

## Correspondence

### **Durability of protective fabrics against dengue vector *Aedes albopictus* in northeastern India**

Sir,

The environmental conditions in the northeastern region of India are highly favourable for mosquito proliferation and survival. The vectors of malaria, Japanese encephalitis and dengue are widely prevalent in the region and there is perennial and uninterrupted transmission of mosquito borne diseases such as malaria<sup>1</sup>. The reduction of man mosquito contact by adopting personal protection measures is recommended as an effective strategy for the management of vector borne diseases. These include the use of topical insect repellents, bed nets and protective clothings impregnated with mosquito repellents. The use of clothings impregnated with permethrin is an effective method of protection against both nocturnal and diurnal insects as the use of bed nets is not always possible<sup>2</sup>. The application of repellents onto clothings reduces the likelihood of allergic reactions and hence is preferable to skin application<sup>3</sup>. Protective clothings treated with permethrin at the rate of 1.25 g/m<sup>2</sup> are recommended for protection from mosquitoes and other arthropods<sup>4</sup>. Permethrin treated clothings have been found to be safe at the suggested exposure levels<sup>5</sup>.

Permethrin impregnated military fabrics have provided 57 and 87 per cent protection against mosquito bites during field trials conducted in Pakistan and Iran, respectively<sup>4,6</sup>. The clothings impregnated with permethrin were found to be helpful in reducing tick bites by 93 per cent in field trials in North Carolina, USA<sup>7</sup>. In India, laboratory and field studies have shown the efficacy of N, N-diethylphenylacetamide (DEPA) impregnated fabrics against mosquitoes<sup>8</sup>. Synthetic pyrethroid (deltamethrin and cyfluthrin) impregnated clothes worn over fabrics were found to provide protection from mosquito bites when evaluated in a military station in Tezpur, northeastern India<sup>9</sup>. A

study on the persistence of permethrin in impregnated military uniforms after repeated washings indicated that more than 50 per cent of the initial permethrin content was lost after five washings<sup>10</sup>. Apart from washings, the durability of protective fabrics is also influenced by the environmental conditions prevailing in a region. The information on the reduction of bioefficacy due to weathering is important for assessing the durability of protective clothings under field conditions. Hence, the present study on the durability of the insect protective fabrics was carried out by subjecting the treated fabrics to the hot and humid environment of northeastern India.

This study was conducted in the Defence Research Laboratory (Defence Research & Development Organization), Tezpur, Assam India. Army uniform fabrics (50% cotton and 50% polyester) obtained from a military station in northeastern India were treated at the rates of 0.5, 0.75, 1, 1.25 and 1.5 g/m<sup>2</sup> by soaking in aqueous emulsions of permethrin 25 per cent emulsifiable concentrate (EC) (Scientific Fertilisers Co. Ltd., Coimbatore, India). 60 × 60 cm pieces of the treated fabrics were pinned to a wooden board and exposed over a period of two years (June 2010 - May 2012) to the ambient weather conditions of Tezpur, northeastern India. The ranges of mean temperature and relative humidity during the study period were 17.6-30.3°C and 67.1-82.5 per cent respectively. The non-weathered fabric was sealed in a polyethylene bag after permethrin treatment and kept in a dark cabinet for two years. Permethrin residues were eluted using acetonitrile from 2 cm<sup>2</sup> pieces of the treated fabrics initially and after 1, 6 months, 1 and 2 years of weathering. The residues were detected using HPLC system (Waters Corporation, Milford, USA) with acetonitrile-water (80:20) as the mobile phase.

World Health Organization bioassay tubes and cones were used as per the guidelines<sup>11,12</sup> to determine the residual efficacy of the impregnated fabrics against dengue vector *Aedes albopictus*. Probit analysis was carried out using IBM SPSS Statistics 19 (IBM, USA) for estimating the median knockdown time (KT<sub>50</sub>). The fabrics providing >95 per cent knockdown at 1 h post-exposure or >80 per cent mortality at 24 h post-exposure were considered to be effective against mosquitoes<sup>11</sup>.

