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REPORT OF THE FIRST WHOPES WORKING GROUP MEETING

WHO/HQ, GENEVA
26-27 June 1997

WORLD HEALTH ORGANIZATION
DIVISION OF CONTROL OF TROPICAL DISEASES (CTD)
WHO PESTICIDE EVALUATION SCHEME (WHOPES)

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1. Objectives

The purpose of the meeting was to review the reports of field trials of pesticides that have reached the final stages of evaluation by the WHO Pesticide Evaluation Scheme (WHOPES) and to make recommendations to the Division of Control of Tropical Diseases (CTD) for their use in vector control.

The reports submitted to the Working Group related to field trials of indoor residual spraying or bednet impregnation with etofenprox (Mitsui Toatsu Chem., Japan) or deltamethrin (AgrEvo, UK) in one or more of the following formulations: etofenprox (20WP, 20% wettable powder; 20EC, 20% emulsifiable concentrate; 10EW, 10% emulsion oil-in-water) for the purpose of *Anopheles* or *Triatoma* control, and deltamethrin (25SC, 2.5% suspension concentrate) for *Triatoma* control.

2. Preparatory work

All the reports to be reviewed were sent to the participants in April 1997 for advance study to facilitate review at the meeting. In addition, each participant was requested to study specifically one or two related reports and to prepare an abstract of each one, critically analyzing the objectives, methodology and the significant contents and main conclusions of the research, with special reference to the objectives of WHOPES.

The abstracts prepared by participants were also circulated prior to the meeting. The list of the supporting/background documents and participants are presented in Annexes I and II respectively.

3. Conduct of the meeting

The meeting took place at WHO Headquarters in Geneva from 26-27 June 1997. Dr K. Behbehani, Director of CTD, opened the meeting on behalf of the Director-General. Dr Yap Han Heng was appointed as chairman and Dr. J.A. Najera as rapporteur.

In his opening address, Director of CTD highlighted the importance of the WHOPES Programme to support the development of new pesticides for public health use and, in particular, those potentially applicable to the renewed efforts in malaria and dengue control; the coordinated efforts for Chagas disease control in the central and southern America; and the continued development of onchocerciasis control. The need for development and evaluation of new products was emphasized, not only to cope with the problem of multiple resistance, but also to optimize the application of new approaches such as use of impregnated bednets. Director of CTD also referred to the re-orientation of the WHOPES to ensure further strengthening of the programme. The need for capacity building of Collaborating Centres in endemic countries was stressed to improve the quality of work and to speed up evaluation of candidate pesticides and their formulations.

Dr M. Zaim, scietist in charge of WHOPES, reviewed the status of development of the two pesticides and their formulations under consideration, recognizing that the trials under review were planned and executed before the recent redefinition of the WHOPES phases (CTD/WHOPES/IC/96.1). Dr Zaim referred to the large amount of scientific knowledge which exists on etofenprox, and referred to the use of deltamethrin SC, which has become the *de facto* reference for *Triatoma infestans* chemical control. In both cases, a considerable body of information has become available from trials in several countries and, therefore, the WHOPES trials were aimed at confirmation and consolidation of existing knowledge. The meeting proceeded to discussion of the reports, which were presented by the participant who had carried out the preliminary review.

4. Background/supportive documents (Annex I)

Phase II trials of residual spraying of etofenprox WP in Burkina Faso and impregnation of bednets with etofenprox EC in Burkina Faso (Bobo-Dioulasso) and Muheza (Tanzania), demonstrated a high repellent effect of this insecticide in preventing entry into experimental huts and inducing exophily, being one of the most repellent pyrethroids

tested in Bobo-Dioulasso, where the strong excito-repellency was shown also on impregnated bednets at the very low dose (25 mg a.i./m²) tested. Nevertheless, it appears that immediate mortality with etofenprox is lower than with other pyrethroids tested and mosquitos had time to bite once inside the house (gorging rate was 94%); similarly, in the experimental huts in Muheza impregnated bednets at a dose of 200 mg a.i./m² showed one of the lowest ability in preventing feeding (geometric mean of *Anopheles* succeeding in feeding 11.5% *vs.* 1.4% with permethrin EC at 500 mg a.i./m²), although it produced a relatively high mortality (geometric mean of 64.6% *vs* 61.2% with permethrin EC); it should be noted that prevention of entry was not reported from Muheza and that results of total mortality in experimental huts did not report separately the mortality inside the hut and after reaching the exit traps.

