

Thesis Title Collection and Preparation of Vespid
 Venom for Skin Diagnostic Test in
 Vespid Sting Allergic Patients.

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ABSTRACT

The venom of *Vespa affinis* L. was collected by two methods: electrostimulation technique and venom sac extraction. The first method was performed by applying low voltage electricity via electrical excitation device to shock the hornets, provoking them to sting and inject venom droplets into the filter paper which was placed underneath the device. Thereafter, venom was extracted from the venom impregnated filter paper. The second method

was performed by direct extraction of venom sacs. The latter method provided much greater quantity of venom than the first one; however, the quality of the venom obtained was inferior due to contaminated sac components. Subsequently, both venom preparations were subjected to preliminary investigation of their constituents by thin-layer chromatography using a solvent system: n-butanol: pyridine: acetic acid: water = 90: 60: 18: 2. The substances identified in TLC of venom collected by electrostimulation were histamine, adrenaline and dopamine, whereas, those of venom sac extract were adrenaline and dopamine. In addition, TLC of venom sac extract also suggested the presence of additional substances other than venom components. Clinical investigation of venom as a diagnostic tool in insect sting allergy was conducted in 15 patients who had experienced insect sting and 10 individuals who denied previous insect sting. The test procedure was done by skin prick technique using three test solutions: venom collected by electrostimulation, venom sac extract and commercial wasp venom, each in concentration of 100 mcg/ml. The result of skin test seemed to correlate with clinical history; however, it could not be expressed in term of statistics because the number of patients, especially those who obviously manifested allergic reaction to sting and likely to produce positive skin test, was too

small. Concurrent with venom skin test, all subjects were investigated for total IgE antibodies and venom specific IgE antibodies in the sera using Phadezym IgE PRIST and Phadezym IgE RAST respectively. The sera level of total IgE antibodies did not correlate with clinical history and it seemed to be of no value in diagnosis of insect sting allergy. Venom specific IgE determination by RAST, which have been reported to be a reliable assessment of insect sting allergy by other authors, failed to give a reliable result in our experiment. The failure of RAST may be due in part to an inappropriate use of antigen disc and technical error.