The initial permethrin content detected in the fabrics treated with permethrin emulsions at the rates of 0.5 to 1.5 g/m<sup>2</sup> were 0.48, 0.74, 1.05, 1.27 and 1.47 g/m<sup>2</sup>, respectively. The mean per cent losses of permethrin due to 1 and 6 months and 1 year of weathering were 3.91 ± 0.57, 12.1 ± 0.74 and 29.2 ± 1.99, respectively. The per cent loss of permethrin from the fabric subjected to two years of weathering was 41 ± 1.71, whereas only 8.22 ± 0.82 per cent of permethrin was lost after two years of storage. The fabric with 1.27 g/m<sup>2</sup> permethrin initially was used for the bioassays, which had 1.24, 1.12, 0.91 and 0.8 g/m<sup>2</sup> permethrin residues after 1 and 6 months, 1 and 2 years after weathering. The tested fabric had 1.16 g/m<sup>2</sup> permethrin after two years of storage (Table I).

The dengue vector, *Ae. albopictus* is a widely prevalent day biting mosquito in northeastern India. The studies using an accelerated weathering instrument have shown that the permethrin-impregnated fabrics provided good protection from the dengue vector *Ae. aegypti* and the malaria vector *Anopheles stephensi* up to 6 weeks<sup>13</sup>. It has also been indicated that the knockdown

effect of the impregnated fabrics against *Ae. aegypti* diminished rapidly as compared to the repellent effect and that *An. stephensi* was more susceptible than *Ae. aegypti*<sup>13</sup>. In the present study, the per cent mosquito knockdown 15 min after exposure (KD<sub>15</sub>) was 100 at the beginning but was reduced to 79.4 ± 3.6, 32.3 ± 2.9, 29.8 ± 3 and 26 ± 2.9 after 1 and 6 months, 1 and 2 years of weathering, respectively. There was 100 per cent mortality of mosquitoes on exposure to the fabric weathered for a year and 97.8 ± 1.3 per cent after 2 years. The treated fabric in storage provided 73 ± 2.5 knockdown and 100 per cent mortality after two years. The median knockdown time is useful for measuring the bioavailability of an insecticide on treated surface and regeneration time and hence a suitable method for the evaluation of insecticide- treated textiles<sup>14</sup>. The median knockdown time (KT<sub>50</sub> with 95% CI) on exposure to the treated fabric was 4.22 (3.24-5.13) minutes at the beginning. The KT<sub>50</sub> were 5.43 (3.18-6.94), 19.3 (17.6-21.3), 24.8 (21.9-28.1) and 28.5 (23-33) minutes, respectively after 1 and, 6 months, 1 and 2 years of weathering. However, exposure of mosquitoes to the fabric under storage resulted in KT<sub>50</sub> of 6.47 (4.15-8.14) minutes (Table I).

Contact exposure for brief periods of time more closely approximates the actual contact while the mosquitoes attempt to feed on human blood<sup>15</sup>. Hence, the mosquito mortality on short-term exposure to the treated fabrics would be useful for assessing the efficacy of the impregnated fabrics. The KD<sub>15</sub>, KD<sub>60</sub> (per cent knockdown 1 h post-exposure) and per cent mortality 24 h post-exposure in cone bioassays with

**Table I.** Mosquito knockdown and mortality on 10 min exposure to protective fabrics subjected to weathering in northeastern India