Although pyrethroids are generally not recommended for larviciding against mosquitos, the Onchocerciasis Control Programme (OCP) in West Africa has included etofenprox EC at a dose of 0.03 mg a.i./l for 10 minutes, among the seven larvicides currently in use. The effects on non-target organisms were investigated, especially on shrimp, usually very susceptible to pyrethroids. On *Caridina sp*, it had no detectable effect in the field. From bioassays it was shown that six times the operational dose is needed to cause 50% mortality of *Caridina* and 30 times this same dose for 95% mortality. Immediately after application, some increased drift (detachment of total fauna) by use of etofenprox was noted; however no disruption was noted in the relative composition of aquatic invertebrate communities. The observed drift was far below that observed for permethrin. The use of etofenprox by OCP was then approved by the Ecological Group (an independent panel of experts).

Etofenprox is a non-ester pyrethroid insecticide with comparable insect toxicity and a similar mode of action to other pyrethroids. Cross-resistance studies on standard susceptible and resistant laboratory strains of *An. gambiae, An. albimanus, An. stephensi,* and *Culex quinquefasciatus* showed no effect of carboxylesterase, elevated esterase, altered acetylcholinesterase or glutathione S-transferase-based resistance mechanisms, while cross

resistance to etofenprox occurred in a pyrethroid-resistant strain of *Cx. quinquefasciatus* with both oxidase and *kdr*-like resistance mechanisms.

Trials on the use of impregnated bednets had been conducted in North Viet Nam, with etofenprox EC and EW formulations, and in South Viet Nam and Cambodia with etofenprox EW, all at a dose of 200 mg a.i./m^2; all trials indicated a marked effect on resting and biting densities and high residual bioassay mortalities between 4 and 6 months with residual effect on wood one or two months longer than on sorptive materials.

Regarding the efficacy of impregnated bednets on long term use and different storage conditions, as well as their residual effect after laboratory-controlled washing, CDC investigated etofenprox 10 EW (300 & 500 mg a.i./m^2) in comparison with permethrin 10% EC (300 & 500 mg a.i./m^2) and deltamethrin 2.5% EC (15 & 25 mg a.i./m^2), on polyester netting material (100 denier), with the following results:

(i) Bioassay tests were carried out every three months for one year on impregnated materials kept under three conditions of storage: (a) in the dark (on a drawer); (b) hanging on a laboratory room, exposed to indirect sunlight; and (c) in a greenhouse where the roof was coated with a semi-opaque substance and a heating/air conditioning system which ensured constant temperature. Results with etofenprox were intermediate between the other two insecticides, with deltamethrin clearly showing a stronger residual effect; etofenprox at 500 mg a.i./m^2, showed a potentially useful residual effect of 6-8 months, although chemical analysis (GLC) showed a faster decay of etofenprox under greenhouse conditions.

(ii) The effect of washing was investigated, by both bioassay tests and chemical analysis, after one to three washing cycles consisting of: placing the netting sample in a jar containing 100 ml of aqueous 2% SDS; mechanical shaking for 5 minutes; rinsing by shaking for 10 minutes in 300 ml of de-ionized water; and wringing and air drying overnight. Results again showed

etofenprox to be intermediate between the other two insecticides. At the dose of 500 mg a.i./m² it caused about 35% mortality after a second wash, with 3 minutes exposure.

5. Insecticides and formulations tested in phases II & III (WHOPES) field trials

The following insecticides/formulations/doses and target vectors were reviewed:

- For indoor residual spraying

1. Against *Anopheles*
   - 1.1 Experimental huts
     - Etofenprox 20WP (100, 200 & 400 mg a.i./m²) **vs** unsprayed hut **(Thailand)**
   - 1.2 Village scale
     - Etofenprox 20WP (300 mg a.i./m²) **vs** DDT 75% WP (2 g a.i./m²) **(Thailand)**
     - Etofenprox 20WP (500 mg a.i./m²) **vs** unsprayed village **(Iran)**
   - 1.3 Large scale
     - Etofenprox 20WP at 300 mg a.i./m² **vs** malathion (2 g a.i./m²) **(Sri Lanka)**
     - Etofenprox 20WP (100 mg a.i./m²) **vs** DDT 75% WP (2 g a.i./m²) **(Philippines)**
     - Etofenprox 20WP (200 mg a.i./m²) **vs** unsprayed village **Indonesia)**
     - Etofenprox 20WP (500 mg a.i./m²) **vs** unsprayed area **(Iran)**

2. Against *Triatoma*
   - 2.1 Village scale
     - Etofenprox 20WP (250, 500 & 750 mg a.i./m²) **vs** etofenprox 20EC (250, 500 & 750 mg a.i./m²) **vs** etofenprox 10EW (250, 500 & 750 mg a.i./m²) **vs** deltamethrin 25SC (25 mg a.i./m²) **vs** unsprayed houses **(NC Bolivia)**

