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COMPARATIVE EFFICACY OF SYNTHETIC PYRETHROID EMULSIFIABLE CONCENTRATE (EC) FORMULATIONS AS THERMAL FOG AGAINST *CULEX QUINQUEFASCIATUS*

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ABSTRACT
Synthetic pyrethroids utilized in household insecticide formulations were assessed for their application as fogging concentrates against laboratory bred *Culex quinquefasciatus*. EC formulations of synthetic pyrethroids namely- d-allethrin, d-trans allethrin, Permethrin and Prallethrin were evaluated for their efficacy at targeted dosages in Peet Grady chamber in comparison to Pyrethrum extract. The study reveals that synthetic pyrethroids especially 2% w/w Permethrin and 2% w/w Prallethrin EC formulation at target dosages hold great promise as space spray (fogging concentrate) application for control of vector mosquitoes in urban areas.

KEY WORDS: Culex, Insecticides Efficay, Pyrethroid

INTRODUCTION

Thermal fogging and ULV space spray are two of the most common methods employed by the Municipal Corporations (local health authority) for rapid reduction of nuisance mosquito (*culex quinquefasciatus*) and other urban vector mosquitoes (*Aedes aegypti* and *Anopheles stephensi*) during epidemic situation. Space spraying applied as thermal fog is therefore an ideal method for bringing about rapid control of vectors in emergency or epidemic situations and may also be used for seasonal control of flying insects, pests (WHO India, 2009).
National Vector Borne Control Program (NVBDCP) in India recommends 2% Pyrethrum extract and Malathion (Tech) for thermal fog and ULV application respectively for control of Dengue and Dengue Hemorrhagic fever (Deobhankar, 1996). Currently there are three thermal fog formulations available in the market for adult mosquito control –Pyrethrum extract 2%, Cyphenothrin 5%EC and Deltamethrin 1.25% EC. (WHO India, 2009).

Presently there is a need of cost effective and efficacious thermal fog formulations that could be find its use in vector control programs. Synthetic Pyrethroids used in household insecticides offer such a potential that could be effectively utilized in thermal fog formulations. In the present study, bio-efficacy of EC formulations containing d-allethrin, Prallethrin, d-trans allethrin and Permethrin was compared with Pyrethrum 2% extract as thermal fog against caged *Culex quinquefasciatus.*

MATERIALS AND METHODS

1. Preparation of EC formulations

Four Emulsifiable Concentrate formulations containing 2% w/w of active ingredients (Prallethrin, d-trans-allethrin, d-allethrin, Permethrin sourced from Ms Shogun Organics Limited) were prepared by adding water and surfactants. Pyrethrum 2% extract was procured from the local market.

2. Preparation of fogging premix

50 ml of each fogging concentrate (as in 1) was diluted with 950 ml of diesel to obtain a total volume of 1 liter (Table 1). In case of Allethrin 100 ml of fogging concentrate was diluted with 850 ml of diesel. The resulting premix was poured into fogging machine for generating fog.

3. Calibration of fogging machine

Potable thermal fogging machine manufactured by Ms Foggers India Limited was used for trials.
(Model: Foggers LOC Handy Fogging Machine, model no: 10116)
Calibration was done as per the method adopted by earlier author (Deobhankar, 1996). Calibration of fogging machine was done to determine the discharge rate (ml/minute) by filling the solution tank and running the machine for 15 minutes and then measuring the quantity of diesel discharged. In 15 minutes, 980 ml diesel was discharged giving the discharge rate of 65.33 ml /minute. For 6 cubic meters of Peet Grady chamber at the rate of 4 ml/cubic meter, 24 ml fogging solution was delivered in 0.367 minutes (corresponding to 22 seconds). This quantity was accurately delivered by measuring time by stop-watch in each case (candidate formulation).

