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COMPARATIVE EFFICACY AND PERSISTENCY OF PERMETHRIN IN OLYSET® NET AND CONVENTIONALLY TREATED NET AGAINST *AEDES AEGYPTI* AND *ANOPHELES STEPHENSI*

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ABSTRACT: The efficacy of the Olyset® net was compared to a net treated conventionally with permethrin 10% emulsifiable concentrate at the World Health Organization recommended dose. The nets were assessed under laboratory conditions against the mosquito vectors *Aedes aegypti* and *Anopheles stephensi*. Knockdown efficacy for the Olyset net was assessed after the first 10 washings and then after the 15th and 20th washings. Data were not collected for the conventionally treated netting after 5 washings because the conventional treatment was no longer effective. The results of the study indicate permethrin effectively persisted on Olyset net for at least 20 washings, confirming the regeneration of pesticide after each wash.

KEY WORDS Olyset net, mosquito, permethrin, washing, persistency

INTRODUCTION

Use of insecticide-treated materials is gaining importance in mosquito control. Pyrethroids are preferred for insecticide-treated materials because of their low mammalian toxicity and rapid insecticidal and excitorepellent impact on mosquitoes. The Olyset® net is a permethrin-impregnated net with a reputation for a long-lasting efficacy, produced by M/s Sumitomo Chemical India Pvt. Ltd., Mumbai, India. The Olyset net was evaluated against the malaria vector *Anopheles stephensi*, Liston, and the yellow fever vector *Aedes aegypti* L., to compare its efficacy to a bed-net conventionally treated with permethrin 10% emulsifiable concentrate (EC).

MATERIALS AND METHODS

Bed-nets were evaluated against *An. stephensi*, a principal vector of urban malaria, and *Ae. aegypti*, a vector for yellow fever. Adults from colonies maintained in the laboratory at a temperature of 24 ± 2°C and 50–70% relative humidity were used for the study.

Market-available nylon bed-netting (325 holes/in.^{2}) was purchased and a portion (1 × 1 m) was cut from the netting and assessed for its absorptive power in water (45 ml) and impregnated with permethrin 10% EC at 5 ml/45 ml to achieve 500 mg active ingredient/m^2 (WHO 1997) under laboratory conditions. As in Hougard et al. (2003), the dipped net portions were carefully handled to ensure an even distribution of the solution and then shade dried horizontally. After drying, the sample was folded and kept at room temperature of 24 ± 2°C and 50–70% humidity for a period of 7 days. Tests began 7 days after impregnation. The same dimension was cut from the Olyset net supplied by M/s Sumitomo Chemical India and tested without modification.

Similar methods were followed for both mosquito vectors and net treatments. Standard WHO plastic cones were fixed on a net sample and 1 to 2 day-old unfed female mosquitoes at 10 insects/cone were released by using an aspirator. Experiments were replicated and 4 cones were run simultaneously for each replication. After an average exposure of 3 min, the mosquitoes were collected in a holding tube and fed with a 10% sugar solution. They were observed for 1 h for knockdown and then for 24 h for mortality. Mosquitoes with broken legs but able to fly were considered alive. Similar exposures were given to the mosquitoes tested on untreated nylon nets from the market, which served as controls.

The 50% knockdown time (KT_{50}) was assessed by using the probit analysis method of Finney (1971) by the using NCSS 2000 package (supplied by NCSS, Kaysville, Utah, USA). Knockdown efficacy for the Olyset net was assessed after the first 10 washings and then after the 15th and 20th washings. Mortality efficacy for both net treatments was assessed for only the first 5 washings because the netting with conventional treatment was no longer effective.

Washing procedure. Washing was done by using a locally available detergent, Rin® (Hindustan Lever LTD., Mumbai, Maharashtra, India) at 0.5 g/liter of water in a washing machine (Whirlpool Whitemagic Super, semiautomatic, Whirlpool India LTD., Pondycherry, India) at 24-h intervals. The washing procedure consisted of 3 min of washing, 2 rinsing cycles each of 2-min duration, and a 1-min spinning cycle.

After washing, the net samples were dried under sunlight for 3–4 h and assessed for efficacy by cone bioassay. During the drying period, the temperature was found to range between 26.21 and 39.28°C.

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Table 1. Knockdown rate of permethrin-treated bed-net (Olyset® net) against mosquito species.[1]

