LABORATORY EVALUATION OF EFFICACY OF BEDNETS IMPREGNATED WITH PYRETHROIDS[1]

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ABSTRACT. The half-life of an insecticide on a treated object can be used as a crucial test for evaluating the residual effect. On cotton and nylon bednets, the half-life of deltamethrin was 65.6 and 55.4 days, whereas the half-life of permethrin on cotton and nylon bednets was 35.0 and 27.4 days. A deltamethrin-impregnated bednet (10 mg/m^2) is effective for 6–7 months and costs less than DDT residual spraying (2g/m^2) on the inner walls of rooms.

INTRODUCTION

Successful trials with permethrin-impregnated clothing against blood-sucking arthropods led to the expansion of the program to impregnate bednets for reducing the human-mosquito contact to prevent the transmission of mosquito-borne diseases. In China many field trials are now under way using deltamethrin- or permethrin-impregnated bednets to reduce malaria. These may replace DDT residual spraying in malaria control programs (Curtis et al. 1989, Rozendaal and Curtis 1989).

This laboratory study evaluated the insecticidal and repellent effects of permethrin and deltamethrin in different fabrics to 4 important mosquito vectors, to find a suitable dosage and to compare the cost-effectiveness of each. Also, a mathematical method is described to predict the residual effectiveness of insecticides on the treated bednets.

MATERIALS AND METHODS

Chemicals used: Deltamethrin, pure powder (99.9%) for bioassays, and K-Othrine (2.5% deltamethrin wp) for impregnating bednets obtained from Roussel-Uclaf Co., and permethrin, 80% technical grade, product of San Ling Co., Shanghai, China, were used.

Netting used: 64–81 mesh/cm^2 of nylon and white cotton netting, thickness of thread is 0.25 mm, was manufactured by the Liuzhou Bednet Factory.

Mosquito species tested: The following 4 species susceptible to pyrethroids, *Anopheles sinensis* Wied., *An. minimus* Theobald, *An. anthropophagus* Xu and Feng and *Culex quinquefasciatus* Say, were studied. All mosquitoes were reared and tested in an insectarium at 26°C and 80% RH.

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Degradation of pyrethroids on nylon and cotton bednets: Theory—The longevity of effectiveness of an insecticide on a treated object is important, and usually it is determined by window trap techniques in the field. We recently developed a new method to determine the half-life of insecticides on treated objects. The degradation of an insecticide on a treated surface corresponds with the first order reaction of chemical dynamics (Lewis and Cowsar 1977), that is:

$$Q_t = Q_o \times e^{-kt} \quad (1)$$

where Q_t = residual amount of insecticide after time t on a treated object, Q_o = beginning dosage, k = factor of first order reaction of chemical dynamics, t = time and $t_{0.5}$ = half-life.

The relation between k and $t_{0.5}$ of insecticides is:

$$k - \ln 2/t_{0.5} = 0.693/t_{0.5} \quad (2)$$

from equation (1), (2) we can get:

$$\log Q_t = \log Q_o - (0.3009/t_{0.5}) \times t \quad (3)$$

$$t_{0.5} = 0.3009/(\log Q_o - \log Q_t) \quad (4)$$

Let the beginning dosage (Q_o) be equal to 100% and the residual amount after t time (Q_t) is $Q_t\%$ = $(Q_t/Q_o) \times 100$, then (3) and (4) is changed to:

and $$\log Q_t = 2 - (0.3009/t_{0.5}) \times t \quad (5)$$

$$t_{0.5} = (0.3009 \times t)/(2 - \log Q_t\%) \quad (6)$$

Procedure for determining the half-life of pyrethroids on an impregnated bednet: One piece of bednet fabric that had been impregnated with an exact dosage of pyrethroid was extracted thoroughly by acetone; the extract was reduced to 2 ml for analysis. Second instar larvae of *Culex quinquefasciatus* were used for determination of the residual amount by a bioassay method (Wu 1987, Rang and Wu 1983); values for $Q_t\%$ and Q_o were substituted into equation (6), and the value of $t_{0.5}$ was calculated. To avoid the error of a single determination, 5–6 replications of different t and $Q_t\%$ were done, and these values were plotted on semi-logarithm coordinates, Q_t% (in logarithm) along y-axis and t (natural number) along x-axis, respectively, making a straight line. From this line the $t_{0.5}$ can be calculated. Alternatively, the exact value of $t_{0.5}$ and its 95% fiducial limit can be calculated from the regression line. The reproducibility and precision of this bioassay method were very good when single instar larvae were used (Rang and Wu 1983).

