The effects of different drying methods and sun exposure on the concentrations of deltamethrin in nets treated with K-O Tab® tablets

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The method used to dry bednets after they have been treated with an insecticide solution may affect the levels of insecticide and the uniformity of the insecticide deposits on the dry nets. In an attempt to see how the drying method may affect the insecticide deposits on the dry net, and to select the best drying method, laboratory and field studies have recently been carried out (in the U.K. and Iran, respectively). Conventional polyester nets were each treated with a deltamethrin solution (made with one K-O Tab® tablet in 500 ml water) and then dried, either while hanging vertically or laid horizontally on the floor, in the sun or shade. The concentrations of deltamethrin in 25-cm^2 samples cut from the dry nets (from the inner folds, surface folds, and top and bottom of each net dried vertically, and from the upper and lower surfaces of each net dried horizontally) were then determined using high-performance liquid chromatography (HPLC).

Drying the treated nets in the sun or the shade did not make a significant difference to deltamethrin concentrations on the nets. Mean deltamethrin concentrations were, however, higher on the lower parts of the nets that had been hung to dry vertically than on the upper parts of these nets, and greater on the upper surfaces of nets dried horizontally than on the lower surfaces of such nets. In general, the layers and folds of the nets that had been on the outside of the drying nets contained more deltamethrin than the inner folds. These patterns are consistent with the hypothesis that deltamethrin tends to accumulate at the points where the water from the insecticide solution evaporates from the drying nets and, also in the case of nets dried vertically, at the lower points of the drying nets (as the result of gravity). In order to obtain an even and adequate distribution of insecticide, it is therefore not necessary to dry the net in the shade. To achieve a uniform deposit of deltamethrin, the drying net should be folded as little as possible and dried quickly.

Instructions on how mosquito nets should be treated with an insecticide generally

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indicate that, after being dipped in a dilute solution of an insecticide, the nets should be dried flat (in order to ensure an even deposit) and in the shade (in order to prevent photo-degradation). There is, however, little evidence to support these common recommendations. Research on net-treatment methods has focused on methods of dipping and washing, with relatively little attention paid to the procedures to be followed for drying (Snow *et al.*, 1988; Lindsay *et al.*, 1991; Miller *et al.*, 1991; Pleass *et al.*, 1993; Curtis *et al.*, 1996; Graham *et al.*, 2005; Kayedi *et al.*, 2007*a*, *b*, 2008).

Most manufacturers of insecticides and bednets advise their customers to dry nets in the shade after insecticide treatment. After trials in The Gambia, Snow *et al.* (1988) recommended that, to prevent breakdown of the insecticide on the nets by ultra-violet light, bednets should be dried in the shade after impregnation with permethrin. Similarly, Miller *et al.* (1991) suggested that, after each wash , insecticide-treated bednets (ITN) should be dried indoors. The conclusion of several other studies (Lindsay *et al.*, 1991; Pleass *et al.*, 1993; Curtis *et al.*, 1996) was that, to be dried after each wash, treated nets should be laid horizontally (on polythene sheets) — to avoid the problem of insecticide being carried, by gravity, to the lower portions of wet nets hung vertically to dry — and indoors — to avoid possible degradation of the insecticide by ultra-violet (UV) light. The full effects of sun exposure on the activity of an insecticide deposited on ITN remain to be elucidated (Kayedi *et al.*, 2008). It is often simply assumed that the UV rays in sunlight may degrade any synthetic pyrethroid deposited on nets because natural pyrethrum is biodegradable and does undergo photo-degradation when exposed to UV light (Barlow *et al.*, 2001). In the present study, several methods of drying nets following deltamethrin treatment were compared, in terms of the insecticide levels, and insecticide-level uniformity, on the dry nets.

MATERIALS AND METHODS

Field studies were conducted in Iran and laboratory studies in the U.K. (at the London School of Hygiene and Tropical Medicine, in London). The same type of bednet [untreated, single size, white, rectangular, polyester nets, from Vestergaard–Frandsen (Koldin, Denmark)] and the same method of deltamethrin treatment [soaking each net in a solution made with one K-O Tab® tablet (Sanofi-Aventis, Guildford, U.K.) in 500 ml water] was used in both countries.

