Journal of the American Mosquito Control Association, 36(1):16–21, 2020

BIO-EFFICACY OF COMMERCIALLY AVAILABLE RESIDUAL INSECTICIDES FOR THE CONTROL OF *AEDES AEGYPTI* IN MEXICO

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ABSTRACT. Commercial aerosolized insecticides can be implemented as a community-based approach to targeted indoor residual spraying against *Aedes aegypti*, but their efficacy on pyrethroid-resistant mosquitoes has not yet been evaluated. Two commercial aerosolized products (H24 Poder Fulminante Ultra Eficaz®, carbamate, and Baygon Ultra Verde®, pyrethroid) were sprayed on common indoor surfaces e.g., cement, plywood, and cloth, and tested for their residual efficacy on susceptible and field-derived pyrethroid-resistant *Ae. aegypti* strains using the WHO cone bioassays. Overall, ≥80% 24-h mortality was observed for both products for at least 4 wk regardless of the mosquito strain or surface type used. H24 Poder Fulminante Ultra Eficaz showed the highest residual potency, sustaining >80% mortality for 7-wk posttreatment regardless of mosquito strain and surface type. For Baygon Ultra Verde, the mean mortality of female *Ae. aegypti* remained >80% for a shorter period (4–6 wk). Nonpyrethroid commercial aerosolized formulations can provide a lasting residual effect indoors compatible with the need for rapid and lasting mosquito control during outbreaks and may be suitable for community-based targeted indoor residual spraying.

KEY WORDS *Aedes aegypti*, aerosol, household insecticides, indoor residual spraying, Mexico, surface spraying

INTRODUCTION

In most tropical urban environments, *Aedes aegypti* L., a vector of dengue, chikungunya, and Zika, is commonly found indoors. Resting behavior of *Ae. aegypti* adults (including the epidemiologically important female population) inside homes is associated with lower heights (<1.5 m) and occurs behind/under furniture, inside wardrobes/closets, and in or around other dark shady objects/areas located frequently within dormitories (Dzul-Manzanilla et al. 2017, Perich et al. 2000). This behavioral trait opens the opportunity to target insecticide applications on such key *Ae. aegypti* resting sites.

House spraying with residual insecticides is a well-known method to kill insects, including mosquitoes, when they land on or crawl over treated surfaces (Rozendaal 1997). Indoor residual spraying (IRS) for *Ae. aegypti*, termed targeted IRS (TIRS) applied to predominantly intradomiciliary resting sites, has been recently recommended by the Vector Control Advisory Group of the World Health Organization to improve the control of *Ae. aegypti* (WHO 2016). This recommendation was essentially made for institutional programs responsible for vector control of *Aedes*-transmitted diseases. Unfortunately, the uptake of TIRS during the Zika outbreak was very limited, primarily due to the poor knowledge from vector control programs about this methodology within *Ae. aegypti* vector management, and the common assumption that this method is costly due to the heavy reliance on personnel who have to gain access to homes to perform interventions, 2 factors that could limit insecticide coverage in urban areas.

Indoor delivery of residual insecticides for TIRS can also be done with household insecticide products (HIPs; e.g., aerosol spray cans) and can be applied by homeowners as part of a community-based control strategy (Gartner et al. 2001, Pai and Hsu 2014). For this purpose, products have to effectively kill adult female mosquitoes at resting sites/surfaces and have a residual effect that is safe, commercially available, and used regularly by homeowners. Studies in the Mexican state of Yucatan have reported the magnitude of community use and expenses associated with HIPs for mosquito control. A large majority (>90%) of households report using commercial insecticides on a regular basis (Gray et al. 2018), from which insecticide canisters were the most common HIPs (70–90%) (Loroño-Pino et al. 2013, Rosecrans et al. 2014, Gray et al. 2018), with an estimated annual average expense per house of approximately $570.00 pesos (Loroño-Pino et al. 2013, Rosecrans et al., 2014), suggestive of an annual market in excess of $75 million pesos (>$5.7 million USD) only for Merida, the state capital (Loroño-Pino et al. 2014).

This study reports on the efficacy of commercially available HIPs applied on different indoor surfaces as residual surface sprays against adult female *Ae. aegypti* (susceptible and field-derived pyrethroid-resistant strains). Information on the efficacy of

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Table 1. Names and active ingredients of commercial aerosol insecticides available in the main supermarkets of Merida, Mexico. An asterisk (*) denotes commercial products selected for this study based on the residual effect information on the label.

