OPERATIONAL NOTE

REPELLENCY OF PERMETHRIN-TREATED BATTLE-DRESS UNIFORMS DURING OPERATION TANDEM THRUST 2001

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ABSTRACT. Successful feeding by mosquitoes through permethrin-treated battle-dress uniforms (BDUs) was observed in the Shoalwater Bay Training Area (SWBTA), Queensland, Australia, during Operation Tandem Thrust 1997 (TT97). Therefore, during Operation Tandem Thrust 2001, the repellency of permethrin-treated BDUs was evaluated. Washing permethrin-treated BDUs 1 time reduced the permethrin concentration by 60%, but permethrin concentration was not reduced further after subsequent washings (up to 20 times). The probing time of *Ochlerotatus vigilax* landing on permethrin-treated BDUs was reduced to the greatest extent on the treated, unwashed uniform (7.5-fold reduction). Treated, washed uniforms also reduced probing time (2.5 times) compared to the untreated control uniform containing no permethrin. Reduced probing time should reduce feeding success and protect operational forces from bothersome mosquitoes and the associated risk of mosquitoborne disease in SWBTA. Observations of successful mosquito feeding made during TT97 could be attributed to the use of old and well-worn BDUs and seasonal differences in mosquito population density.

KEY WORDS Personal protective measures, U.S. Military, Ross River virus, Australia, Ochlerotatus vigilax

Several thousand members of the U.S. Armed Forces deploy periodically with the Australian Armed Forces to the Shoalwater Bay Training Area (SWBTA), Queensland, Australia, for Exercise Tandem Thrust. During this exercise, operational forces come in close contact with arthropods that carry disease agents, produce toxic venom, or have annoying behaviors. However, the main concern of military medical entomologists during this exercise has been the prevention of endemic arthropodborne viral diseases, specifically those caused by Ross River (RR) virus, Barmah Forest (BF) virus, and dengue virus. During Operation Tandem Thrust 1997 (TT97), there were 9 confirmed clinical cases of infection with RR virus and 1 confirmed clinical case of infection with BF virus. Ross River virus was isolated from 2 human cases and from the following 4 mosquito species, Ochlerotatus vigilax (Skuse), Oc. procax (Theobald), Verrallina funereus (Skuse), and Culex annulirostris (Skuse).

Personnel of the U.S. Armed Forces protect

themselves from biting arthropods and vector-borne disease with repellents applied to clothing and exposed skin. Clothing is treated with the pyrethroid insecticide permethrin (3-(phenoxyphenyl) methyl (\pm) -*cis*, *trans*-3-(2,2-dichloroethenyl)-2,2-dimethyl-cyclopropanecarboxylate), and exposed skin is treated with deet (*N*,*N*-diethyl-*m*-toluamide or *N*,*N*-diethyl-3-methylbenzamide). Although this is the best system developed to date for the protection of deployed personnel, results vary depending on environmental conditions (AFPMB 2002).

Permethrin binds tightly and persistently to uniform material (Schreck et al. 1978a, 1980a). At high concentrations, permethrin will kill many kinds of terrestrial arthropods, but at lower concentrations, permethrin is a contact repellent. This repellency reduces the biting rate of arthropods, such as mosquitoes, ticks, fleas, and lice (Schreck et al. 1978b, 1980b; Sholdt et al. 1989). Therefore, permethrin treatment of uniform material protects deployed military personnel from bothersome arthropods and accompanying diseases.

During TT97, military medical entomologists observed a high frequency of successful mosquito feeding through permethrin-treated battle-dress uniforms (BDUs). This led to concerns that permethrin-treated BDUs were ineffective in SWBTA and potentially could lead to a high level of transmission of mosquito-borne disease agents. Therefore, this study was designed to evaluate the effective repellency of permethrin-treated BDUs during Operation Tandem Thrust 2001 (TT01).

Five hot-weather BDUs were treated with permethrin (151 ml, 40% emulsifiable concentrate) diluted in 7.57 liters of water and applied with a 2gal hand sprayer by following the standard procedure for treating BDUs (AFPMB 2002). Each

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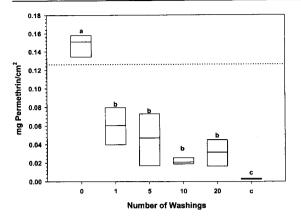


Fig. 1. Median \pm 95% CI concentration of permethrin present in treated, washed and unwashed battle-dress uniforms. The dotted line indicates the treatment target permethrin concentration of 0.125 mg/cm². The control uniform (c) was not treated with permethrin. Medians with different letters are significantly different (P < 0.05).

uniform side (front and back) was sprayed at 55 psi for 50 sec with a course fan spray and allowed to air dry. Four of the treated BDUs were washed in Tide[®] laundry detergent (Proctor and Gamble, Cincinnati, OH) in a standard domestic clothes washer (Heavy Duty Imperial[®], Whirlpool Corporation, Benton Harbor, MI) at normal cycle in cold water 1, 5, 10, or 20 times. After washing, uniforms were allowed to air dry. A 6th BDU was not treated and served as a control. Each BDU was placed into a plastic bag and kept cool and dry until used in this study.

