institutes of Health, Bethesda, Md.

Rabbit antibodies

Immunization of rabbits. Two New Zealand white rabbits weighing 8 to 10 pounds were immunized with each venom preparation by intradermal injection in multiple sites at 2- to 3-wk intervals. For each injection, 0.2 mg of yellow jacket venom or 0.5 mg of hornet venom in 0.5 ml of saline was suspended in an equal volume of complete Freund's adjuvant. A higher hornet venom dose had to be given to obtain a peak antibody response (titer $\geq 1/32$ in double-diffusion gel precipitation). Blood samples were obtained from 10 to 14 days after the fourth and fifth injections.

Double-diffusion gel preparation. Double-diffusion gel precipitation in 1% agarose was done as described previously.²

Immunoelectrophoresis. Plates of 8 by 10 cm were coated with 12 ml of 1% agarose in acetate buffer, pH 5, 0.025M. A 75- μ g volume of antigen was applied to each well. Electrophoresis was run at 10 V/cm for 50 min in acetate buffer, pH 5, 0.05M. Troughs were filled with 150 μ l of the respective antisera. Immunodiffusion was carried out for 24 hr at room temperature in a moist chamber. The plates were then washed, dried, and stained with 0.2% Coomassie Brilliant Blue. When the troughs were filled with normal rabbit serum instead of antiserum, no protein-staining precipitation lines resulted with any of the venoms.

IgG-radioimmunoassay. Rabbit serum venom-specific IgG was determined by a modification of the solid-phase radioimmunoassay using ¹²⁵I-labeled protein A.⁷ Details of this modification are described below. One milligram of yellow jacket venom protein and 2 mg of hornet venom protein, obtained by electrical stimulation, were used to couple 200 cyanogen bromide-activated discs. Serial two-fold dilutions of each rabbit antiserum starting at a dilution of 1:1000 were prepared in 1% ovalbumin. Aliquots (100 μ l) of each dilution were added to the homologous venom disc and incubated for 2 hr at room temperature. After the discs were washed three times with buffered saline containing 0.2% ovalbumin and 1% Tween, 50 ng of ¹²⁵I-labeled protein A in 50 μ l of diluent were added. After a 4-hr incubation, the discs were washed three times and the residual radioactivity was counted.

A dilution of each rabbit antiserum that bound the same amount of protein A with the homologous discs was selected (4000 cpm, 7% of total radioactivity). This dilution

was then used for incubation with the discs coupled with the other venoms.

Human IgE antibodies

RAST. Sera from 84 patients from Western New York State with systemic allergic reactions after stings by yellow jackets and hornets were obtained before starting immunotherapy. Specific IgE antibodies to yellow jacket and hornet venoms were measured by RAST as previously described.⁸ The same cellulose discs used for the rabbit IgG antibody assay were used. RAST results were expressed in percent of net binding of a serum pool from ragweed-sensitive patients to ragweed discs. A net binding to the venom disc $>10\%$ was considered significant. The background activity bound when normal human serum was assayed was 0.7% for ragweed discs and varied between 0.7% and 1.5% of the total activity added for the venom discs. The maximal binding of the ragweed standard went up to 15%.

RAST inhibition. The RAST inhibition method was performed as described elsewhere.⁹ Briefly, 100 μ l of serial threefold dilutions of the venoms starting at 625 μ g of venom protein were used for absorption of 50 μ l of the individual sera from two patients with high levels of IgE antibodies against yellow jacket and hornet venoms and for absorption of 50 μ l of a serum pool obtained from six patients with high titers of IgE antibodies reacting with both venoms.

Human IgG antibodies

IgG radioimmunoassay. Human serum venom-specific IgG was determined by a modification of the solid-phase radioimmunoassay using ¹²⁵I radiolabeled protein A. A volume of 30 μ g of protein A was labeled with approximately 1200 μ Ci of ¹²⁵I for a period of 2 min, using chloramine-T and the reagents supplied by Zymed Laboratories, Burlingame, Calif. After filtration through a Sephadex G-25 column equilibrated with 0.2% ovalbumin, the stock preparation was stored at 4° C at a concentration of 1 μ g of protein/ml in 0.2% ovalbumin.

