Evaluation of Bistar 80SC (Bifenthrin) as a Tent Treatment for Protection Against Mosquitoes in Northern Territory, Australia

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J. Med. Entomol. 45(6): 1087–1091 (2008)

ABSTRACT A field trial to assess the efficacy of Bistar 80 SC as a barrier treatment of Australian military tents was conducted over 10 d at Mount Bundey Military Training Area, Northern Territory, Australia, in March 2003. Four pairs of standard eight-person tents were erected, with a single tent in each pair treated with 0.1% Bistar 80 SC as a course spray, and the remainder left as untreated control tents. Carbon dioxide-baited traps were operated in each tent nightly, and biting collections conducted over 8 nights. There was a mean increase in protection of 81% for mosquitoes entering treated tents and 90.4% increase in protection against biting of predominantly *Culex annulirostris* Skuse. In addition, bifenthrin applied to the military tents enhances the protection of occupants against bites from this important arbovirus vector.

KEY WORDS bifenthrin, Bistar 80 SC, tent treatment, Australia, *Culex annulirostris*

The protection of humans from mosquitoes by applying insecticides inside and around dwellings and community areas has been used for many years. Indoor residual spraying with DDT was used to control malaria by reducing the longevity of mosquitoes in many communities during the 1970s (WHO 1975).

Recently, various insecticides were applied to vegetation as barrier treatment against mosquitoes (Anderson et al. 1991, Perich et al. 1993). The use of insecticides, especially permethrin, has been shown to enhance the barrier effect of tents in preventing the entry and biting of mosquitoes within and around treated tents.

This method was first evaluated with the application of repellents such as *N,N*-diethyl-3-methylbenzamide (deet) on tent fabrics. Sholdt et al. (1977) showed that mosquito bites were reduced in and near tents treated with the repellent deet. The treatment of the inner walls of tents with permethrin reduced the nuisance of mosquitoes and probably invasive pests (Schreck 1991) and provided good protection against vectors of malaria (Hewitt et al. 1995).

Bifenthrin [2-methylbifenyl-c-ylmethyl (Z)-(IRS)-*cis*-3 (2-chloro-*c,c,c*-trifuroprop-1-enyl)-2,2-dimethyl cyclopropane carboxylate] is a non-α cyano pyrethroid used against a range of agricultural pests and recently as an insecticide treatment for mosquito bednets (Hougard et al. 2002). This chemical has a relatively low-irritant and knockdown effect compared with permethrin and deltamethrin. Bifenthrin causes a higher mortality by allowing mosquitoes to rest on treated surfaces for longer periods (WHO 2001).

The purpose of this study was to assess the efficacy of a bifenthrin suspension concentrate (Bistar 80 SC; FMC Chemicals, Murrarie, Queensland, Australia) treatment to military tents and their Hessian visual barrier fencing against mosquitoes at Mount Bundey Training Area, Northern Territory, Australia.

Materials and Methods

Study Site. The study was conducted at the edge of a forest at Mount Bundey Training Area, Northern Territory (131° 50′ E, 12° 52′ S), between 18 and 27 March 2003 (Fig. 1). This site was located ≈145 km east of Darwin, Northern Territory, and was primarily native woodland forest. This area was characterized by a monsoon climate with a distinct wet (November–April) and dry season (May–October). The study was timed to coincide with the occurrence of a high density of adult mosquitoes at the end of the wet season. Ethical approval was obtained from the Australian Defense Human Research Ethics Committee (ADHREC) and the study was conducted under ADHREC protocol number 306/02.

The opinions expressed herein are those of the authors and do not necessarily reflect those of the Defense Health Service (Australia) or any Defense policy. Mention of a commercial product does not constitute an endorsement of the product by the Australian Defence Force. The volunteers gave informed consent to participate in the study.

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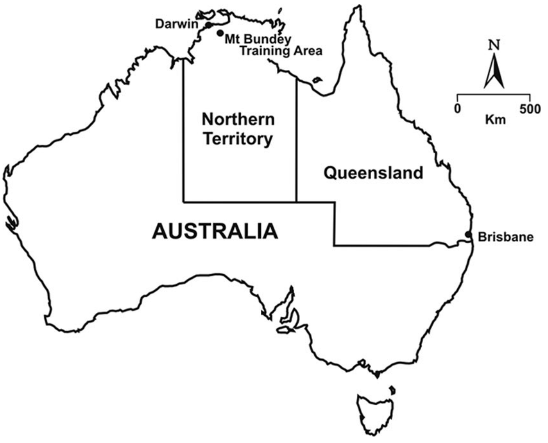

Fig. 1. Map of Australia showing location of the study site, Mount Bundey Training area, Northern Territory, Australia.