Washings	Mosquito species	KT_{50} (min)	95% CI (min)	Regression equation
0	*Ae. aegypti*	<1	—[2]	$y = 6.0239 + 0.8112 \log x$
	An. stephensi	2.26	1.72–2.80	$y = 4.2886 + 2.0128 \log x$
1	*Ae. aegypti*	<1	—	$y = 5.2272 + 1.5176 \log x$
	An. stephensi	6.25	5.79–6.71	$y = 2.3929 + 3.2767 \log x$
2	*Ae. aegypti*	<1	—	$y = 7.2064 + 0.1924 \log x$
	An. stephensi	4.68	4.18–5.08	$y = 3.1504 + 2.7606 \log x$
3	*Ae. aegypti*	<1	—	$y = 5.8549 + 1.0827 \log x$
	An. stephensi	4.16	3.62–4.72	$y = 3.5840 + 2.2856 \log x$
4	*Ae. aegypti*	<1	—	$y = 7.2064 + 0.1924 \log x$
	An. stephensi	<1	—	$y = 5.4283 + 0.8731 \log x$
5	*Ae. aegypti*	<1	—	$y = 5.9301 + 0.9236 \log x$
	An. stephensi	1.55	1.02–2.08	$y = 4.7039 + 1.5610 \log x$
6	*Ae. aegypti*	<1	—	$y = 6.4699 + 0.6284 \log x$
	An. stephensi	1.88	1.32–2.44	$y = 4.5575 + 1.6057 \log x$
7	*Ae. aegypti*	<1	—	$y = 6.6263 + 0.5394 \log x$
	An. stephensi	1.63	1.09–2.17	$y = 4.6727 + 1.5400 \log x$
8	*Ae. aegypti*	<1	—	$y = 6.3943 + 0.6588 \log x$
	An. stephensi	1.52	0.98–2.06	$y = 4.7333 + 1.4620 \log x$
9	*Ae. aegypti*	<1	—	$y = 6.0711 + 0.9105 \log x$
	An. stephensi	1.25	0.71–1.79	$y = 4.8769 + 1.2367 \log x$
10	*Ae. aegypti*	<1	—	$y = 5.9209 + 0.9433 \log x$
	An. stephensi	1.39	0.84–1.94	$y = 4.8134 + 1.2854 \log x$
15	*Ae. aegypti*	2.63	1.99–3.27	$y = 4.3548 + 1.5355 \log x$
	An. stephensi	3.26	2.34–4.18	$y + 4.4434 + 1.0832 \log x$
20	*Ae. aegypti*	2.45	1.52–3.38	$y = 4.6469 + 0.9085 \log x$
	An. stephensi	11.07	9.78–12.36	$y = 3.6669 + 1.2767 \log x$

[1] KT_{50}, time required to knockdown 50% of the test population after exposure; *Ae.*, *Aedes*; *An.*, *Anopheles*; *y*, probit knockdown; *x*, time
[2] —, values more than 60 min

RESULTS

The Olyset net exhibited excellent knockdown and mortality. Table 1 presents the knockdown results for the Olyset net. The KT_{50} values ranged from <1 to 11.07 min for *An. stephensi* and <1 to 2.63 min for *Ae. aegypti*. Table 2 presents the knockdown results for the conventionally treated bed-net. The KT_{50} values ranged from <1 to >60 min for *Ae. aegypti* and 6.41 to >60 min for *An. stephensi*. Figure 1 presents the mortality results for both net treatments against *An. stephensi*. Figure 2 presents the mortality results for both net treatments against *Ae. aegypti*. Mortality percentages against both species remained at 100% for the Olyset net, whereas those for the conventionally treated net declined substantially after each washing.

DISCUSSION

Olyset is a new concept for long-lasting insecticide-treated nets. The insecticide is incorporated into the polyethylene polymer before yarn extrusion. The insecticide is then slowly released from the polymer, allowing the availability of insecticide

Table 2. Knockdown rate of conventionally permethrin-treated bed-net against mosquito species.[1]

Washings	Mosquito species	KT_{50} (min)	95% CI (min)	Regression equation
0	*Ae. aegypti*	<1	—[2]	$y = 6.2676 + 0.8108 \log x$
	An. stephensi	6.41	5.96–6.86	$y = 2.2556 + 3.4027 \log x$
1	*Ae. aegypti*	23.39	22.46–24.32	$y = 1.1789 + 2.7912 \log x$
	An. stephensi	30.42	29.39–31.45	$y = 0.3196 + 3.1556 \log x$
2	*Ae. aegypti*	57.36	53.65–61.07	$y = 0.8281 + 2.3722 \log x$
	An. stephensi	>60	—	$y = -0.6045 + 2.8643 \log x$
3	*Ae. aegypti*	>60	—	$y = -0.4227 + 2.5989 \log x$
	An. stephensi	>60	—	$y = 0.4748 + 1.7411 \log x$
4	*Ae. aegypti*	>60	—	$y = 0.5194 + 1.6166 \log x$
	An. stephensi	>60	—	$y = 1.8137 + 0.5753 \log x$
5	*Ae. aegypti*	>60	—	$y = 0.6476 + 1.6202 \log x$
	An. stephensi	>60	—	$y = 2.3731 + 0.1056 \log x$

[1] KT_{50}, time required to knockdown 50% of the population after exposure; *Ae.*, *Aedes*; *An.*, *Anopheles*; *y*, probit knockdown; *x*, time
[2] —, values more than 60 min