Preliminary experiments were carried out to determine the optimal amount of radiolabeled protein A for these studies. The stock ¹²⁵I-labeled protein A was initially diluted 1:5 in 0.2% ovalbumin (200 ng/ml). To 0.5-ml aliquots of this dilution were added 0.5-ml aliquots of either 0.1% ovalbumin or serial 10-fold dilutions of unlabeled protein A ranging in concentration from 0.2 to 200 μ g/ml. The final concentrations of total protein A ranged from 0.1 to 100 μ g/ml.

Initially, 50- μ l aliquots of a 1:40 dilution of a serum with a high titer of venom-specific IgG or 50 μ l of a similarly diluted normal serum were added to venom-coupled paper discs. The discs were incubated for 3 hr at room temperature and then washed three times with normal saline, after which 100 μ l of each protein A solution were added to two positive and to two control serum discs. After overnight incubation at room temperature the radioactivity was measured. The percent of binding increased with decreasing total protein A, relative to the constant amount of radiolabeled protein A, for both positive and control discs.

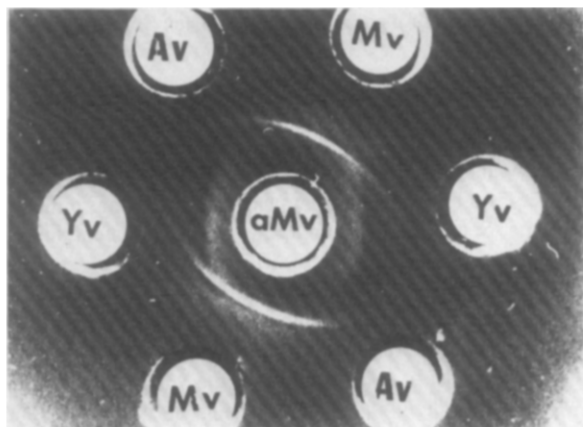


FIG. 1. Double-diffusion gel precipitation experiment showing the reaction between anti-bald-faced hornet venom rabbit serum (*aMv*) and bald-faced hornet venom (*Mv*), yellow hornet venom (*Av*), and yellow jacket venom (*Yv*). The rabbit antiserum was undiluted; the venoms were used in concentrations of 400 μ g of protein/ml.

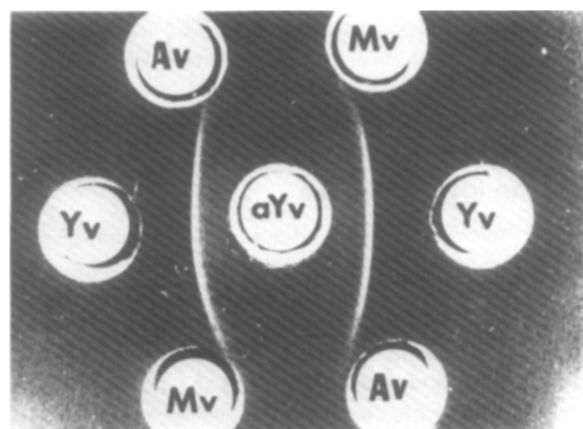


FIG. 2. Double-diffusion gel precipitation experiment showing the reaction between anti-yellow jacket venom rabbit serum (*aYv*) and bald-faced hornet venom (*Mv*), yellow hornet venom (*Av*), and yellow jacket venom (*Yv*). The rabbit antiserum was undiluted; the venoms were used in concentrations of 400 μ g of protein/ml.

The ratios of specific/control counts were increased from 1:1 at a concentration of 100 μ g of protein A to a ratio of 5:1 at a protein A content of 1 μ g/ml and then dropped to 2:1 with further decrease in protein A content.