Chemical Tested. The insecticide used was Bistar 80 SC is a suspension concentrate containing 80 g/liter bifenthrin, produced by FMC Chemicals, Murrarie, Queensland, Australia. The concentrated product was diluted in water to produce a 0.1% active ingredient (AI) formulation for application.

Test Procedures. Eight standard 4.25- by 4.5-m Australian military canvas tents were erected in pairs at the edge of the Mount Bundey Training Area air strip, with each pair ≈250–300 m apart (Fig. 2a). For two pairs of tents, a barrier of Hessian fencing was erected around the perimeter of each (Fig. 2b) measuring 10- by 10-m square and 1.5 m in height. The remaining two pairs of tents were without Hession barriers (Fig. 3).

The insecticide was prepared as an emulsion in water by placing 125 ml of formulation into 10 liter of water in a Solo backpack power sprayer (Solo, Sindelfingen, Germany), which was calibrated by the Centre for Pesticide Application and Safety (University of Queensland, Gatton Campus) to deliver a dilute spray comprised of large (150–200-μm) droplets. The chemical was then sprayed onto a single tent and its Hessian fence in each pair, whereas each remaining tent and Hessian fence was left untreated. The outside and inside walls and the inside ceiling of treated tents were sprayed with the diluted formulation of Bistar 80

Fig. 2. Position of tents at the edge of airstrip and forest at Mount Bundey Training area, Northern Territory, Australia. (a) Tents without Hessian fence. (b) Tents with Hessian fence.

Fig. 3. A person applying bifenthrin to tent without Hessian fencing barrier.

SC to just before the point of runoff. The outside surface of the roof of tents was not treated. Both sides of the Hessian fences were treated until beading of the liquid formulation was observed on Hessian fibers. The treated area of the four tents and two Hessian fences was calculated to be 587 m^2.

Mosquito Collections. Each night a single encephalitis virus surveillance trap (EVS) by using a carbon dioxide bait was hung from the ceiling rafter inside each tent. The traps were placed each day for 10 d after application of insecticide.

An assessment of the effect of insecticide on the biting of mosquitoes was conducted using human biting catches. For each treatment and control tent, one person collected landing mosquitoes from one exposed lower leg and foot by using an aspirator while seated in the middle of the tent for 20 min. On the first four nights, two people collected by alternating between treated and untreated tents of each replicate pair commencing at dusk (≈1830 hours) and continuing on a 30-min cycle until all tents were sampled. On the final four nights, only one person collected from all tents.

All collected mosquitoes were returned to a field laboratory, counted, and identified using the keys of Lee et al. (1980–1989).

Statistical Analysis. Comparison of the overall number of mosquitoes biting or entering treated and untreated control tents used a two-way analysis of variance (ANOVA) on log $(x + 1)$-transformed data. The percentage of reduction in entry and biting was calculated using the following formula: total number bites/collected in control tents − total bites/collected in treated tent divided by the total in control, expressed as a percentage. The percentage of entry/biting of mosquitoes were compared using a two-way ANOVA for repeated measures. Because the data were expressed as a percentage, an arcsine transformation was conducted on the data before analysis.

Results

In total, 700 ml of the test emulsion (8% chemical concentrate) was used to treat test tents and Hessian screening, with an estimated combined area of 587 m^2. The average application rate for the trial was 96 mg/m^2.

Table 1. Mosquito species and overall number collected at Mount Bundey Training Area, Northern Territory, Australia, March 2003

Species	No. collected	
	EVS traps	Landing
Aedes alternans	1	0
Ae. lineatopennis	13	2
Ae. mallochi	9	1
Ae. normanensis	2,276	260
Ae. vigilax	22	2
Anopheles amictus	6	0
An. annulipies	5	0
An. bancroftii	23	1
An. farauti	7	0
An. hilli	1	0
An. meraukensis	76	12
An. novaguinensis	144	8
Coquillettidia xanthogaster	2,896	16
Culex annulirostris	15,698	1,990
Cx. bitaeniorhynchus	2	0
Cx. orbostiensis	45	0
Cx. sitiens subgroup	299	13
Mansonia uniformis	137	17
Verrallina reesi	6	2
Verrallina sp.	1	0
Undetermined	446	2
Total	22,113	2,326