Contact toxicity of bednets impregnated with different dosages of a pyrethroid: Pieces of bednet netting (16 × 15 cm) were soaked in a concentrated solution of pyrethroids and were dried by laying them flat on a nonabsorbent surface. Fifty 3- to 7-day-old female mosquitoes fed on 10% sugar solution were anaesthetized with CO_2 and placed in the test chamber lined with the treated netting. Mortality was recorded over time to calculate the KT_{50} and KT_{95} value by probit analysis (Finney 1952).

Behavior of mosquitoes to bednets impregnated with pyrethroids: A guinea pig with its back shaved and protected by a thick cloth to prevent unnecessary mosquito bites was restrained. A piece of pyrethroid-treated bednet was placed over the shaved back. The animal was then placed in a cage (45 cm^3) with hungry female mosquitoes overnight. Blood feeding and mortality were recorded after 24 h.

Tensile strength test: The pyrethroid-impregnated bednets were hung over a bed in a room for 12 months and were then sent to The Nanning Institute of Textiles to check for their tensile strength according to International Standard Organization standard no. ISO-5081-77.

RESULTS

The residual amounts of pyrethroids on cotton and nylon fabrics over time are shown in Table 1, the half-life of deltamethrin in cotton and nylon bednets was 65.6 and 55.4 days, whereas the half-life of permethrin on cotton and nylon bednets was 35.0 and 27.4 days, respectively. The chi-square test showed that the rate of loss of the pyrethroids was very much like the first order reaction of chemical dynamics.

Toxicities of the pyrethroids to 4 species of mosquitoes (KT_{50} and KT_{95}) are shown in Tables 2 and 3. It was found that the toxic action of pyrethroids to mosquitoes varied with the type of fabric used. The nylon bednet impregnated with a pyrethroid had a faster toxic effect than a cotton bednet, particularly with permethrin. Deltamethrin was more toxic than permethrin at the same dosage levels.

Table 1. Half-life of pyrethroids on different bednet fabrics.

Insecticide	Material of bednet	% residue remaining after indicated days						Equation	Half-life	
		0	2	8	14	43	57	log y = a − bx	(days)	χ^2
Permethrin	Cotton	100.0	88.7	86.0	78.0	68.0	22.0	log y = 2.0095 − 0.0089x	35.0	0.0424*
	Nylon	100.0	93.3	87.0	67.3	46.4	18.0	log y = 2.0121 − 0.0114x	27.4	0.0229*
Deltamethrin	Cotton	100.0	99.8	74.8	87.5	65.0	53.4	log y = 1.9816 − 0.0043x	65.6	0.0040*
	Nylon	100.0	88.0	90.5	75.7	59.0	48.0	log y = 1.9769 − 0.0050x	55.4	0.0012*

* Data fit theory; there are no significant differences $P > 0.99$.

Table 2. KT_{50} values (minutes) of pyrethroids to 4 species of mosquitoes.

Species	Insecticide	Material of bednet	Dosage (mg/m^2) 0.005	0.05	0.5	5.0	10.0	20.0	50.0	100.0
An. minimus	Permethrin	Nylon	18.7	14.1	11.4	3.9	—	—	—	—
		Cotton	29.2	28.4	17.3	—	11.0	—	—	—
	Deltamethrin	Nylon	—	17.6	9.6	—	7.0	—	—	—
		Cotton	21.3	16.6	13.5	—	6.6	—	—	—
An. sinensis	Permethrin	Nylon	—	32.5	22.7	8.7	7.7	—	—	—
		Cotton	—	63.5	44.3	32.2	23.6	—	—	—
	Deltamethrin	Nylon	—	34.3	17.5	10.5	7.9	—	—	—
		Cotton	—	40.1	20.0	8.4	6.9	—	—	—
An. anthropophagus	Permethrin	Nylon	—	57.7	21.5	11.8	9.0	—	—	—
		Cotton	—	52.9	25.7	17.8	14.2	—	—	—
	Deltamethrin	Nylon	—	33.4	15.9	14.5	10.4	—	—	—
		Cotton	—	23.5	13.7	9.0	5.9	—	—	—
Cx. quinquefasciatus	Permethrin	Nylon	—	—	—	—	23.0	15.1	11.9	7.7
		Cotton	—	—	—	—	75.2	32.4	16.6	13.9
	Deltamethrin	Nylon	—	44.5	25.9	9.7	8.8	—	—	—
		Cotton	—	41.6	22.0	8.0	8.8	—	—	—

Table 3. KT_{95} values (minutes) of pyrethroids to 4 species of mosquitoes.