- Deltamethrin 25SC (25 mg a.i./m²) **(S. Bolivia)**

- For bednet impregnation against anopheline vectors

1. Experimental huts
   - Etofenprox 20EC (100, 200 & 400 mg a.i./m²) **(Thailand)**
   - Etofenprox 20EC **vs** Etofenprox 10EW (300 mg a.i./m²) **(Thailand)**
2. Village scale
   - Etofenprox 10EW (300 mg a.i./m²) **(Thailand)**
3. Large scale
   - Etofenprox 20EC (200 mg a.i./m²) **(Indonesia)**

6. Main results

As mentioned above, these trials were conducted before the redefinition of WHOPES requirements, most were planned as part of the efforts of National Antimalaria Programmes to find alternative insecticides to maintain or improve their vector control operations in their "problem areas" for malaria control, instead of being planned with the purpose of exploring the characteristics of the insecticide. Evaluation aimed at determining whether or not transmission was interrupted, following the methods commonly used in traditional antimalarial programmes.

It is noteworthy that most of the malaria control problem areas, outside tropical Africa, are only partially responsive to indoor residual spraying, due to exophilic vectors, dispersal and high mobility of the population and/or difficult accessibility; malaria transmission is often highly variable from year to year and from locality to locality, with a tendency to periodic epidemic outbreaks, due to ecological and climatic variability. Many of these problem areas are situated in international borders, in arid areas with nomadic or semi-nomadic populations, in areas of economic expansion, such as agricultural developments attracting temporary labour forces, or in jungle areas invaded by mining, timber and other forest exploitations.

Although the programmes responsible to control malaria in those areas feel an urgent need to test new insecticides, their ecological and social conditions and the unstable and relatively low transmission, makes evaluation of the direct effects of the insecticides, on the disease, rather difficult, requiring long periods of observation. Although such studies provide, valuable information, they could be more profitably planned after the characteristics of the insecticide, which determine its effectiveness and the conditions and limitations on use have been quantified.

The reviewed trials were carried out in relatively small areas and in the short period of one year; most antimalarial programmes could not allow to have an unsprayed comparison area and the trials had to attempt to compare the test insecticide with those normally used by the programme (DDT or malathion) which, being also partially effective, reduced the margin of comparison. Moreover, most trials had no baseline information or only limited to one or two months before the beginning of the intervention; this often lead to the problem that a great part of the period of observation occurred during a natural decline in vector activity.

All trials showed a total disappearance of indoor resting mosquitos immediately after spraying, maintained for several months and only partially recovering at the end of the trial. Although quite variable results were obtained in different trials, all of them confirmed the high persistence of etofenprox on non-sorbent materials (*e.g.* wood and bamboo), while considerably less on cement and mud surfaces.

None of the trials conducted observations on total house mortality or on the modifications of vector behaviour in relation to house entering and biting, which could have been induced by the insecticide; the use of exit traps was limited to the observation of delayed mortality and a partial observation of success in feeding, since there had not been a parallel collection of fed mosquitos having died before reaching the exit traps.

The main results are summarized below:

(a) **for *Anopheles* control:**

Operational dose

As indicated above, the selected doses for village or large-scale testing of etofenprox varied from 100 to 500 mg a.i./m² for residual spraying of anopheline control and 250 and 750 mg a.i./m² for triatomine control; impregnation of bednets was tested at doses of 100, 200 and 300 mg a.i./m².

Although panel bioassay tests show a rather clear dose-residual effect relation, this was not so clear in the limited comparisons made in experimental huts but, as mentioned above, full comparisons could not be made in the absence of complete assessment of mortality and the success of feeding.

In the case of impregnated bednets, comparisons were mainly limited to residual bioassay mortality or, in the case of the two village scale trials, to the mass effect on vector densities and malaria transmission. No measurement was made on the prevention of man-vector contact due to the repellent effect of the insecticide, in spite of the very high effect shown in Burkina Faso phase II trials at a very low dose (25 mg a.i./m²), nor on total mortality and success in feeding (as had been assessed in stage II in Tanzania).

Coverage and actual dose achieved

Reported coverage was high, over 80% in all trials and over 90% in most; it compared favorably with either DDT or malathion, due to the absence of objectionable odour and discoloration of sprayed surfaces, although some white markings were observed in the Thailand experimental hut studies, on sprayed surfaces at the dose of 400 mg a.i./m². Knowledge, attitude and practice surveys (KAP), conducted in some trials, confirmed the greater acceptability of etofenprox, compared with DDT and malathion.