The procedure was further modified after several titrations of a dilution of the positive control serum. A serum dilution of 1:50 was subsequently chosen, with a first incubation time of 2 hr. The protein A (50 μ l containing 50 ng of protein A) incubation time was also studied. With decreasing incubation periods, nonspecific background binding was diminished, suggesting an optimal incubation time of 90 min. With this modification, the counts per minute ratio of positive to control serum is of the order of 30:1 for a serum containing 100 units of bee venom-specific IgG antibody/ml and 15:1 for serum with 50 units.

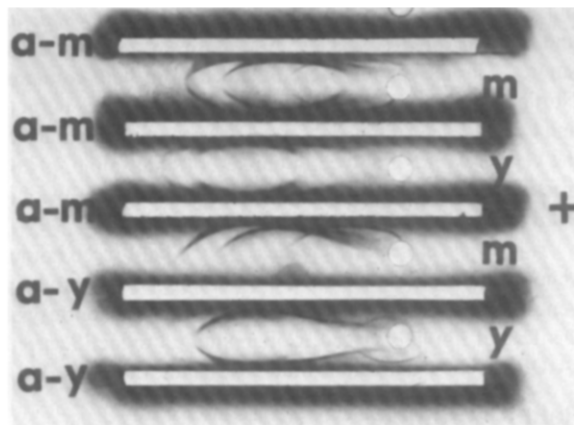


FIG. 3. Immunoelectrophoresis of venoms from yellow jackets (*y*) and bald-faced hornets (*m*) against rabbit antiserum to these venoms (*a-y* and *a-m*).

The same venom-coupled discs used for the RAST and rabbit antisera analyses were used. Preliminary experiments were done to determine the optimal amount of venom for coupling to the discs (5 μ g of yellow jacket and 10 μ g of hornet venom per disc). A serum with a high titer of yellow jacket venom-specific IgG was used as a positive standard. A 50- μ l volume of a 1:50 dilution of serum or the test sera in borate-buffered saline containing 0.2% ovalbumin and 1% Tween were added to each tube containing a venom-coupled disc. After incubation for 2 hr at room temperature, the discs were washed three times in buffered saline, and 50 μ l of radiolabeled protein A were then added. After a 90-min incubation at room temperature, the discs were washed three times with buffered saline and the radioactivity bound was determined in a gamma counter. The background activity bound for normal human serum varied between 0.9% (yellow jacket discs) and 1.7% (yellow hornet discs) of the total activity added. The binding of the standard serum at 1:50 dilution was 12.8% of total activity. Results are expressed in percent net binding of the total activity.

Sera were obtained from 14 insect-sensitive patients prior to and after immunization with yellow jacket venom. Venom-specific IgG was determined with discs coupled with either yellow jacket, bald-faced hornet, or yellow hornet discs.

Inhibition of yellow jacket venom-specific IgG. Sera from two patients with high titers of yellow jacket venom-specific IgG were used for these assays. A 100- μ l volume of serial 10-fold dilutions of yellow jacket or bald-faced hornet venoms were used to absorb 50 μ l of 1:50 dilution of the patients' sera. After overnight incubation at 4°C, discs were then added and the procedure continued as described above.

RESULTS

Rabbit antibodies

Fig. 1 shows the double-diffusion gel precipitation experiment between anti-bald-faced hornet venom rabbit serum and bald-faced hornet, yellow hornet,