Species	Insecticides	Material of bednet	Dosage (mg/m^2) 0.005	0.05	0.5	5.0	10.0	20.0	50.0	100.0
An. minimus	Permethrin	Nylon	40.8	37.5	24.8	7.1	—	—	—	—
		Cotton	46.0	47.5	31.7	—	16.8	—	—	—
	Deltamethrin	Nylon	—	29.9	20.0	—	16.8	—	—	—
		Cotton	35.0	32.3	29.6	—	12.3	—	—	—
An. sinensis	Permethrin	Nylon	—	81.0	52.5	14.2	15.2	—	—	—
		Cotton	—	117.6	78.4	54.8	47.9	—	—	—
	Deltamethrin	Nylon	—	71.2	37.6	30.4	17.1	—	—	—
		Cotton	—	63.4	47.5	14.8	10.8	—	—	—
An. anthropophagus	Permethrin	Nylon	—	94.6	48.6	18.6	21.2	—	—	—
		Cotton	—	117.3	66.8	25.5	27.2	—	—	—
	Deltamethrin	Nylon	—	66.5	30.8	27.8	15.7	—	—	—
		Cotton	—	70.0	31.6	17.1	12.2	—	—	—
Cx. quinquefasciatus	Permethrin	Nylon	—	—	—	—	38.6	22.0	14.2	11.3
		Cotton	—	—	—	—	130.5	49.6	26.0	20.0
	Deltamethrin	Nylon	—	83.2	41.9	15.8	17.5	—	—	—
		Cotton	—	52.4	45.0	10.6	20.1	—	—	—

Table 4. Repellent effect of bednets treated by pyrethroids.

Insecticide	Dosage mg/m²	Species	No. of expts.	No. of mosquitoes	Blood feeding rate %	% mortality	
						Fed	Unfed
Deltamethrin	5.0	*Cx. quinq.**	1	268	0	0	70.5
	5.0	*Cx. quinq.*	2	260	4.6	0	68.1
	0.5	*Cx. quinq.*	1	166	3.6	0	0
	0.1	*Cx. quinq.*	1	244	0	0	0
	0.1	*Cx. quinq.*	2	312	7.1	4.5	0
	0.1	*An. sin.***	1	125	4.8	33.5	3.4
Permethrin	5.0	*Cx. quinq.*	1	214	0	0	0
	5.0	*Cx. quinq.*	2	78	0	0	0
	5.0	*Cx. quinq.*	3	393	3.8	0	0
	5.0	*An. sin.*	1	170	4.1	14.3	2.5
	1.0	*Cx. quinq.*	1	217	43.3	0	0
Untreated	0	*Cx. quinq.*	1	210	91.9	0	0
	0	*Cx. quinq.*	2	164	93.3	0	0
	0	*Cx. quinq.*	3	216	90.7	0	0
	0	*An. sin.*	1	85	18.8	0	0

* *Culex quinquefasciatus.*
** *Anopheles sinensis.*

Table 5. Breaking strength of bednet cloth impregnated with pyrethroids (tested 1 year after impregnation).

Insecticide and dosage	Type of fabric	Samples tested	Breaking strength* $\bar{x} \pm$ SD	*t*-value
Deltamethrin	Cotton	10	164.7 ± 14.0	1.57 NS**
50 mg/m²	Nylon	11	111.6 ± 16.6	1.88 NS
Permethrin	Cotton	10	152.3 ± 16.4	0.44 NS
400 mg/m²	Nylon	11	109.1 ± 10.5	2.14 NS
Untreated	Cotton	8	146.5 ± 7.3	
	Nylon	10	89.3 ± 7.7	

* Newton/5 × 20 cm.
** Not significant when compared with untreated fabric.

Table 6. Cost-effectiveness comparison between residual spraying and bednet impregnation.

Method	No. of treatments*	Cost (yuan)		Ratio
		Chemical	Labor	
DDT residual spraying (2 g/m²)	2	4.20	0.60	1
Deltamethrin impregnation				
(10 mg/m²)	1	0.72	—	0.15
(5 mg/m²)	1	0.36	—	0.08
Permethrin impregnation				
(800 mg/m²)	1	12.60	—	2.63
(400 mg/m²)	1	6.30	—	1.31

* For a 150-day protective period. It was estimated that each home had an inner wall surface of 150 m² to be sprayed or 2.5 bednets to be impregnated.