Field Studies Carried Out in Iran

In Iran, 20 nets were treated and then dried overnight in the shade — 10 folded once lengthways and hung vertically, over a line (with the fold over the line) and 10 laid flat on the floor, on plastic sheets. After drying for 12 h, 5×5-cm samples were cut (two each from the top outer surface, bottom outer surface, top inner surface and bottom inner surface of each net dried vertically and two each from the central upper and central lower surfaces of each net dried horizontally. Each of the 120 samples (plus two cut from an untreated net, as controls) was sealed in a plastic bag, labelled and sent to London, where high-performance liquid chromatography (HPLC) was used to evaluate the sample's deltamethrin content (Graham *et al.*, 2005).

Laboratory Studies Carried Out in the U.K.

In London, another 32 nets were treated. Each wet net was then folded three times (twice vertically and once horizontally) and dried while hung vertically, over a line (eight nets in the sun and eight in the shade) or laid horizontally on plastic sheets on the floor/ground (eight nets in the sun and eight in the shade). Although all the nets were left to dry for 24 h, those left 'in the sun' were only exposed to direct sunlight for about 10 h. After drying, square (5×5-cm)

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samples were cut from each net and tested, like the Iranian samples, for deltamethrin content, by HPLC. Eight samples were cut from each net that had been dried vertically (two each from the top outer surface, bottom outer surface, top inner surface and bottom inner surface, as in Iran) and six from each net that had been dried horizontally (two each from the centre of the upper surface and the centre of the lower surface and one each from the upper and lower sections of the middle surface).

COLOUR CHANGES

After the London nets were dried, they were found to have brown patches (between white areas) on their outer surfaces (whether dried vertically or horizontally). Since the colour of the treatment solution made by dissolving one K-O Tab tablet in 500 ml water is brown, it was thought that the brown staining may indicate areas of net that had relatively high levels of deltamethrin. To explore this possibility, two nets were each 'treated' with 500 ml tap water and dried vertically (one net) or horizontally (one net), to see if the brown staining occurred in the absence of insecticide. As no brown colouration was detected on either of these nets, further samples were cut from some of the other ITN (some from the white areas of netting and some from the brown) and assayed for deltamethrin content.

Statistical Methods

Each net sample was weighed so that its deltamethrin content could be expressed both as mg/m^2 net surface and as g/g of netting.

As the distribution of the concentrations recorded showed positive skewness and the log-transformed concentrations were not found to be normally distributed (Kolmogorov–Smirnov test; $P<0.001$), non-parametric Mann–Whitney *U*-tests were used to determine the statistical significance of the differences observed. To reduce the inflation of statistical errors as the result of the multiple comparisons, only *P*-values of <0.01 were considered indicative of statistically significant differences.

RESULTS

Field Studies Carried Out in Iran

The mean deltamethrin concentration on the nets dried vertically in Iran was very similar to that on the nets dried flat (Table 1). The upper and lower sections of the Iranian nets also had similar insecticide concentrations, whether the nets were dried vertically or horizontally ($P>0.06$ for each; Table 1). With the nets dried vertically, however, the surfaces of the net that were outside during the drying had significantly higher concentrations of deltamethrin than the inner surfaces (Table 1).

Laboratory Studies Carried Out in the U.K.

In the London-based experiments, the mean deltamethrin concentration on the nets dried in the sun was very similar to that on the nets dried in the shade (whether the nets were dried horizontally or vertically; $P>0.45$ for each comparison; Table 2).

The concentration of deltamethrin on the lower section of a net dried vertically in London was almost twice that on the upper section ($P<0.001$; Table 2), whereas the concentration of deltamethrin on the upper surface of a net dried horizontally was more than double that on the lower surface ($P<0.001$; Table 2). Also in London, as in Iran, the concentration of deltamethrin on the outer surface of a net dried vertically was significantly greater than that on the inner surface of such a net ($P<0.001$; Table 2).

As expected, the brown patches of netting held significantly more deltamethrin than the white, with mean (S.D.) concentrations of 56.7 (26.6) and 55.1 (29.4) mg/m^2, respectively ($P<0.001$).