4. Test insects

Culex quinquefasciatus mosquitoes for bio-efficacy studies were reared in laboratory at temperature of 28 ±1◦ C with a relative humidity of 60-70 % and light: day photoperiod of 14:10 hours. Larvae were reared in plastic trays filled with de-chlorinated tap water. Yeast powder was provided as nutrient for mosquito Pupae were collected and allowed to emerge in a gauze cage of 62 cm x 48cm x 48 cm. A diet of 5% sucrose solution was provided for the adults.

4. Bio-assay

Fogging trials were conducted in Peet Grady chambers. All six sides of the chamber are of glass. The chamber is provided with a duct to vent the fog after the test is completed. The chamber is maintained at 27 ± 1◦ centigrade and

relative humidity of 55 ±5% during the test period. The chamber is well lit to facilitate observation during the trial (Figure -2).

Prior to initiation of experiment the Peet Grady chambers were checked for decontamination by exposing caged mosquitoes for a period of 30 minutes. Mosquitoes were caged in bullets that comprised of polypropylene cylinder admeasuring 9 cm diameter X 5 cm height with both faces covered with stainless steel gauze to prevent escape of mosquitoes and facilitate penetration of fog. Each bullet contained 5 adult female mosquitoes (5-6 day old). 10 such bullets were hanged at the center of the room at a height of 90 cm (approximately 3 feet) from the ground level.
Experiment was initiated if the knockdown was observed to be less than 10%. If the observed knockdown exceeded 10% the chamber was sanitized with detergent and vented to conform to decontamination norms.

After each experiment the test chambers were decontaminated with detergent water and vented adequately. Decontamination check was carried out as described above. Once the contamination check was done test mosquitoes – *Culex quinquefasciatus* were introduced in the Peet Grady chamber using ten bullets each containing five adult female mosquitoes (5-6 day old). All the bullets were hanged in Peet Grady chamber in the center at a height of 90 cm (approximately 3 feet from the ground level). All the ports in the test chamber were placed in shut-off mode and exhaust system was turned off. Pre-calibrated fogging machine was started outside the test area and was allowed to run for 5 minutes to ensure uniform fog generation. Measured quantity of fog was introduced in the test chamber using stop watch and the mosquitoes were exposed to fog for 2 minutes and 5 minutes. Five bullets were removed after an exposure of 2 minutes and the rest 5 bullets at the 5^{th} minute.

Control consisted of exposing equal number of mosquitoes in separate chamber with fog containing only diesel solution with no active ingredient. Number of mosquitoes knocked down (KD) and the number of dead mosquitoes (mortality) was recorded for each bullet containing 5 mosquitoes after 2 minutes and 5 minutes of exposure period and mortality after 24 hours. The observations recorded were translated into mean percentages for 2 sets of readings. For observing, mortality mosquitoes were transferred to clean cages and provided with 5% sucrose solution.

RESULTS AND DISCUSSIONS

The summarized results depicting mean values for % knockdown and mortality for 2 minute and 5 minute exposure period for two sets of experiments are shown in Table 2.

The graphical representation (Figure-1) clearly indicates that 2% Permethrin EC formulation gives higher knock-down of mosquitoes coupled with higher percentage mortality after 24 hours for both 2 minute and 5 minute exposure period. Pyrethrum extract which is used as a reference standard gives least knock-down and mortality values among the formulations tested. Although, the 2 minute exposure values for other formulations (Prallethrin, d-trans-allethrin and Allethrin) are not satisfactory, the 5 minute exposure level gives greater than 80% knockdown and mortality values with Prallethrin 2% EC formulation being better than the other two (d-trans-allethrin and Allethrin).