Wall scrapings, expressed as mg a.i. of insecticide per kg of sprayed material showed a considerable variation from 27.5 to 264 mg a.i./kg of mud wall, in the samples taken in the first week after spraying, in the Sri Lanka trial; on average, results showed a general decline with time, nevertheless, difficulties in standardization of sampling procedure does not allow to determine how much of the great variability is due to variation in actual dose and how much to differences in scraping technique.

Regarding bednet impregnation, chemical analysis of twelve samples taken in the Thai trial, showed a range of 99.6-177 mg a.i./m^2, compared with a target dose of 300 mg/m^2; on the average cotton gave higher etofenprox content: 144.1 ($\pm$ 29) versus 130.8 ($\pm$ 24) mg a.i./m^2 on nylon nets.

Vector density

Various methods were used to assess changes in overall vector density and the influence of the insecticide used, either as indoor residual spray or as impregnated bednets. Only man-landing (biting) rates were used in all anti-anopheline trials; other methods, such as animal-bait collections, artificial outdoor resting places and larval collections, were used in some trials and mostly gave inconclusive results.

Man-biting rates were also difficult to interpret in view of the lack of baseline data, the similarity in results between test and comparison areas, the small size and the lack of isolation of the test areas. Nevertheless, the fact that all trials show some decline in vector densities points to an effect of the insecticide.

The village scale trial in Iran, conducted in ecologically isolated villages and including comparison with an unsprayed village, showed more clearly the effect of etofenprox in reducing man-biting rates after spraying while they continued to increase in the unsprayed villages.

It should be noted that the impact of indoor spraying on total mosquito densities depends to a great extent on the importance that the

house, and other sprayed shelters, have as main day time resting places; in a very arid area, like that of the Iran trial, when the killing and repellent effects of the insecticide succeeds in reducing the most suitable day-time resting place for the vector, thus, its survival rate is greatly reduced.

Malaria incidence and prevalence

As mentioned above, most trials were carried out in areas of low and unstable malaria transmission and, although most of them showed a reduction in malaria cases in the test areas, it is difficult to estimate the contribution of spraying to those reductions.

The Indonesian trial seems to have been conducted in relatively isolated villages in an area where movement of population was mainly migratory, showing a marked population decline during the period of study. Although there is no long term baseline information, periodic prevalence surveys show a continuous decline in parasite rates and in the proportion of *Plasmodium falciparum* in both the sprayed area and the village using impregnated bednets, while both indicators remain quite stable in the control area.

As with the impact on total mosquito densities, the expected effect on malaria transmission should be due to the joint action of the repellent and killing effects of the insecticide, which are expected to contribute to a reduction of biting rates and to an increased mortality of those which succeed in feeding.

Residual effects, as measured by bio-assays

All trials confirmed the long lasting residual effect of etofenprox on wood and other non-sorbent substances; results are available from a number of tests indicating that the residual effect is progressively shorter in a series of substrates such as wood-corrugated iron-thatch-cement-mud; it should be noted that tests on corrugated iron were made on sheets kept inside an experimental hut and not on an actual hut roof, where exposure to extreme temperature could have a deleterious effect on insecticide deposits.

Most bioassay test sequences in several areas, show an obvious drop in mortality, around the third or fourth month after spraying, followed by an increase in subsequent months. It is quite likely that, as spraying is generally done during the main transmission season, the period of mortality decline in bioassay test may coincide with the hottest (driest) period of the year, when the effect of the insecticide may have been reduced by high temperature or its surface availability also reduced by increased absorption due to low humidity.

Comparative trials of EC 20% and EW 10% formulations for bednet impregnation, were made to test their resistance to washing with water alone and with water and detergent. Both formulations gave the same residual mortality in unwashed nets (47-68% mortality for 6 months), while EW10 showed more resistance to washing, 20-40% for EW10 versus 2.5-12% mortality for EC20. Differences between water and detergent washing were very small for the EW formulation while they were very noticeable for the EC; mortality after washing with detergent was about half of that after washing with water alone.

(b) for *Triatoma* control

The field effectiveness of both deltamethrin 25SC and the three formulations of etofenprox was determined by the search of triatomines in treated houses and by the longitudinal study of mortality of wall bioassays. After treatment, only houses that had been previously found infected were inspected. At 45 days post-spray, there was still 25% house infestation in 4 of the 9 etofenprox dose-formulation combinations, but not in the houses sprayed with deltamethrin. At 115 days overall infestation decreased to 5.6% and thereafter progressively increased to 14.0% at 365 days. House infestations were lowest for WP at 750 mg a.i./m^2 (none of 10 houses infested after 45 days); for all other combinations of etofenprox, except for the lowest dose (250 mg a.i./m^2) of EC, only one of the ten inspected houses became infested after 284 days.