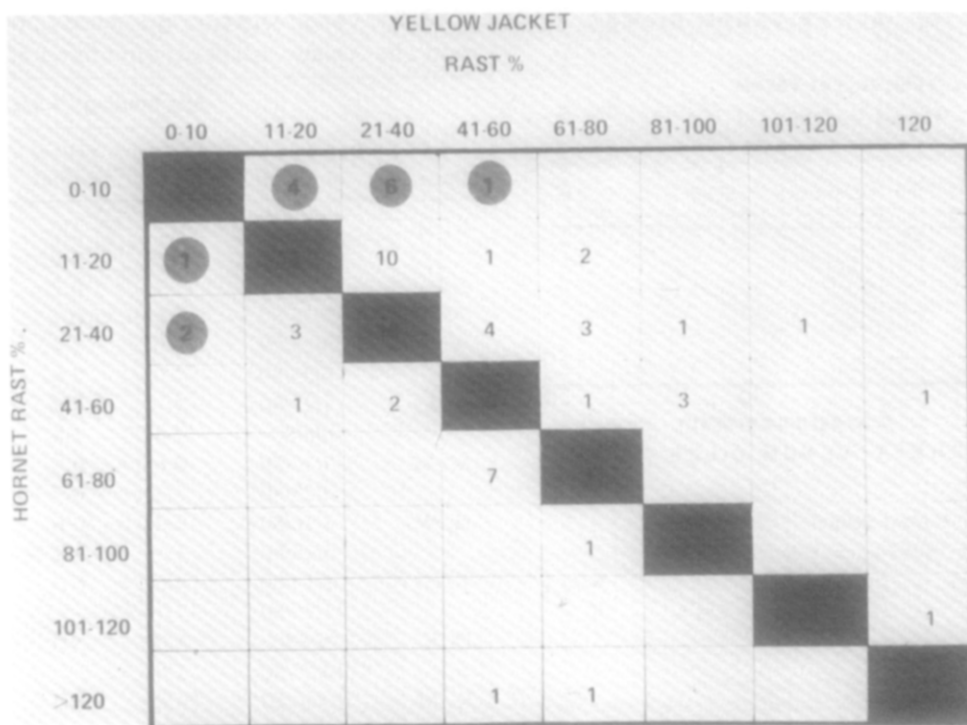


FIG. 4. Direct RAST results of 84 sera from vespid-sensitive patients, expressed as percent of a positive standard. Yellow jacket RAST results are shown along the horizontal axis; hornet RAST results along the vertical. Results greater than 10% are considered significant. Numbers in the dark squares indicate sera reacting equally to yellow jacket and hornet venoms. Numbers in dark circles indicate sera reacting to only one venom.

and yellow jacket venoms. There are several lines of precipitation in the homologous system, at least one of which appears to be unique to bald-faced hornet. Other lines of precipitation appear to crossreact with yellow hornet venom and with yellow jacket venom. Similar results were found when yellow hornet venom antiserum was used for this experiment, with a unique antigen also present in the yellow hornet venom.

Fig. 2 shows the reaction of anti-yellow jacket venom rabbit serum with yellow jacket venom and the two hornet venoms. In this experiment, there is one strong band of precipitation between yellow jacket venom and its antiserum and no lines of precipitation can be seen with either of the hornet venoms. Similar results were obtained with venom sac extracts of yellow jacket, bald-faced hornet, and yellow hornet and their corresponding antisera.

The immunoelectrophoretic results are shown in Fig. 3. With hornet venom antiserum, five precipitin bands are detected in hornet venom and three in yellow jacket venom. Antiserum to yellow jacket venom and yellow jacket venom react with at least three precipitin lines. Yellow jacket antiserum does not appear to react with hornet venom under these conditions.

The IgG radioimmunoassay results were similar.

Anti-bald-faced hornet venom rabbit serum and anti-yellow hornet venom rabbit serum bound strongly to both hornet discs and bound to a lesser degree to yellow jacket venom discs. Anti-yellow jacket venom rabbit serum bound strongly to yellow jacket discs, bound only minimally to bald-faced hornet discs, and did not bind to yellow hornet discs.