The repellency of permethrin-treated BDUs was tested at Freshwater Camp, SWBTA. Each BDU was donned in sequential order by the same individual starting with the untreated BDU, followed by the BDUs washed 20, 10, 5, 1, and 0 times. For each uniform, the arrival and departure time of 10 individual *Oc. vigilax* landing in a square area encompassing the shoulders and extending to the lower back was recorded by a 2nd person. This study was started at 1700 h and was completed by 1800 h. After data were recorded, the back of each BDU was cut out for permethrin concentration analysis.

The concentration of permethrin from each uniform was determined by cutting 4 fabric swatches, weighing 1.2–1.9 g, from each uniform sample and placing these swatches individually in 10 ml of acetonitrile (Burdick & Jackson, Muskegon, MI) for 24 h at room temperature with occasional agitation. The acetonitrile extract was then filtered through a 0.45-µm filter and subjected to high-performance liquid chromatography analysis (1100 series isocratic pump, auto-sampler, and UV-Vis variable wavelength detector set at 237nm, Agilent Technologies Deutschland GmbH, Waldbronn, Germany; Waters Nova-Pak[®] C18 analytical column, 4 µm, 5 × 100 mm Radial-Pak[®] Cartridge, Waters

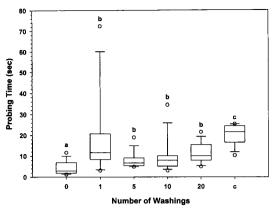


Fig. 2. Median \pm 95% CI probing time of mosquitoes exposed to permethrin-treated battle-dress uniforms. Uniforms were treated with permethrin and subsequently washed. The control uniform (c) was not treated with permethrin. Error bars represent standard deviation of the mean. Open circles represent the range of the data set.

Corp., Milford, MA). Data on probing time and concentration of permethrin-treated BDUs were analyzed by Kruskal–Wallis 1-way analysis of variance on ranks procedure, and median separations were performed by using the Student–Newman– Keuls method (SPSS 1997).

The permethrin concentration of the unwashed BDU averaged 0.147 mg/cm², with a maximum concentration of 0.159 and a minimum of 0.129 mg/cm² (Fig. 1). This was above the target concentration of 0.125 mg/cm² specified in the treatment protocols of the AFPMB (AFPMB 2002).

Washing significantly reduced the concentration of permethrin (H = 18.7; df = 5; P = 0.002; Fig. 1). The unwashed BDU contained a higher median concentration of permethrin (0.148 mg/cm²) than all other washing combinations. However, no significant differences were found in the median concentrations of BDUs washed 1, 5, 10, and 20 times (0.060, 0.046, 0.022, 0.031 mg/cm², respectively).

Probing time (time spent on the uniform) of Oc.vigilax was reduced by permethrin (H = 26.0; df = 5; P < 0.001; Fig. 2). Individuals landing on the permethrin-treated unwashed BDU had the shortest median probing time at 2.9 sec. No significant changes were found in the probing times of mosquitoes exposed to BDUs washed 1, 5, 10, or 20 times (11.7, 6.7, 7.9, and 10.1 sec, respectively). Ochlerotatus vigilax that landed on the untreated BDU had the longest probing time at 21.6 sec.

Ochlerotatus vigilax was the main mosquito species present during this study. Only 1 individual of a different species was observed, Coquillettidia xanthogaster (Edwards). A 3-night light-trap survey of Freshwater Camp showed that 1,425 (92.1%) of 1,547 mosquitoes trapped were Oc. vigilax. After a total of 9 nights of trapping in inland and coastal areas of the SWBTA, 82% of the mosquitoes trapped were *Oc. vigilax*. The only other species captured during TT01 in which RR virus was isolated was *Oc. procax* and only 2 individuals (0.11%) were identified from the 9 nights of trapping.

This study demonstrated that permethrin-treated washed and unwashed BDUs reduced the probing time of Oc. vigilax in SWBTA by 58 and 87%, respectively. Therefore, permethrin-treated BDUs will decrease the biting frequency and transmission of disease among deployed personnel exposed to this mosquito in the SWBTA. Observations of successful feeding by mosquitoes during TT97 could be attributed to several factors. Tandem Thrust 97 was held in the hot, humid summer, whereas TT01 was held in the cool, dry fall. The mosquito density was much higher and the species composition was different during TT97 than in TT01. Additionally, common problems that lower the efficacy of permethrin treatment, such as improper treatment, dry cleaning, and the use of older, well-worn BDUs, may have contributed to the perception that the permethrin treatment was not functioning properly.

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