The two trials showed that despite some indications of a dose-response, the bioassay and infestation rates for most dose-formulation

combinations were similar to deltamethrin applied at 25 mg a.i./m², so that the lowest dose (250 mg a.i./m²) of etofenprox WP or SC could be used for *T. infestans* control. Residual activity of deltamethrin, in southern Bolivia, remained high for 12 months, although mortality was considerably reduced during the hot period of the year. The cost, per house per year of etofenprox WP20 at 250 mg a.i./m² appears to be similar to that of the application of deltamethrin SC at 25 mg a.i./m².

7. Pending questions

As indicated above, there remain, not only for etofenprox, but still for most pyrethroids, important lacunae in our understanding of the dynamics of man-vector contact and vector survival in the presence of indoor spraying or the use of impregnated bednets; such questions would require detailed investigation in experimental huts under well selected, different ecological conditions.

In spite of being the subject on which more information is available, there still remain some pending questions on the residual effect of etofenprox WP and deltamethrin SC, on different sprayed surfaces, under different climatic conditions, particularly temperature and humidity; it is necessary to standardize the sampling methods and determine the value in the field of chemical analysis of wall scrapings or bednet material; it should be also necessary to standardize, for WHOPES trials the minimum requirements of bioassay testing (*e.g.* time of contact or the time to knockdown).

There seems to be no accepted criteria for the selection of operational doses, except the carrying out of a previous series of bioassay tests, which are only a measure of immediate contact mortality. This lack of criteria is particularly obvious in the case of doses for impregnation of bednets, which seem to be based on those used for residual spraying, in spite of the very different mode of action of the two methods of insecticide use.

The repellent effect of the etofenprox acquires a predominant role in the use of bednets for personal protection, or for disease prevention by the additive effect of individual protection; in those

cases, the dose required for impregnation may be considerably lower than those used in the reported trials; phase II trials in Bobo-Dioulasso showed a very high repellent activity with a dose as low as 25 mg a.i./m².

More information is needed on relative advantages of EW and EC formulations of etofenprox for bednet impregnation; although there are few indications of longer residual effect of EC than of EW formulations (North Viet Nam trial refers), there is strong evidence (both from CDC and from the Thai trial) that EW may be more resistant to washing and, in general, non-solvent based formulations are considerably less irritant and less smelly and, therefore, preferred by users.

It is necessary for WHOPES to develop methodological guidelines for field trials aimed at adequate characterization of new insecticides, as well as for the selection of insecticides to be used for disease control under main eco-epidemiological types.

8. Conclusions

1. The accumulated experience and trials reviewed confirmed that etofenprox 20 WP is a safe and effective insecticide for use in indoor residual spraying in vector control operations for both malaria and Chagas disease control. The use of etofenprox as a larvicide for *Simulium* control has already been established by the Onchocerciasis Control Programme in West Africa.

2. Similarly the safety and effectiveness of deltamethrin 25 SC for *Triatoma infestans* control is recognized and a field trial showed that etofenprox at a dose of 250 mg a.i./m² could be as cost-effective as the standard deltamethrin at 25 mg a.i./m².

3. Etofenprox WP has been effectively used at dosages of 100-300 mg a.i./m² for malaria vector control. Nevertheless, the operational dose to be used depends on local conditions and needs of the control programme:

- sprayable surface materials can alter the residual effect, the insecticide is more lasting on non-sorbent surfaces, like wood and shortest in mud or cement.

- high temperature also reduces the effectiveness of pyrethroids and low humidity may reduce their surface availability.

- length of transmission season may be an argument to increase the dose of application to cover the necessary period of control instead of conducting two rounds at a lower dose.

4. The very low toxicity of etofenprox and its effectiveness shown in field trials demonstrates its suitability for bednet impregnation, nevertheless, there are a number of pending questions which should be resolved in order to guide the choice of formulation.

5. The implementation of well controlled experimental hut studies of impregnated bednets to test various concentrations and to compare etofenprox EW and EC formulations in different contrasting ecological situations is recommended, *e.g.* humid *vs* arid, with enough endophilic and anthropophilic mosquitos, studying the impact on entry into the hut, total hut mortality and success in feeding (distinguishing for both dead and fed mosquitos, those collected inside the hut from those in exit traps, as well as delayed mortality).

Annex I

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Annex II

List of participants

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