Repellent effects of impregnated bednets on mosquito landings were observed even when the dosage was as low as 1/100 of the treatment dosage (deltamethrin = 0.1 mg/m²) (Table 4). There were no corrosive effects on the bednet fabric at a dosage of 50 mg/m², which was 5 times the recommended dosage 1 year after impregnation (Table 5).

DISCUSSION

When 20 min is set as a critical KT_{50} value for the 4 species of mosquitoes, then from Table 2 the dosage for impregnating bednets would be 0.5 mg/m² for deltamethrin and 20 mg/m² for permethrin. For example, to calculate protective time for 150 days (t), substitute the above values into equation (3) and as a result, the impregnating dosage for deltamethrin would be 2.44 mg/m^2 whereas for permethrin it would be 389.25 mg/m^2. Generally, the recommended dosage for permethrin ranges from 200 to 1,000 mg/m^2 and for deltamethrin from 15 to 25 mg/m^2 (Rozendaal and Curtis 1989). To assure effectiveness, the theoretical dosage should be doubled; therefore, the dosage of permethrin would be 800 mg/m^2 compared with deltamethrin (5 mg/m^2). These numbers are in close proximity to the recommended dosage that was obtained from the field trials.

The cost-effectiveness was calculated and compared with DDT residual spraying at a dosage of 2 g/m^2 on the inner wall of the room as shown in Table 6. DDT residual spraying was a classical method of mosquito control for malaria eradication, but it is only suitable for areas where the vector has not yet developed resistance to DDT. DDT has almost lost its toxic effect against *Anopheles sinensis*. In 1987 (Wu, unpublished data), it was found in some rural areas of Guangxi and Hubei provinces that the mortality of *An. sinensis* was 20–40% after forced contact for 1 h when the surface was treated with DDT at 2 g/m^2. However, the KT_{95} values of 4 important vector mosquitoes were only 30–50 min when they contacted deltamethrin-treated bednets at a dosage of 0.5 mg/m^2. If the impregnating dosage of deltamethrin is increased to 5 mg/m^2 (double the theoretical dosage), the protective time will be longer, and the impregnation can be easily done at one-fifth the cost of DDT residual spraying.

Besides the toxic action, the pyrethroid-impregnated bednets provided protection without causing mortality, thus exhibiting repellent effects against resting mosquitoes on the bednet during daytime and at night. This is particularly important in rural areas because the mosquitoes that are not killed will be repelled by the bednet.

Usually the bednets are washed once a year, so impregnating the bednets just after washing would be an ideal time. It should be made sure that the dosage is no more than 10 mg/m^2. This dosage is enough to provide a 6- to 7-month protective period without causing any damage to the fabric of the bednet. The cost of impregnating with permethrin at a dosage of 400 mg/m^2 will be 1.3 times the cost of DDT residual spraying.

In conclusion, the half-life of deltamethrin and permethrin on the cotton and nylon fabric was 65.6, 55.4 and 35.0, 27.4 days, respectively. Bednets impregnated with deltamethrin at a dosage of 5–10 mg/m^2 provided 5–6 months' protection against resting mosquitoes on the net during the day and preventing mosquito bites through the net at night. It is cheaper than DDT residual spraying on the inside walls of the house, easy to undertake without causing any damage to the fabric, and is acceptable.

REFERENCES CITED

Curtis, C. F., J. D. Lines et al. 1989. Impregnated bednets and curtains against malaria mosquitoes. pp. 5–47. *In:* C. F. Curtis (ed.). Appropriate technology for vector control. CRC Press Inc., Boca Raton, FL.

Finney, D. J. 1952. Probit analysis. 2nd ed. Cambridge Univ. Press.

Lewis, D. H. and D. R. Cowsar. 1977. Principles of controlled release pesticides. pp. 1–16. *In:* H. B. Scher (ed.). Controlled release of pesticides. ACS Symposium Series 53, American Chemical Society Washington, DC.

Rang, T. Q. and Wu Neng. 1983. Observation on the quality of water effecting the results of bioassay. (In Chinese). Insect Knowledge 1983(6):266–267.

Rozendaal, J. A. and C. F. Curtis. 1989. Recent research on impregnated mosquito nets. J. Am. Mosq. Control Assoc. 5:500–507.

Wu Neng. 1987. The relation between dosage and reaction time in bioassay of insecticides. (In Chinese). Insect Knowledge 1987(4):234–237.