In total, 22,113 mosquitoes from six genera and >20 species were collected over 80 trap nights in treated and untreated tents (Table 1). The predominant species collected was *Culex annulirostris* Skuse (71% of collection), with fewer numbers of *Coquillettidia xanthogaster* (Edwards) (13%) and *Aedes normanensis* (Taylor) (10%). In total, 2,326 mosquitoes from six genera and >13 species were collected by human collectors during 64 tent collections over eight nights (Table 1). *Cx. annulirostris* was also the most abundant species collected (85%), with *Ae normanensis* the next most abundant (12%). The overall mean number of mosquitoes collected each night in EVS traps was 86.3 ± 9.2 in treated tents and 467.8 ± 43.0 in untreated (control) tents. An overall mean of 5.6 ± 0.7 mosquitoes were collected nightly on humans in treated tents and 67.1 ± 7.1 mosquitoes in untreated tents. The collection of mosquitoes each night was relatively uniform, although overall more mosquitoes were collected in the last three nights of the study (Table 2).

There was no significant difference between the percentage reduction in entry of mosquitoes to tents due to the presence of Hessian ($F = 1.36$; df = 1, 20; $P = 0.26$) or collection day ($F = 1.37$; df = 9, 20; $P = 0.27$), and no interaction between day and presence or absence of Hessian ($F = 0.66$; df = 9, 20; $P = 0.73$). In addition, the percentage reduction in biting was not significantly different due to the presence of Hessian ($F = 1.72$; df = 1, 16; $P = 0.21$) and day of collection ($F = 0.84$; df = 7, 16; $P = 0.57$). Because the presence or absence of a Hessian barrier did not significantly affect the entry or biting of mosquitoes the results for all of the tents were combined and are shown in Fig.

Table 2. Mean ± SE number of mosquitoes collected nightly by EVS traps and human landing at Mount Bundey Training Area, Northern Territory, Australia, March 2003

Night after insecticide application	Bifenthrin-treated tents		Untreated (control) tents	
	EVS traps	Landing	EVS traps	Landing
1	13.0 ± 2.9a		188.5 ± 71.5a	
2	34.3 ± 5.0a,b	7.3 ± 2.3a,b	189.8 ± 40.1a	48.8 ± 9.3a
3	45.5 ± 14.9a,b	5.3 ± 1.5a,b	329.8 ± 58.8a	62.5 ± 5.0a
4	43.8 ± 5.0a,b	2.5 ± 0.7a	330.5 ± 98.4a	62.5 ± 23.5a
5	78.8 ± 11.1b,c	1.8 ± 0.8a	480.3 ± 104.4a,b	49.0 ± 11.6a
6	107.0 ± 12.6c,d		541.8 ± 72.1a,b	
7	82.5 ± 8.2b,c	4.0 ± 0.7a,b	402.8 ± 63.9a,b	48.8 ± 13.1a
8	139.3 ± 13.7d,e	5.8 ± 3.2a,b	597.3 ± 151.8a,b	69.5 ± 36.7a
9	142.3 ± 7.9d,e	8.5 ± 1.0a,b	843.0 ± 130.8b	80.3 ± 15.7a
10	176.8 ± 36.8e	10.0 ± 0.9b	774.3 ± 89.3a,b	115.3 ± 20.2a

Means in each column followed by the same letter are not significantly different using one-way ANOVA and Student–Newman–Keuls method ($P < 0.05$).

4. Significantly more mosquitoes were collected entering ($F = 429.2.$; df = 1, 60; $P < 0.001$) and biting ($F = 208.1$; df = 1, 48; $P < 0.001$) in untreated tents than in the treated tents. Overall, the mean reduction in mosquitoes entering treated tents was 81.0%, and mean reduction of mosquitoes biting within treated tents was 90.4%.

During landing collections in treated and untreated tents, some interesting differences in mosquito behavior were observed. Although the human collector was seated in treated tents waiting for mosquitoes to land, many mosquitoes were observed to enter the window and door openings, land on the tent ceiling but remain there for the remainder of the collecting period without attempting to feed. Toward the end of the 20-min collecting period, up to 20 mosquitoes were observed clinging mostly to the treated tent ceiling and walls. Those mosquitoes that were collected in treated tents flew directly through openings to the human collector without first landing on a treated tent surface. In untreated tents, mosquitoes also were observed entering tent openings with many landing on either ceiling or walls, however only for a brief period (<60 s), then flying onto the human collector to feed.

