

Evaluation of two 80% DEET formulations of Bushman insect repellent against *Aedes aegypti* mosquitoes in laboratory trials.

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Introduction

The repellency of two 80% DEET (N, N-diethyl-m-toluamide) formulations of insect repellent (Bushman Heavy Duty (800g/kg Diethyltoluamide) and Bushman Plus (800g/kg Diethyltoluamide plus sunscreen) North Queensland Laboratories, Cairns, Australia) were tested against *Aedes aegypti*. This species of mosquito is the most commonly used for insect repellent testing in Australia and overseas.

Methods

The repellent trials were carried out using human volunteers under laboratory conditions of 25°C and approximately 60%RH. The trials were based on the methods described in Frances *et al.* (1993).

Cages (30cm x 30cm x 25cm), each containing approximately 100, 5-7 day old *Aedes aegypti* mosquitoes, were prepared approximately 14 hours before the trial. A cotton pad soaked in 10% sugar solution was included in each cage and removed 1 hour before the commencement of the trial.

An untreated forearm of each volunteer was first inserted into a cage and the number of landing mosquitoes over 60 seconds was recorded. A minimum of 10 landings is recommended as a requirement to establish a test subject's suitability for repellency trials.

While wearing gloves, one gram (1.0g) of the formulation was applied evenly to the forearm (between wrist and elbow). The forearm was then consecutively exposed to a cage of mosquitoes for 3 minutes (control arm exposed for 1 minute). The total number of landings (when a mosquito remains on the skin for more than 3 seconds) and total number of bites (when the probing mosquito begins to break the skin) were

recorded. Mosquitoes were knocked off the skin before blood was imbibed. Treated and untreated forearms were exposed for 3 minutes every 60 minutes until 3 bites were recorded.

The results were analysed to determine Repellent Efficacy and Complete Protection Time as described in Barnard *et al.* (2002) *J. Med Ent.* 39: 895-899.

Repellent Efficacy:

$$\%R = ((C-T)/C) \times 100$$

where:

C = (total number of mosquitoes landing on control subject in 1 minute) x 3

T = total number of mosquitoes landing on treatment subject in 3 minutes

Complete Protection Time:

CPT = mean time from repellent application until a total of three bites (probing) was recorded.

Results

The mean landing rate on untreated control forearms (prior to commencement of trial) was 45.5±7.8 mosquitoes per minute.

The trial was conducted over 15 hours with overall mean landing rate on control forearms 29.4±7.9 mosquitoes per minute (range 23.3 to 45.5 mosquitoes per minute) (Figure 1).

The mean protection time for 80% DEET Bushman Heavy Duty lotion formulation was 830 ± 20.2 minutes (range 780 – 900 minutes) with no replicate cages recording any bites up to 660 minutes. The mean repellency rate remained above 90% up to 660 minutes (Figure 1) with no mosquito landings recorded in any of the replicate cages up to 420 minutes.

The mean protection time for 80% DEET Bushman's Plus (with sunscreen) lotion formulation was 770 ± 54.8 minutes (range 540 – 900 minutes) with no replicate cages recording any bites up to 480 minutes. The mean repellency rate remained above 90% up to 600 minutes (Figure 1) but there were small numbers of landing mosquitoes (less than 1 per minute) after the first 240 minutes.

There was no significant difference in the mean protection time between the 80% DEET (830±20.2minutes) and 80% DEET+Sunscreens (770±54.8minutes) $F_{1,11}=1.27$ $P=0.286$.