Name	Ingredients	Manufacturer
H24 Poder Fulminante Ultra Eficaz®*	Propoxur = 4.60 g/kg, Tetrametrina = 1.03 g/kg, Fenvalerato = 4.55 g/kg	Industrias H24®
H24 Matacucarachas Acción prolongada contra insectos rastreros®	Propoxur = 1.57 g/kg, Prallethrin = 0.93 g/kg	Industrias H24®
H24 Doméstico Acción Inmediata®	Tetramethrin = 2.99 g/kg, Cifenothrin = 1.00 g/kg	Industrias H24®
H24 Poder Citronox®	Tetramethrin = 2.99 g/kg, Cifenothrin = 1.00 g/kg	Industrias H24®
Raid Max®	Cypermethrin = 1.0 g/kg, Imiprothrin = 2.5 g/kg	SC Johnson®
Raid Acción Total mata cucarachas, moscas y mosquitos®	Imiprothrin = 2.0 g/kg, Cypermethrin = 1.0 g/kg, Prallethrin = 0.3g/kg	SC Johnson®
Raid Matamoscas y mosquitos Aerosol®	Phenothrin = 1.2 g/kg, Prallethrin = 0.5 g/kg, Tetramethrin =1.1 g/kg	SC Johnson®
Baygon Ultra Verde®*	Cypermethrin = 1.0 g/kg, Imiprothrin = 0.5 g/kg	SC Johnson®
Baygon Total Insectos voladores y rastreros uso domestico®	Cyfluthrin = 0.015 g/kg, Imiprothrin = 0.05 g/kg	SC Johnson®
Oko®	Tetramethrin = 2.5 g/kg, Cifenothrin = 1.5 g/kg	Vishen de México®

commercial insecticides with the potential to control indoor-resting *Aedes* can help inform local communities and support integrated vector control programs. The recent renewed interest in TIRS implementation and community involvement make such studies important and appropriate.

MATERIALS AND METHODS

Insecticides/products selection

A search for commercially available products was carried out in the main supermarkets of Mérida (Table 1). Two commercial products, with a residual effect as stated on the label, were selected: 1) H24 Poder Fulminante Ultra Eficaz® (429 ml containing the carbamate propoxur [4.60 g/kg], the pyrethroid tetramethrin [1.03 g/kg], and fenvalerate [4.55 g/kg]), recommended for domestic control of household pests (cockroaches, spiders, ants, scorpions, bugs, and fleas) with a labeled residual effect for at least 30 days, and 2) Baygon Ultra Verde® (400 ml containing the pyrethroids cypermethrin [1 g/kg] and imiprotrine [0.5 g/kg], recommended for the control of mosquitoes, flies, cockroaches, ants, fleas, wasps, bed bugs, and moths) with a labeled residual effect of 6 wk. Both products are registered for household use at the Federal Commission for the Protection against Sanitary Risk of Mexico (H24 Poder Fulminante Ultra Eficaz RSCO-DOM-MEZC-1159-301-305-0.1915 Industrias H24, SA de CV, and Baygon Ultra Verde RSCO-MEZC-1167-0468-305-0.431 SC Johnson, S. de RL de CV) and considered within the toxicological category 5, or generally nontoxic (COFEPRIS 2019).

Residual application on surfaces

Based on the knowledge of common *Ae. aegypti* resting sites, the 3 different substrates/surfaces used in this study were cement with vinyl paint (typical construction material of walls), plywood (doors and furniture), and cloth (as in furniture, curtains, etc.). One-square-meter panels of each surface (acknowledging that our design involves pseudo-replicated data) were manufactured and placed on a wall at 1.5 m high. Between evaluations, they were stored in black bags in a dark room at an average temperature of 25°C and 80% RH.

Single blinding was used in this study in that the applicants did not know which brands of the aerosol they were administering. All cans were painted white and identified as A or B. Products were applied 30 cm from the point of application and discharged for a 10-sec period according to the label directions. Each panel of the different substrates was sprayed uniformly in horizontal strips with a flow of 1.9 g (SD = 0.15) per second to leave an estimated dose of 0.01 g per square meter. Panels were allowed to dry for 24 h before the first evaluation (see below).