Weathering period	Per cent loss of permethrin	Permethrin residue in the tested cloth (g/m <sup>2</sup> )	Mosquito knockdown (%) <sup>#</sup>	Mosquito mortality (%)	Median knockdown time (min) (95% CI)
0 month	0	1.27	100	100	4.22 (3.24-5.13)
1 month	3.91 ± 0.57	1.24	79.4 ± 3.6	100	5.43 (3.18-6.94)
6 months	12.1 ± 0.74	1.12	32.3 ± 2.9	100	19.3 (17.6-21.3)
1 year	29.2 ± 1.99	0.91	29.8 ± 3	100	24.8 (21.9-28.1)
2 years	41 ± 1.71	0.80	26 ± 2.9	97.8 ± 1.3	28.5 (23-33)
2 years storage	8.22 ± 0.82	1.16	73 ± 2.5	100	6.47 (4.15-8.14)

<sup>#</sup>15 min after exposure to treated clothes  
Values are mean ± SEM

**Table II.** Mosquito knockdown and mortality on three minutes exposure to protective fabrics subjected to weathering in northeastern India

Duration of weathering	Mosquito knockdown (%)		Mosquito mortality (%)
	15 min post-exposure	1 h post-exposure	
0 month	49.4 ± 2.9	100	100
1 month	26.8 ± 2.1	94.6 ± 2	100
6 months	15.3 ± 2.6	80.6 ± 4.2	94.5 ± 3.3
1 year	2 ± 1.2	43 ± 2.3	79.2 ± 2.8
2 years	0	37.3 ± 2.7	67.6 ± 2.9
2 years storage	33.4 ± 2.7	100	100

Values are mean ± SEM

three minutes exposure were  $49.4 \pm 2.9$ , 100 and 100 at the beginning. The  $KD_{15}$  after 1 month, 6 months, 1 year and 2 years of weathering were  $26.8 \pm 2.1$ ,  $15.3 \pm 2.6$ ,  $2 \pm 1.2$  and 0, respectively, whereas the corresponding  $KD_{60}$  were  $94.6 \pm 2$ ,  $80.6 \pm 4.2$ ,  $43 \pm 2.3$  and  $37.3 \pm 2.7$ . The per cent mortality, which was 100 after 1 month of weathering, was reduced to  $94.5 \pm 3.3$ ,  $79.2 \pm 2.8$  and  $67.6 \pm 2.9$  after 6 months, 1 and 2 years of weathering. The non-weathered fabric after 2 years of storage provided  $KD_{15}$  of  $33.4 \pm 2.7$ , while  $KD_{60}$  and per cent mortality were 100 each (Table II).

The tested fabric weathered for a year retained only  $0.91\text{g/m}^2$  of permethrin, which was 27.2% below the recommended dose of  $1.25\text{g/m}^2$ . Three minutes exposure to this fabric resulted in <95 per cent mosquito knockdown at 1 h and <80 per cent mortality at 24 h post-exposure, which did not meet the WHO criteria for efficacy<sup>11</sup>. Hence, the fabrics may be retreated with permethrin a year after the first treatment at the recommended rate of  $1.25\text{g/m}^2$ . However, it should be noted that washings may further reduce the durability of the impregnated fabrics. Earlier studies have shown that permethrin-impregnated fabrics need to be treated again after 5 washings to retain their bioefficacy against mosquitoes<sup>10</sup>. Hence, re-impregnation of fabrics with permethrin is necessary after one year or five washings whichever is earlier. The permethrin treatment may be done during February-March well before the build-up of the vector population so as to ensure adequate protection from dengue and other disease vectors. The non-weathered fabric retained its efficacy after 2 years with complete knockdown 1 hour and complete mortality 24 h post-exposure. This indicated that the efficacy of the insect protective clothings could last beyond two years if stored properly.

The durability of permethrin impregnated clothes on exposure to weather and in storage as shown in the present study would be useful for devising a clothing impregnation schedule for disease endemic areas of northeastern India. However, there is a need to develop safer and more effective formulations for long lasting impregnation of clothings for use in areas with high density of disease vectors.