Human IgE antibodies

Fig. 4 shows the results of direct RAST examining 84 sera with yellow jacket and bald-faced hornet venoms. Most patients had only one systemic reaction after a sting. The serum was obtained within 3 mo of the sting reaction. Most sera reacted with both venoms and there was a good correlation between the RAST results with the two venoms ($r = 0.63$, $p < 0.001$). Three sera reacted with hornet venom only, and 11 sera reacted with yellow jacket venom only. RAST titers were generally higher with the yellow jacket than with the hornet venom in the majority of the patients.

In Fig. 5, RAST inhibition experiments with sera from two patients with high-titer yellow jacket venom- and hornet venom-specific IgE are shown. With both sera examined, yellow jacket venom was a

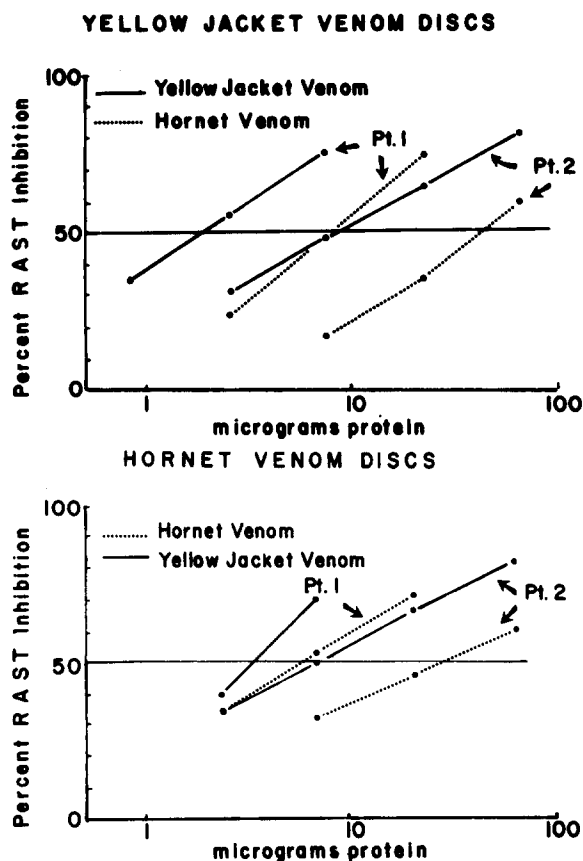


FIG. 5. RAST inhibition experiments showing inhibition of the reaction between two sera from insect-sensitive patients and discs coupled with yellow jacket venom and bald-faced hornet venom by yellow jacket and bald-faced hornet venoms. With both sera and both discs, yellow jacket venom was a more potent inhibitor.

more potent inhibitor than hornet venom, using both yellow jacket venom- and hornet venom-coupled discs. Similarly, with the serum pool obtained from patients with high-titered antibodies, yellow jacket venom was a more potent inhibitor than hornet venom with both yellow jacket venom- and hornet venom-coupled discs (Fig. 6).

Human IgG antibodies

Serum was obtained from 14 patients prior to and after yellow jacket immunization. All patients were receiving a top dose of between 50 and 100 μ g of venom when the second serum sample was obtained. Paired sera were examined at the same time. The results are shown in Table I.

After therapy, all 14 patients had increased titers of yellow jacket venom-specific IgG. Most paired sera showed increased titers of bald-faced hornet- and yellow hornet-specific IgG. The paired t test was used for analysis of the change in antibody titers, comparing pretreatment and posttreatment sera. For yellow jacket venom- and yellow hornet venom-specific

TABLE I. Venom-specific IgG activity prior to and after yellow jacket venom immunotherapy