Mosquito strains

Two strains of *Ae. aegypti* were used: the susceptible New Orleans (NO) strain and the field-derived San Lorenzo (SL) strain, which is resistant to pyrethroids (Vazquez-Prokopec et al. 2017, Deming et al. 2016). The NO strain was obtained from a colony established at the Collaborative Unit for Entomological Bioassays of the Autonomous University of Yucatan (UCBE-UADY) since 2012, originally provided by the Centers for Disease Control and Prevention (CDC), Atlanta, GA, USA. The SL strain was obtained from ovitraps placed at the study site and reared in the insectary of UCBE-UADY.

Assessment of residual activity

World Health Organization cone bioassays (WHO 2006) were used to monitor the residual efficacy of each product on sprayed panels and different

substrates. On each substrate type, 3 cones were affixed to evaluate the persistence of insecticides. Groups of 10 female *Aedes aegypti* of both strains (2–3 days old, provided only sugar solution and non-bloodfed) were introduced into the cones using mouth aspirators. A set of non-insecticide-treated substrates with the same dimensions and characteristics was used as control.

After being exposed for 30 min, mosquitoes were removed from the cones, returned to paper cups, kept under insectary conditions, and fed with a 10% sucrose solution. Mortality was recorded after 24 h. A mosquito was considered alive if it was able to fly. Bioassays were conducted after 24 h and once per week posttreatment. The WHO considers a mortality of 80% as the cutoff for effective insecticidal effect of indoor residual spraying (WHO 2006), so the number of weeks each product was effective was calculated. Bioassays were extended until mortality was $\leq$ 50% within all treatments, which occurred after a 3-month period. All bioassays were carried out at UCBE-UADY.

Data analysis

The mortality of mosquitoes at 24 h (mortality rate) was calculated as the proportion of dead mosquitoes against the total number exposed to treated surfaces. If the mortality of the control group was 5% to 20%, results were corrected by the Abbott formula (1925), and if this was more than 20%, tests were repeated. Mortality rates were calculated from the total number of dead and living mosquitoes across all replicates and stratified by strain, surface, and insecticide. The persistence of both insecticides on the surfaces was calculated from the percentage of mortality in mosquitoes exposed to different types of treated surfaces. All data were statistically analyzed using a Friedman chi square test ($\chi 2$) using SPSS v. 10.0 (SPSS Inc. Chicago, USA) to estimate the comparisons of mortality rates, paired by insecticide and type of surface. A P value < 0.05 was considered statistically significant.

RESULTS

Overall, both insecticide products provided $\geq$80% of effective insecticidal effect at least for 4 wk regardless of the strain and the surface type used (Table 2). No mortality was observed in the control groups. For H24 Poder Fulminante Ultra Eficaz the mean mortality of female *Aedes* mosquitos remained >80% during a 7-wk period posttreatment regardless of strain and surface type. As shown in Table 2, the efficacy of this product varied only slightly between surfaces and strains. The best residual efficacy (>80% mortality) and maximum duration of effectiveness (10 wk = 70 days) were observed on plywood with the *Ae. aegypti* susceptible strain (Table 2). The mean *Ae. aegypti* mortality can be summarized in the following order: plywood > cloth > cement for both susceptible and field-derived strains. Complete (100%) mortality was maintained with H24 Poder Fulminante Ultra Eficaz on the plywood surface in the first 5 wk (susceptible strain) or 6 wk (field-derived strain). Similarly, complete mortality was maintained for 4 (susceptible) or 5 (field-derived) wk in cement and cloth surfaces (Table 2).

For Baygon Ultra Verde, the mean mortality of female *Aedes* mosquitos remained >80% for a shorter period (4–6 wk) but at least for a month. As shown in Table 2, the efficacy of this product applied on common indoor surfaces varied slightly between surfaces and strains. The best residual efficacy (>80% mortality) and maximum duration of effectiveness (6 wk = 42 days) were observed on cloth with the *Ae. aegypti* field-derived strain. The longest complete mortality with Baygon Ultra Verde was observed also on cloth surfaces (4 wk) with the field-derived *Ae. aegypti* strain. Complete mortality was maintained for only 3–4 wk in cement and cloth surfaces, respectively (Table 2).