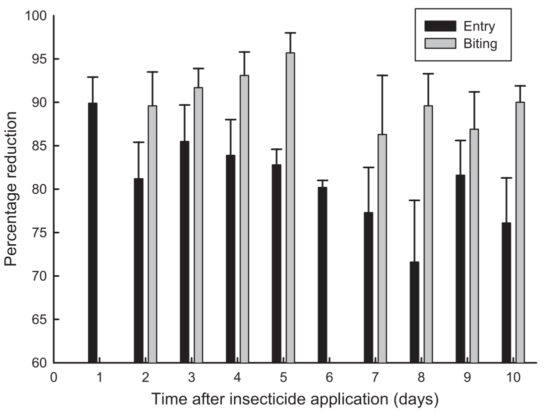

Fig. 4. Mean percentage ± SE reduction in biting and entry of mosquitoes into tents treated with Bifenthrin at Mount Bundey Training Area, Northern Territory, Australia, March, 2003.

Discussion

The purpose of the study was to assess the efficacy of the 0.1% (AI) suspension concentrate formulation of bifenthrin applied to military tents and Hessian screening to protect personnel inside the tents from mosquito bites. The study showed that there was a significant reduction in mosquitoes collected by EVS traps (81%) and significant reduction in biting (90.4%) inside treated tents over 10 d posttreatment.

For the field conditions at Mount Bundey that included relatively high mosquito numbers and application to porous materials, the recommended label application rate for Bistar 80 SC was 125 ml of concentrate diluted in 10 liters of water and applied at 1 liter of formulation per 10 m^2 of surface to be treated. The calculated concentration of chemical at this rate was 100 mg/m^2. The calculated average field application rate (96 mg/m^2) was therefore 4% lower but within the typical accuracy for hand held application equipment.

The behavior of mosquitoes entering tents treated with bifenthrin was observed in this study through the course of biting collections. In bifenthrin-treated (50 mg/m^2) bednet efficacy trials in North Cameroon, Africa, biting inhibition for *Anopheles funestus* Giles and *Anopheles gambiae* Giles s.l. of >60% was measured by Chouaibou et al. (2006) and attributed to the irritant effect of bifenthrin even if less irritating than for other pyrethroids. The striking difference in behavior between mosquitoes landing and remaining on treated tent ceilings and walls and those readily feeding in untreated tents may partly be explained by the slight pyrethroid irritation produced by bifenthrin (Chouaibou et al. 2006). However, their study reported a high proportion of mosquitoes exiting huts containing bifenthrin-treated nets, which was not the behavior observed with tents where the entire internal surface was treated. By landing directly on tent surfaces treated at the higher dose (96 mg/m^2) in the current study those mosquitoes may have been experiencing greater or more rapid loss of function including motor as well as feeding.

Culex annulirostris is an important vector of Japanese encephalitis (van den Hurk et al. 2003), Ross

River virus (Russell 2002) and Barmah Forest virus (Russell 1995) in Australia. The results in this study showed that this species readily entered military tents to obtain a bloodmeal and that the application of bifenthrin to tent fabric significantly reduced the entry and biting of this mosquito in treated tents over a 10-d period. At Wide Bay Military Training Area, Queensland, Australia, a longer term comparative study of the effects of treating military tents with permethrin and bifenthrin was conducted. That study showed that barrier tent treatments provided a reasonable increase in preventing the entry of mosquitoes (predominately *Aedes vigilax* Skuse) for at least 4 wk and that there was no significant difference in the protection provided by either bifenthrin or permethrin (Frances 2007). The use of bifenthrin as a barrier application to vegetation to protect individual occupants of houses from biting flies has been successfully evaluated against biting midges and mosquitoes in Australia (Standfast et al. 2003) and mosquitoes in the United States (Trout et al. 2007).

The current study, although only conducted over 10 d, showed that bifenthrin applied to military tents in the short term enhanced the protection of human occupants against biting *Cx. annulirostris*, an important arbovirus vector in Australia.

Acknowledgments

We thank N. W. Beebe, D.G.E. Waterson, and A. K. van den Hurk for field assistance, and FMC Chemicals (Australia) for funding assistance.

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Received 15 April 2008; accepted 18 June 2008.