Patient	Dates serum obtained	Net binding (% total activity)		
		Yellow jacket	Yellow hornet	Bald-faced hornet
R. M.	8/31/79	0.5	1.0	0.4
	5/27/80	3.9	3.0	1.5
J. P.	2/22/80	1.2	0.2	0
	5/28/80	2.1	0.4	0.2
W. S.	9/25/79	1.0	1.4	2.0
	6/25/80	2.0	2.6	2.2
K. S.	1/22/80	1.7	0.9	0.6
	6/6/80	2.4	1.8	1.0
M. W.	7/17/79	0.6	0	0.1
	4/23/80	2.3	0.7	0.3
H. W.	1/16/80	0.2	0.6	0.6
	5/28/80	1.5	3.2	2.7
T. C.	9/15/80	4.5	ND	1.6
	11/26/80	17.1		3.3
D. K.	5/9/80	0.0	ND	0.2
	10/27/80	1.8		1.7
K. M.	10/10/80	4.8	ND	0.6
	3/18/81	7.8		0.4
J. M.	9/3/80	3.6	ND	0.9
	3/31/80	6.6		0.7
J. O.	4/24/78	0.05	ND	0.6
	7/30/79	4.10		1.2
A. P.	8/6/80	0.1	ND	0.3
	1/5/81	2.6		0.7
J. T.	10/3/80	1.8	ND	2.1
	4/24/81	3.4		2.3
S. T.	2/17/81	0.21	ND	0.8
	5/6/81	2.10		1.5

ND = not done.

IgG this increase was significant ($p < 0.02$). For bald-faced hornet venom, the change in IgG was not significant ($p > 0.05$).

Fig. 7 shows the inhibition of yellow jacket venom-specific IgG by bald-faced hornet and yellow jacket venoms. Two sera that had high-titered yellow jacket-specific IgG were examined. In both cases, yellow jacket venom inhibited to a greater extent than bald-faced hornet venom. The difference in inhibiting potency was especially striking in patient 3.

DISCUSSION

The rabbit antisera raised against both hornet venoms reacted with these venoms with a number of precipitin bands. Several precipitin bands also were found to crossreact when antisera against hornet venoms were assayed with yellow jacket venom. In addition, each of the hornet venoms appeared to have antigens of its own. Interestingly, anti-yellow jacket venom rabbit serum, although reacting with yellow

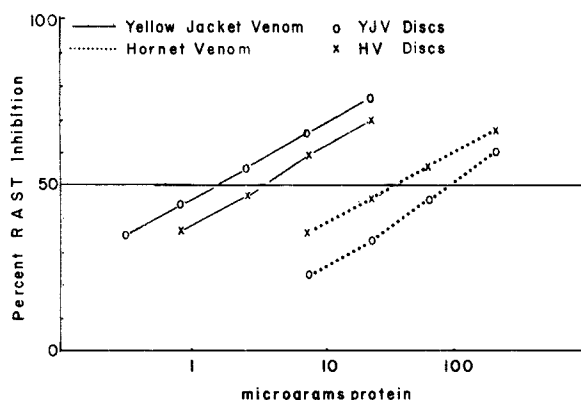


FIG. 6. RAST inhibition experiments showing inhibition of the reaction between pooled sera from six insect-sensitive patients and discs coupled with yellow jacket and bald-faced hornet venoms. Yellow jacket venom was a more potent inhibitor.

jacket venom, failed to react with hornet venom. These immunodiffusion experiment results were confirmed by measurements of rabbit IgG antibodies by radioimmunoassay. These latter experiments showed binding of anti-hornet venom rabbit serum to both hornet and yellow jacket venom discs but only minimal binding of anti-yellow jacket venom rabbit serum to hornet venom discs.

These results suggest that the rabbit recognizes an antigen in hornet venom which is similar to that found in yellow jacket venom. On the other hand, the yellow jacket venom apparently contained a different antigen that does not crossreact with hornet venom. The same results were found with the venoms obtained by venom sac extraction. It is possible that the lower dose of yellow jacket venom needed to induce adequate rabbit antibody response is responsible for this discrepancy. Perhaps higher venom-dose immunization would have resulted in further antibody formation with a broader range of specificity.