When mean *Ae. aegypti* mortality was compared between strains, H24 Poder Fulminante Ultra Eficaz showed best performance on all surfaces, with significantly ($P < 0.05$) higher mortalities of the susceptible strain from weeks 3 and 4 (painted cement $\chi 2 = 9.2–18.7$; plywood $\chi 2 = 8.2–12.4$; cloth $\chi 2 = 9.5–18.2$) and from weeks 3–5 in the pyrethroid resistant strain (painted cement $\chi 2 = 9.3–14.5$; plywood $\chi 2 = 8.7–17.2$; cloth $\chi 2 = 10.3–11.5$).

DISCUSSION

Both products evaluated here proved that, when applied according to label directions, they can be effective in the control of *Ae. aegypti* females when sprayed on surface materials typically used as indoor-resting sites. The 24-h mortality rates of both products persisted for at least 30 days, providing an important result with regards to the potential for HIPs to be used as part of a community-based TIRS implementation.

As far as we are aware, this is the first study evaluating the biological and residual efficacy of commercially available surface spray insecticides in Mexico for the control of *Ae. aegypti*, with cone bioassay tests and including a product formulated with a nonpyrethroid molecule H24 Poder Fulminante Ultra Eficaz: Propoxur 4.6 g/kg (carbamate) + Tetramethrin (pyrethroid) + Fenvalerate (pyrethroid). This product showed the highest residual activity and remained effective over 7 wk regardless of the strain and surface type. Furthermore, complete mortality was maintained during the first 5 wk after spraying.

Kuri-Morales et al. (2018) reported the efficacy of 13 household aerosol insecticides, when used as space sprays, against field-derived *Ae. aegypti* populations from Morelos, Mexico. That study followed a different study design and methodology than ours, e.g., designed to evaluate space spray

Table 2. Mean 24-h mortality (mean ± SD) of 2 *Ae. aegypti* strains exposed to 2 commercially available products in Mexico as sprayed on common indoor surfaces at various posttreatment intervals. Significant differences between HIPs ($P < 0.05$) are denoted by an asterisk (*) in the week of observation.