**Reji Gopalakrishnan\***, **A.K. Chaurasia**,  
**Indra Baruah & Vijay Veer**  
Defence Research Laboratory  
(Defence Research &  
Development Organisation)  
PB No. 2, Tezpur, Assam 784 001, India  
\*For correspondence:  
gknreji@gmail.com

## References

1. Dev V, Bhattacharyya PC, Talukdar R. Transmission of malaria and its control in the northeastern region of India. *J Assoc Physicians India* 2003; 51 : 1073-6.
2. Penetier C, Chabi J, Martin T, Chandre F, Rogier C, Hougard JM, et al. New protective battle-dress impregnated against mosquito vector bites. *Parasit Vectors* 2010; 3 : 81.
3. Rozendaal JA. *Vector control: methods for use by individuals and communities*. Geneva: World Health Organization; 1997.
4. Khoobdel M, Shayeghi M, Ladonni H, Rassi Y, Vatandoost H, Alipour HK. The efficacy of permethrin-treated military uniforms as a personal protection against *Culex pipiens* (Diptera: Culicidae) and its environmental consequences. *Int J Environ Sci Technol* 2005; 2 : 161-7.
5. Young GD, Evans S. Safety and efficacy of DEET and permethrin in the prevention of arthropod attack. *Mil Med* 1998; 163 : 324-30.
6. Sholdt LL, Schreck CE, Qureshi A, Mammino S, Aziz A, Iqbal M. Field bioassay of permethrin – treated uniforms and a new

- extended duration repellent against mosquitoes in Pakistan. *J Am Mosq Control Assoc* 1988; 4 : 233-6.
7. Vaughn MF, Meshnick SR. Pilot study assessing the effectiveness of long-lasting permethrin-impregnated clothing for the prevention of tick bites. *Vector Borne Zoonotic Dis* 2011; 11 : 869-75.
  8. Rao KM, Prakash S, Kumar S, Suryanarayana MV, Bhagwat MM, Gharia MM, *et al.* N,N-diethylphenylacetamide in treated fabrics as a repellent against *Aedes aegypti* and *Culex quinquefasciatus* (Diptera: Culicidae). *J Med Entomol* 1991; 28 : 142-6.
  9. Bhatnagar A, Mehta VK. Efficacy of deltamethrin and cyfluthrin impregnated cloth over uniform against mosquito bites. *Med J Armed Forces India* 2007; 63 : 120-2.
  10. Gopalakrishnan R, Chaurasia AK, Baruah I, Veer V. Evaluation of permethrin-impregnated military uniforms for contact toxicity against mosquitoes and persistence in repeated washings. *Int J Environ Sci Tech* 2013. DOI:10.1007/s13762-013-0337-6.
  11. WHO. *Guidelines for laboratory and field testing of long-lasting insecticidal mosquito nets*. WHO/ CDS/ WHOPES/ GCDPP/ 2005.11. Geneva: World Health Organization; 2005.
  12. WHO. *Guidelines for testing mosquito adulticides for indoor residual spraying and treatment of mosquito nets*. WHO/ CDS/ NTD/ WHOPES/GCDPP/ 2006.3. Geneva: World Health Organization; 2006.
  13. Gupta RK, Rutledge LC, Reifenrath WG, Gutierrez GA, Korte DW Jr. Effects of weathering on fabrics treated with permethrin for protection against mosquitoes. *J Am Mosq Control Assoc* 1989; 5 : 176-9.
  14. Skovmand O, Bonnet J, Pigeon O, Corbel V. Median knock-down time as a new method for evaluating insecticide-treated textiles for mosquito control. *Malar J* 2008; 7 : 114.
  15. Fryauff DJ, Shoukry MA, Hanafi HA, Choi YM, Kamel KE, Schreck CE. Contact toxicity of permethrin-impregnated military uniforms to *Culex pipiens* (Diptera: Culicidae) and *Phlebotomus papatasi* (Diptera: Psychododae): effects of laundering and time of exposure. *J Am Mosq Control Assoc* 1996; 12 : 84-90.