

## HORNET VENOMS: LETHALITIES AND LETHAL CAPACITIES

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J. O. SCHMIDT, S. YAMANE, M. MATSUURA and C. K. STARR. Hornet venoms: lethality and lethal capacities. *Toxicon* 24, 950–954, 1986.—The i.v. LD<sub>50</sub> values to mice of pure venoms of *Vespa mandarinia japonica*, *V. simillima xanthoptera*, *V. tropica deusta* and *V. l. luctuosa* were, respectively, 4.1, 3.1, 2.8 and 1.6 mg/kg. The LD<sub>50</sub> value of 1.6 mg/kg distinguishes the venom of *V. luctuosa* as the most lethal known wasp venom. To measure the absolute lethality of a single sting, a new index, called lethal capacity, based on the amount of venom possessed by an individual and its lethality is presented. *V. mandarinia* and *V. tropica* are the most venomous known insects, with a lethal capacity of one sting from *V. mandarinia* delivering an LD<sub>50</sub> (i.v.) dosage of venom to 270 g of mouse. The lethal capacity for an entire hornet colony, called colony lethal capacity, for *V. tropica* is 84 kg of mouse/colony.

HORNETS in the genus *Vespa* are the largest stinging insects in the world. These often conspicuously colored species attract both lay and scientific interest, are sometimes serious predators of honey bees (MATSUURA and SAKAGAMI, 1973; ISHAY *et al.*, 1967) and their stings can induce allergic reactions in hypersensitive individuals (ORI and HIYAMA, 1977; ABE, 1985; see SCHMIDT, 1986b, for general discussion). Extensive literature pertaining to the genus *Vespa* (SPRADBERY, 1973; EDWARDS, 1980; MATSUURA and YAMANE, 1984) and specifically to the four species of *Vespa* used in this study (MATSUURA, 1984; KOJIMA and YAMANE, 1980; KOJIMA, 1982) is available. With a few exceptions (e.g. KAWAI *et al.*, 1980; ABE *et al.*, 1982; ABE and KAWAI, 1983), little is known about the venoms of the hornets of eastern Asia and their venom lethality are unknown. We report here the results from four diverse East Asian species and determine their capacities as defensive agents.

Worker hornets of *V. mandarinia japonica* Radoszkowski, *V. simillima xanthoptera* Cameron and of *V. tropica deusta* Lepeletier and *V. l. luctuosa* de Saussure were collected, respectively, 11–13 August 1980 in Hitachi Daigo and Mito, Japan, 17 August 1980 in Yuasa, Wakayama Prefecture, Japan, and 1 June 1985 in Inang, Sorsogon Province, Philippines. Immediately upon collection the workers of all species were placed on ice. Venom was extracted from cold-anesthetized live hornets or from live frozen (–10° to –20°C) individuals. All venoms were collected as pure fluids expressed from the sting tip into 5 or 10 µl microcapillary tubes. The microcapillary tube contents were then emptied

TABLE 1. VENOM YIELDS FOR FOUR SPECIES OF HORNETS

Species	Mean workers/ mature colony	Mean $\mu\text{l}$ venom/ individual	% dry wt in liquid venom	Venom dry wt/ worker (mg)
<i>V. mandarinia</i>	250	4.1	26.8	1.10
<i>    japonica</i>				
<i>V. simillima</i>	500	~2	21.2	~.42
<i>    xanthoptera</i>				
<i>V. tropica</i>	300	~3	~25.6	~.77
<i>    deusta</i>				
<i>V. luctuosa</i>	200	~1.5	28.8	~.43
<i>    luctuosa</i>				

into 250  $\mu\text{l}$  polyethylene Eppendorf centrifuge tubes, freeze dried and stored desiccated in the dark at  $-20^{\circ}\text{C}$  until used. To obtain values of per cent dry matter in the liquid venom, known quantities of liquid venom were dried and weighed. LD<sub>50</sub> values at 24 hr to Swiss white mice (six mice per dose) were determined by the method of REED and MUENCH (1938) with 95% confidence intervals calculated by the modification of PIZZI (1950) and means compared by the method described in WOOLF (1968).

When we disturbed the nests of *V. mandarinia*, defending workers flew towards us and when near hovered and made loud clicking sounds with their mandibles. These aposematic clicks were warning threats that usually preceded actual physical attacks. The nest from Hitachi Daigo contained the queen and 51 workers, from which 62  $\mu\text{l}$  clear venom and 8  $\mu\text{l}$  milky venom were obtained. When only those workers with full reservoirs were considered (18), an average yield of 3.5  $\mu\text{l}$  per worker with a maximum of 7  $\mu\text{l}$  was obtained. The nest of *V. mandarinia* from Mito contained the queen plus 161 workers which could be separated into groups of: (a) 104 good venom-yielding individuals with a mean venom content of 4.7  $\mu\text{l}$  and a maximum of 12.5  $\mu\text{l}$  per individual; (b) 19 failures that for one reason or another yielded no venom; (c) 16 teneral adults that produced no venom; (d) 19 milky venom-yielding individuals with a mean venom content of 4.7  $\mu\text{l}$  per individual; (e) 3 individuals crushed during the collecting. Excluding teneral and destroyed individuals, the overall average was 4.1  $\mu\text{l}$  venom per worker (Table 1).

Venom yields from *V. simillima* were smaller and less consistent than from *V. mandarinia*. The best extraction of *V. simillima* venom was from 21 individuals with a mean of 1.4  $\mu\text{l}$  per individual. This value is undoubtedly low and was probably caused by the hornets spraying venom during the collecting and handling processes. The maximum individual venom yielded for *V. simillima* was 4  $\mu\text{l}$  and we estimate the mean to be about 2  $\mu\text{l}$ .

Actual venom yields per hornet for *V. tropica* were not obtainable, due to the hurried and primitive conditions in the field. In qualitative terms they yielded large amounts of venom and an estimate of 2–4  $\mu\text{l}$  per individual would be reasonable. The collection and transport procedures for *V. luctuosa* entirely precluded quantitative venom measurement (85 hornets were required to produce 40  $\mu\text{l}$  venom). Under normal conditions we would expect about 1–2  $\mu\text{l}$  venom per individual from these small hornets.

The dry weights of 25  $\mu\text{l}$  fresh venom were 6.69 and 5.31 mg, respectively, for *V. mandarinia* and *V. simillima* and for 20  $\mu\text{l}$  of venom from *V. luctuosa* 5.75 mg. The *V. tropica* were wet when milked and some water was collected with venom from the sting tip, hence the value of 29.0 mg dry weight for 200  $\mu\text{l}$  venom is unrealistically low. In Table 1 the mean value of 25.6 for the three other venoms was used for *V. tropica* to allow estimated calculations of other values.