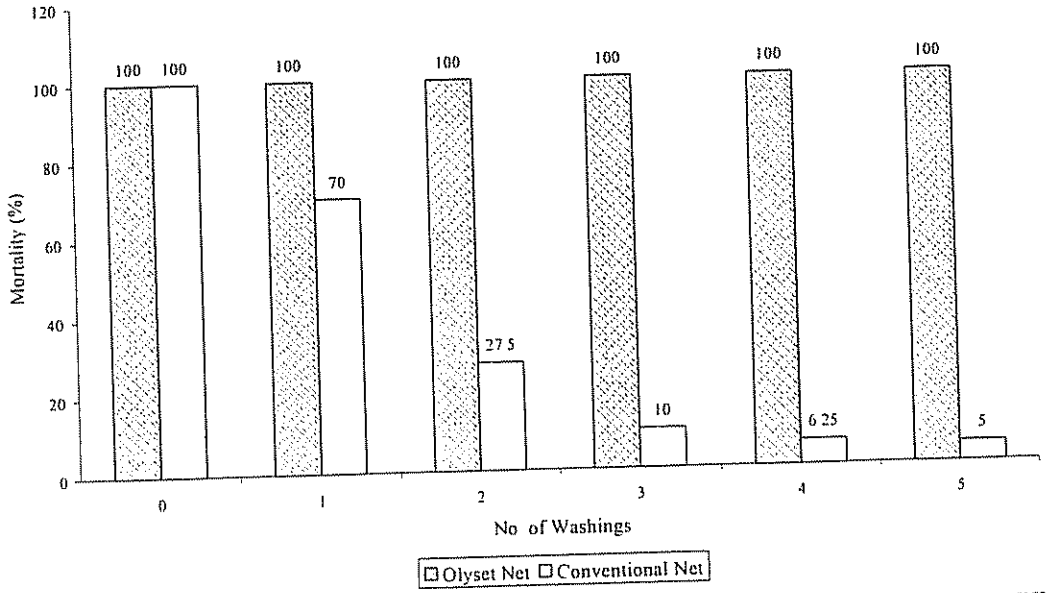

Fig 1 Mortality of *Anopheles stephensi* when exposed to Olyset® net and conventionally treated net at different washings.

at the surface (WHO 2001) after washing and subsequent drying under sunlight.

There is converging evidence that after 3 or 4 washes, insecticide is removed from the nets treated by conventional dipping (WHO 2003). In the present study, efficacy from the netting treated conventionally declined substantially after each washing. This implies that there is no regeneration of permethrin, unlike Olyset net. Previously, the Olyset net was found to lose 76% of permethrin when washed once and regained 82% when exposed to strong sunlight (WHO 2001). The results of the present study support the concept of regeneration of permethrin on the Olyset net and confirm it is better than the net treated conventionally in exhibiting long-lasting effectiveness after subsequent washes.

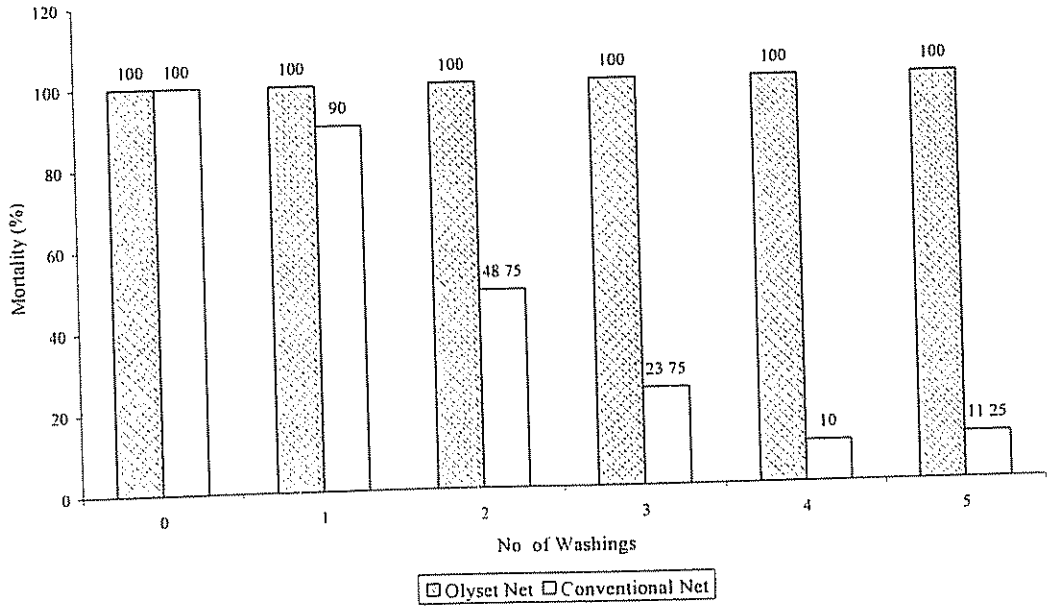

Fig 2 Mortality of *Aedes aegypti* when exposed to Olyset® net and conventionally treated net at different washings.

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