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TABLE 1. *The results of the field studies in Iran — deltamethrin concentrations stratified by drying position in the shade*

Drying position and source of sample	Mean (S.D.) deltamethrin concentration (mg/m^2)	*P*	Mean (S.D.) deltamethrin concentration (g/g)	*P*
VERTICAL				
Upper	14.7 (4.7)		0.04 (0.01)	
Lower	14.7 (7.5)	0.86	0.04 (0.02)	0.86
Inside	11.8 (3.6)		0.03 (0.009)	
Outside	17.4 (7.1)	0.001	0.04 (0.02)	0.001
All	14.7 (6.2)	–	0.04 (0.016)	–
HORIZONTAL				
Upper	15.7 (2.9)		0.04 (0.007)	
Lower	14.3 (4.1)	0.06	0.04 (0.01)	0.07
All	14.9 (3.5)	–	0.04 (0.009)	–

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DISCUSSION

Mean deltamethrin concentrations were greater in the lower parts of nets hung to dry vertically than in the upper parts (significantly so in London), and were also greater on the upper surface of nets dried horizontally than on the lower (significantly so in London). In vertical drying it appears that gravity drags the insecticide solution downwards while the water is evaporating whereas in flat drying the effects of gravity are outweighed by the effects of the evaporation pressure that draws the insecticide solution to the upper, exposed surface. The effects of gravity and evaporation pressure were, in fact, visible to the naked eye, since the higher-than-average concentrations of insecticide that they caused led to a brown colouration on the dry nets — on the top of folds in the upper surface of nets dried flat and on the outer surface of nets dried vertically. It was probably evaporation pressure that also made deltamethrin concentrations about twice as high on the outside of nets dried vertically than on the inside of such nets ($P \leqslant 0.001$ in both London and Iran). In general, the outer layers and folds of the nets contained more deltamethrin than the inner folds, consistent with the

TABLE 2. *The results of the laboratory studies in London — deltamethrin concentrations stratified by drying position*

Drying position and source of sample	Mean (S.D.) deltamethrin concentration (mg/m^2)	*P*	Mean (S.D.) deltamethrin concentration (g/g)	*P*
VERTICAL				
Net dried in sun	41.6 (41.5)		1.3 (1.3)	
Net dried in shade	31.8 (28.3)	0.478	1.1 (0.9)	0.526
Upper	24.6 (13.1)		0.8 (0.4)	
Lower	48.9 (45.8)	0.001	1.6 (1.4)	<0.001
Inside	25.7 (29.8)		0.8 (0.9)	
Outside	47.7 (37.9)	<0.001	1.6 (1.2)	<0.001
All	36.7 (35.7)	–	1.2 (1.1)	–
HORIZONTAL				
Net dried in sun	49.5 (49.2)		1.5 (1.4)	
Net dried in shade	43.6 (66.0)	0.878	1.3 (1.9)	0.735
Upper	65.7 (72.2)		1.9 (2.1)	
Lower	27.4 (29.0)	<0.001	0.8 (0.8)	<0.001
All	46.6 (58.1)	–	1.4 (1.7)	–

hypothesis that deltamethrin accumulates at the points where the water evaporates from the drying net.

As expected from the results of earlier studies (Kayedi, 2004; Kayedi *et al.*, 2008), exposure of the drying nets to the sun had no significant effect on the concentration of deltamethrin in the nets, whether the nets were dried vertically on a line or flat on the ground (Table 2). Deltamethrin is, however, a second-generation pyrethroid and probably the most sun-resistant of all pyrethroids. If permethrin — a first-generation pyrethroid that is more UV-susceptible than deltamethrin (Snow *et al.*, 1988; Barlow *et al.*, 2001) — had been used in the present study, sun exposure while the nets were drying may well have had a marked detrimental effect on insecticide deposits on the dry nets.

Curiously, only in London were (1) the vertically dried nets found to have significantly higher insecticide concentrations on their lower sections than on their upper sections and (2) the flat-dried nets found to have significantly higher insecticide concentrations on their upper sections than on their lower sections (Table 1). In Iran, where nets hung on a line were only folded once (not three times, as in London) and where drying temperatures were relatively high (>45°C, compared with 19°C in London), the wet nets probably dried much more rapidly than their counterparts in London. Such rapid drying would presumably reduce the flow of insecticide solution caused by gravity or evaporation pressure and tend to fix insecticide concentrations at a uniform level across the nets.

In conclusion, in order to obtain an even distribution of deltamethrin at an adequate level, it is not necessary to dry treated nets in the shade. To achieve a fairly uniform deposit of deltamethrin, the drying net should be folded as little as possible, to minimise drying times.

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