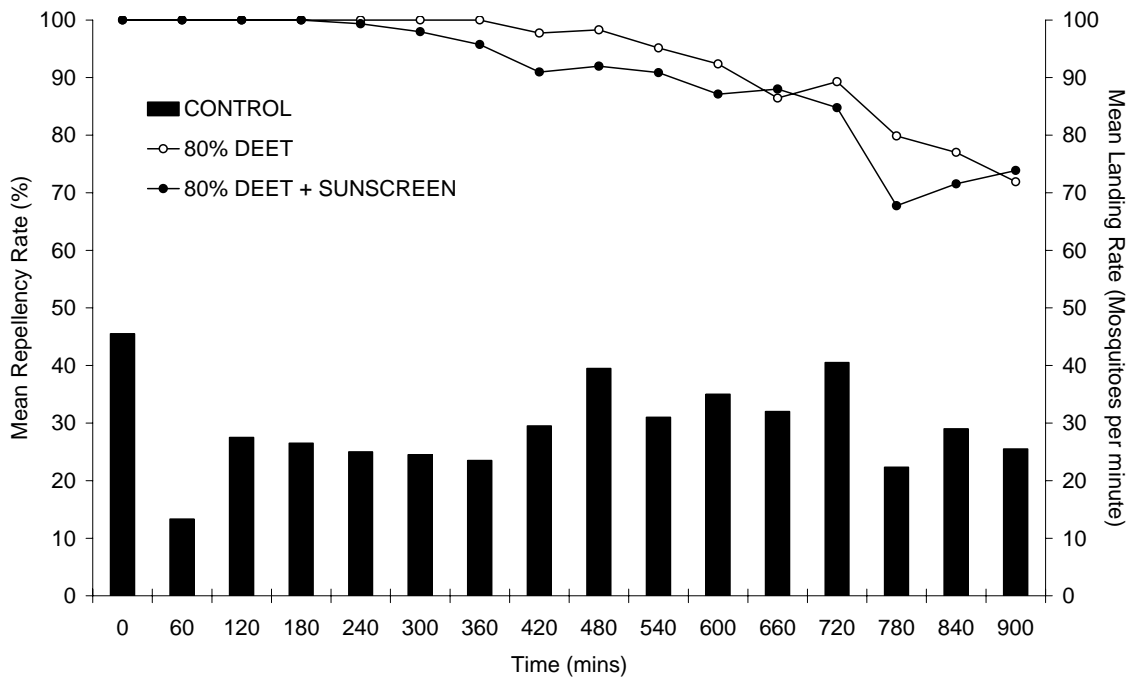


Figure 1: Mean repellency rate of two 80% DEET lotion formulations (Bushman Heavy Duty and Bushman Plus with Sunscreen) insect repellent against *Aedes aegypti* over 900minutes.

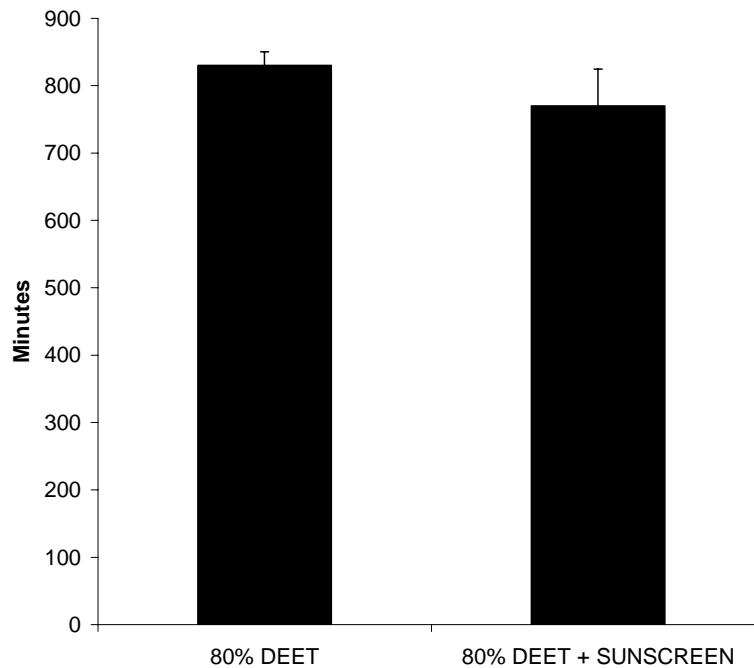


Figure 2: Mean protection time of two 80% DEET lotion formulations (Bushman Heavy Duty and Bushman Plus with Sunscreen) insect repellent against *Aedes aegypti*.

Conclusion

The results of these laboratory trials indicate that the two formulations of 80% DEET insect repellent (Bushman Heavy Duty and Bushman Plus with sunscreen) provides exceptional protection against biting *Ae. aegypti* for up to 15 hours (900minutes).

While the mean repellency rate of the 80% DEET Bushman Plus with sunscreen was slightly lower than the 80% DEET Bushman Heavy Duty repellent, there was no statistical difference in the mean protection time of the two products. The combined mean protection time for these two products was 13.3 hours (800 minutes).

As these trials were conducted under laboratory conditions, the repellents were tested against densities of mosquitoes rarely experienced under normal outdoor situations. The protection times determined in these trials are likely to be greater in situations where mosquito populations are low.

References

- Barnard, D. R., Bernier, U. R., Posey, K. H., and Xue, R. (2002) Repellency of IR3535, KBR3023, *para*-methane-3,8-diol, and Deet to Black Salt Marsh Mosquitoes (Diptera: Culicidae) in the Everglades National Park. *Journal of Medical Entomology*. 39: 895-899.
- Frances, S. P., Eikarat, N., Sripongsai, B. and Eamsila, C. (1993) Response of *Anopheles dirus* and *Aedes albopictus* to repellents in the laboratory. *Journal of the American Mosquito Control Association*. 9: 474-476.