

Evaluation of two pine oil-based formulations of Kitz against various life stages of *Aedes aegypti* (Linnaeus)

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Abstract: Two formulations, A and B, of a product Kitz®, containing pine oil, oleic acid and a wetting agent, were evaluated against various life stages of *Aedes aegypti*. Kitz B additionally contained 5% neem oil. Total mortality of eggs, first instar larvae, fourth instar larvae and pupae was obtained at 1, 0.1, 0.1 and 1% of Kitz A respectively. The concentrations of Kitz B that caused 100% mortality for these corresponding stages were 0.2, 0.2, 0.1 and 1% respectively. There was no significant difference between the two formulations in their effect on the mortality of the juvenile stages. Oviposition by female mosquitoes was drastically reduced at concentrations of 1% for both formulations, although it was not totally prevented. The presence of neem in Kitz® did not increase the product's toxicity to the juvenile stages; nor did it improve its efficacy as a repellent against oviposition by female mosquitoes.

Introduction

Aedes aegypti is the most important mosquito vector in Singapore. The present routine control programme consists of source reduction by removing stagnant water in and around buildings, supplemented with legislation and health education for the public. The emphasis of the current mosquito control programme is on non-chemical methods which have proved to be a success for many years. However, during dengue haemorrhagic fever outbreaks, which occur in cycles of about five years, fogging with insecticides has to be carried out (Chan, 1990). Thus the use of products that are relatively safe for non-target organisms to replace chemical insecticides is of immense practical significance. A non-toxic compound with surface-active properties, MonoxCi Foam, has been found to be effective in killing the larvae and particularly the pupae of another mosquito species, *Culex fatigans* (*C. quinquefasciatus*) (Ho and Wong, 1985).

A new non-toxic product, Kitz®, containing plant extracts and a wetting agent, is a natural disinfectant used commercially for cleaning purposes. However, the presence of pine oil, oleic acid and a wetting agent could possibly render the product effective against juvenile stages of mosquitoes. Pine timber has been reported to be protected from attack by termites due to the presence of terpenes (Moore, 1965). Zaheer *et al.* (1987) also noted the repellent properties of terpenes obtained from the needles of *Pinus roxburghii* against termites. Moreover, pine oil and oleic acid delayed and reduced attack on logs by ambrosia beetles (Nijhalt, 1980).

In this study, two formulations, A and B, of Kitz were evaluated against various life stages of *A. aegypti*. Kitz B also contained neem which is a well-established insect repellent and antifeedant.

Materials and Methods

Insects: *Aedes aegypti* larvae were hatched from eggs obtained from a laboratory colony in the insectary. The pan-reared larvae were fed with finely ground wheat germs, cereals (Nestum) and beef liver (1:1:1, dry weight). The pupae were picked and placed in plastic bowls containing water and housed in Perspex cages. The adult mosquitoes that emerged were fed on a diet of 10% sucrose supplemented with 560mg/l vitamin B-complex. 10-day old gravid females were fed to repletion on mouse blood three days prior to experimentation.

Test formulations: Both formulations of Kitz contained pine oil as the principal ingredient, with smaller amounts of oleic acid and other compounds that acted as a wetting agent. Kitz B additionally contained 5% neem oil. Each formulation was diluted with tap water to give concentrations ranging from 0.03-2.0%. The controls consisted of tap water only.

Mortality of eggs, larvae, pupae: Eggs, half-day old first and fourth instar larvae and pupae were placed in 500, 300 and 150 ml of the test solution respectively. 200 eggs and 20 juveniles were used for each Kitz concentration. All treatments and controls were replicated five times. Daily observations were made until end-point mortality was reached, as determined by the inability of each stage to develop to the adult stage.

Oviposition by females: 10 blood-fed, gravid female mosquitoes were enclosed in a Perspex cage and allowed to oviposit on a cone made of filter paper standing on a petri dish containing 40 ml of the test solution. All controls and treatments were replicated five times. The females were removed after three days and the number of eggs laid in each dish was recorded.

All rearing and experiments were carried out at 26-28°C, 70-90% RH.

Results

Total kill (LC₁₀₀) of the eggs was achieved at 1% of Kitz A and 0.2% of Kitz B (Table 1). At lower concentrations, there was no significant difference between Kitz A and Kitz B ($p > 0.005$) in their effect on egg mortality. Tables 2 and 3 compare the mortality of first and fourth

Table 1: Effects of Kitz A and Kitz B on egg mortality of *A. aegypti*.

Kitz concentration (%)	Mean % kill (S.D.)	
	Kitz A	Kitz B
1.0	100	100
0.2	79.7 (14.3)	100
0.1	29.4 (16.4)	58.9 (24.8)
0.02	10.2 (4.1)	12.2 (7.3)
Control	2.0 (3.8)	9.7 (4.3)

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