Our results with rabbit antisera may differ somewhat from those found by King et al.¹⁰ These investigators found similar protein composition in venoms (obtained by sac extraction) from bald-faced hornets, yellow hornets, and yellow jackets. They described three basic proteins: hyaluronidase, phospholipase, and an unidentified antigen, "antigen 5." With rabbit antisera, in one set of rabbits there was strong reaction with the hyaluronidase fraction and very weak cross-reaction with the phospholipase and antigen 5. Anti-yellow jacket rabbit serum did react with these hornet fractions. However, in repeat experiments with two sets of rabbits, crossreactivity between the venoms could not be detected.

In the RAST, most sera from patients with vespid-sensitivity reacted with both yellow jacket

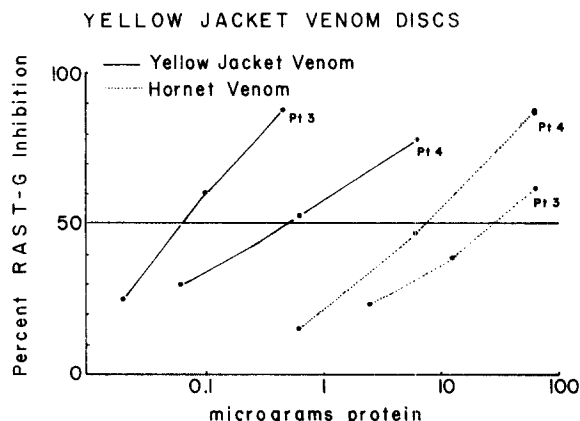


FIG. 7. Inhibition of the reaction between human serum yellow jacket venom-specific IgG and yellow jacket venom-coupled discs by yellow jacket and bald-faced hornet venoms.

and hornet venoms. There were only a few sera reacting with yellow jacket and hornet venoms alone. Further RAST studies with dialyzed venom as the coupling antigen showed even a better correlation between yellow jacket and hornet venom results. These results confirm the common clinical observation that most patients react to the majority of vespid venoms in skin testing even after a single sting reaction.

The direct RAST results were confirmed by RAST inhibition studies. Both venoms significantly inhibited the reaction with both yellow jacket venom- and hornet venom-coupled discs. Since yellow jacket venom was a better inhibitor even with hornet venom-coupled discs, it appeared to be the primary sensitizing agent for the two patients studied. Similar results were obtained with the serum pool.

Kern et al.¹¹ studied the cross-sensitivity of the IgE antibody response to various vespid venoms, including yellow jacket, bald-faced hornet, yellow hornet, and *Polistes*. These authors found that using the criterion of the venom concentration required for 50% histamine release from cells of insect-allergic patients, no allergic crossreactivity among the venoms existed except for a modest association between yellow hornet and bald-faced hornet venoms. Despite these findings, 13 of 18 patients did react to three or four venoms tested. These latter results did suggest the presence of multiple major allergens, several of which might crossreact. The reason that the histamine-release results did not show a correlation as good as that for RAST results may be related to problems of the histamine-release technique or to differences in the actual allergenic content of the venoms used.

We attempted to determine whether the human system differed from the rabbit system in IgG antibody activity. The majority of patients who mounted an

IgG antibody response to yellow jacket venom after immunotherapy with yellow jacket venom also showed a rising titer when tested with bald-faced hornet or yellow hornet venoms. Inhibition studies also showed that bald-faced hornet venom would inhibit the yellow jacket IgG radioimmunoassay to some extent, although not to the same degree as did the homologous venom. Sera were not available from patients receiving immunotherapy with hornet venom only. Thus studies of crossreactivity of human IgG antibody response to hornet venom could not be done.

Golden et al.¹² also showed that selected patients with multiple positive vespid-venom skin tests are primarily sensitive to yellow jacket venom. The patients were selected on the basis of RAST inhibition profiles, which failed to show specific IgE antibody reaction to wasp or hornet venoms. One group of patients tolerated a wasp-sting challenge after treatment with yellow jacket venom only or with yellow jacket and hornet venoms. The second group tolerated hornet-sting challenges after yellow jacket venom therapy.