Synthetic pyrethroids used in household insecticides have hitherto not been used for their application in thermal fog formulations due to the apprehension of being degraded during application as thermal fog. The results clearly depict potential of such household synthetic pyrethroids to be used for vector control application in the control of mosquito borne diseases due to their efficacy at lower dosages which have less harmful impact on environment. Unlike other organophosphate compounds (malathion and fenitrothion) the use of synthetic pyrethroids also eliminates unpleasant smell thereby making them more acceptable to the community. Formulations (Resigen) containing multiple actives (S-Bio-allethrin and Permethrin and Piperonly butoxide) have been found to be effective for adult control (WHO, 1995). As *Culex quinquefasciatus* is known to be a much robust species than *Aedes aegypti* and *Anopheles stephensi* the thermal fog formulations of Permethrin and Prallethrin could be easily applied in situations that warrants control of *Aedes* and *Anopheles* species of mosquitoes in epidemic situation.

CONCLUSION

Household synthetic pyrethroids show promise in thermal fog applications for vector control when used in Emulsifiable concentrate (EC) formulations. 2% w/w Permethrin and 2% w/w Prallethrin "Emulsifiable concentrates" when used in working dilutions of 1: 19 (1 part of 2% EC + 19 parts of diesel) results in greater than 80% knock-down and mortality against caged *Culex quinquefasciatus* at 5 minute exposure time. Studies with higher concentrations (2 parts of 2% EC +19 parts of diesel) would necessarily give higher knock down and mortality counts at lesser exposure time. The cost impact of increasing the concentration versus knock-down efficacy needs to be studied.

Table 1: Preparation of fogging premix

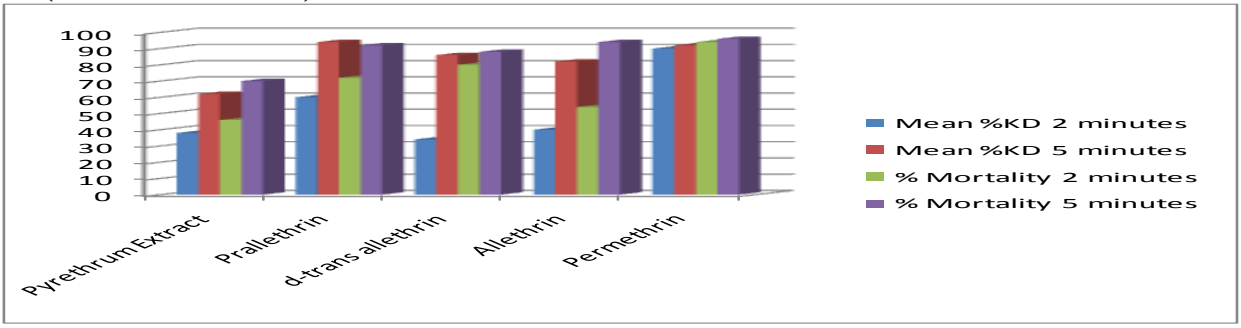
EC formulation	Amount (ml)	Diesel (ml)	Total volume (ml)
Pyrethrum extract (market sample)	50	950	**1000**
Prallethrin 2% w/w EC	50	950	**1000**
d-trans allethrin 2% w/w EC	50	950	1000
d-allethrin 2% w/w EC	50	950	**1000**
Permethrin 2% w/w EC	50	950	**1000**

EC= Emulsifiable concentrate

Table- 2. Knock-down of different Pyrethroids EC formulations against Culex quinquefasciatus.
(Summarized trial results of 2 sets of experiments (each set containing 10 bullets with5 mosquitoes each)

EC Formulations 2% w/w	Minutes of exposure			
	Mean % knock-down		Mean % mortality after 24 hours	
	2	5	2	5
Control	0	0	0	0
Pyrethrum extract	38	62	46	70
Prallethrin	60	94	72	92
d-trans allethrin	34	86	80	88
Allethrin	40	82	54	94
Permethrin	90	92	94	96

Figure -1 Graphical representation of mean % knockdown and mortality against various EC formulations and exposure times (2 minute and 5 minutes)



Y axis= Mean percentage Knock-down/Mortality (after 24 hours)
X axis= Various EC formulations with 2 minute and 5 minute exposure to thermal fog

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