These results suggest that there are crossreacting antigens in yellow jacket and hornet venoms that account for the majority of the reactions from these vespids. This knowledge does not entirely resolve the choice of appropriate immunotherapy. Species-specific antigens are present and may induce sensitization. Treatment with only one venom might therefore be insufficient. On the other hand, treatment with multiple venoms is expensive and may sensitize patients with a single sensitivity to new antigens. This problem may only be resolved definitively when purified venom fractions are available for evaluation. As noted, RAST inhibition profiles have been suggested as one means of resolving this question of unique or cross-sensitivity. However, unique minor antigens might not be detected by such a technique in patients with concurrent sensitization to common major antigens. Furthermore, this technique is too cumbersome and costly to do on a routine basis.

These results also suggest that in the patient population studied, yellow jacket venom was the primary

sensitizing agent. This conclusion is in agreement with our clinical observations that most patients in our area are stung by yellow jackets. The rational conclusions from these observations would be that yellow jacket venom is the appropriate immunotherapeutic antigen for the majority of these patients.

REFERENCES

1. Lichtenstein LM, Valentine MD, Sobotka AK: Insect allergy: the state of the art. *J ALLERGY CLIN IMMUNOL* **64**:5, 1979.
2. Wicher K, Reisman RE, Wypych J, Elliott W, Steger R, Matthews RS, Arbesman CE: Comparison of the immunogenicity of venoms of various species of yellow jackets (genus *Vespula*). *J ALLERGY CLIN IMMUNOL* **66**:244, 1980.
3. Mueller U, Elliott W, Reisman R, Ishay J, Walsh S, Steger R, Wypych J, Arbesman CE: Comparison of biochemical and immunologic properties of venoms from four hornet species. *J ALLERGY CLIN IMMUNOL* **69**:290, 1981.
4. Benton AW, Morse RA, Stewart DJ: Venom collection from honeybees. *Science* **142**:228, 1963.
5. Eskridge EM, Elliott WB, Elliott AH, Eskridge PB, Doerr JC, Schneller N, Reisman RE: Adaptation of the electrical stimulation procedure for the collection of vespid venoms. *Toxicon* (in press, 1982).
6. Simon RP, Benton AW: A method for mass collection of wasp venoms. *Ann Entomol Soc Am* **62**:277, 1969.
7. Hamilton RG, Sobotka AK, Adkinson NF Jr: Solid-phase radioimmunoassay for quantitation of antigen-specific IgG in human serum with ¹²⁵I protein A from *Staphylococcus aureus*. *J Immunol* **122**:1173, 1979.
8. Light WC, Reisman RE, Rosario NH, Arbesman CE: Comparison of the allergenic properties of bee venom and whole bee body extract. *Clin Allergy* **6**:293, 1976.
9. Wypych JI, Reisman RE, Elliott WB, Steger RJ, Arbesman CE: Immunologic and biochemical evaluation of the potency of whole insect body extracts. *J ALLERGY CLIN IMMUNOL* **63**:267, 1979.
10. King TP, Sobotka AK, Alagon A, Kochoumian L, Lichtenstein LM: Protein allergens of white-faced hornet, yellow hornet and yellow jacket venoms. *Biochemistry* **17**:5165, 1978.
11. Kern F, Sobotka AK, Valentine MD, Benton AW, Lichtenstein LM: Allergy to insect sting. III. Allergic cross-reactivity among the vespid venoms. *J ALLERGY CLIN IMMUNOL* **57**:554, 1976.
12. Golden DBK, Valentine MD, Kagey-Sobotka A, Lichtenstein LM: Cross reactivity of vespid venoms, in *Proceedings of the 37th Annual Meeting of the American Academy of Allergy*, San Francisco, 1981, p. 57.