	Susceptible New Orleans strain			Field-derived pyrethroid-resistant San Lorenzo strain		
Time interval	H24 Poder total®	Baygon Ultra Verde®	$\chi 2$ (df = 3)	H24 Poder total®	Baygon Ultra Verde®	$\chi 2$ (df = 3)
Painted cement						
24 h	100 ± 0	100 ± 0	NS[1]	100 ± 0	100 ± 0	NS
1 wk	100 ± 0	100 ± 0	NS	100 ± 0	100 ± 0	NS
2 wk	100 ± 0	100 ± 0	NS	100 ± 0	100 ± 0	NS
3 wk	100 ± 0	100 ± 0	NS	100 ± 0	100 ± 0	NS
4 wk	100 ± 0	87.5 ± 25	12.33*	100 ± 0	87.5 ± 25	11.14*
5 wk	100 ± 0	70 ± 5	9.21*	97.5 ± 5	70 ± 5	14.53*
6 wk	90 ± 0	60 ± 9.6	18.73*	87.5 ± 2	65 ± 0	12.21*
7 wk	85 ± 5	60 ± 9.6	16.33*	82.5 ± 9.57	57.5 ± 5	11.73*
8 wk	80 ± 5	55 ± 3	11.71*	75 ± 5	35 ± 5	9.33*
9 wk	77.5 ± 8.6	50 ± 0	11.55*	75 ± 5	0	—
10 wk	62.5 ± 5	0	—	55 ± 5.8	0	—
11 wk	55 ± 3	0	—	22.5 ± 7.1	0	—
12 wk	53 ± 3	0	—	0	0	—
13 wk	40 ± 5	0	—	0	0	—
Plywood						
24 h	100 ± 0	100 ± 0	NS	100 ± 0	100 ± 0	NS
1 wk	100 ± 0	100 ± 0	NS	100 ± 0	100 ± 0	NS
2 wk	100 ± 0	100 ± 0	NS	100 ± 0	100 ± 0	NS
3 wk	100 ± 0	95 ± 0	12.45*	100 ± 0	80 ± 5	17.23*
4 wk	100 ± 0	90 ± 0	12.33*	100 ± 0	70 ± 5	15.23*
5 wk	100 ± 0	80 ± 5	11.02*	100 ± 0	62.5 ± 5	12.33*
6 wk	100 ± 0	70 ± 5	10.33*	92.5 ± 0	60.5 ± 9.6	11.21*
7 wk	92.5 ± 0	55 ± 3	9.32*	90 ± 0	55 ± 5.8	11.13*
8 wk	87.5 ± 25	50 ± 0	8.32*	75 ± 9.5	45.5 ± 5	8.75*
9 wk	80 ± 5	47.5 ± 5	8.26*	70 ± 5	0	—
10 wk	80 ± 5	0	—	60 ± 9.6	0	—
11 wk	72 ± 5	0	—	15 ± 10.1	0	—
12 wk	60 ± 9.6	0	—	0	0	—
13 wk	42.5 ± 5	0	—	0	0	—
Cloth						
24 h	100 ± 0	100 ± 0	NS	100 ± 0	100 ± 0	NS
1 wk	100 ± 0	100 ± 0	NS	100 ± 0	100 ± 0	NS
2 wk	100 ± 0	100 ± 0	NS	100 ± 0	100 ± 0	NS
3 wk	100 ± 0	94	18.21*	100 ± 0	100 ± 0	NS
4 wk	100 ± 0	87.5 ± 25	14.24*	100 ± 0	100 ± 0	NS
5 wk	100 ± 0	80 ± 5	12.33*	100 ± 0	97.5 ± 5	11.56*
6 wk	97.5 ± 0	72.5 ± 5	12.02*	92.5 ± 0	87 ± 5	11.21*
7 wk	92.5 ± 0	70 ± 5	12.06*	90 ± 0	62.5 ± 5	10.54*
8 wk	90 ± 0	70 ± 5	11.37*	75 ± 9.5	42.5 ± 5	10.33*
9 wk	80 ± 5	42.5 ± 5	9.53*	57 ± 5	0	—
10 wk	80 ± 5	0	—	55 ± 5.8	0	—
11 wk	60 ± 9.6	0	—	48.5 ± 5	0	—
12 wk	52.5 ± 5	0	—	0	0	—
13 wk	45 ± 5	0	—	0	0	—

[1] NS = not significant.

applications to caged mosquitoes located inside bedrooms. Kuri-Morales et al. (2018) reported that none of the products produced complete mortality (100%) after 30 min of exposure; only 3 products killed 100% of the individuals after a 24-h exposure. The authors discussed the relationship between low efficacy and the susceptibility status of their *Ae. aegypti* populations (which were field-derived and pyrethroid-resistant). Other factors that arguably could have an effect in efficacy were differences between active ingredients and formulations and the possible failure of the insecticide droplets to reach in a sufficient amount hidden places or surpass obstacles. Gray et al. (2018) did perform surface spray experiments on *Ae. aegypti*, selecting 2 commercial products containing tetramethrin, allethrin, and phenothrin (Raid® House and Garden, coded as "space spray formulation") and cypermeth-

rin and imiprothrin (Baygon Multi-Insect Killer, coded as "residual spray formulation"), and recorded 24-h mortality within 1 wk only. More information regarding efficacy/susceptibility to the formulations tested here was not available.

Although ISS with commercial aerosol-products has been recognized as a potential community-based vector control tool, few studies have explored its value (Ansari et al. 1997, Osaka et al. 1999, Samuel et al. 2017). Interior spraying of houses with residual insecticides as part of an *Ae. aegypti*–integrated vector management was implemented by the Tropical Public Health Unit in north Queensland, Australia. Interior spraying, in conjunction with source reduction and larval control, resulted in marked declines in local dengue transmission when implemented during epidemics (Hanna et al. 1998, 2001; Ritchie et al. 2002). Assays in experimental houses in Mexico have shown that TIRS on lower walls and typical *Ae. aegypti* resting sites achieves high efficacy (> 80% mortality for 4 months) with a significant reduction in insecticide use and duration of application in comparison to the "classic" IRS of all walls (Dunbar et al. 2019). However, the use of commercially available residual insecticide aerosol spray cans was not recommended for dengue vector control in the past, as it was thought that adult *Ae. aegypti* often rest on nonsprayable surfaces in houses (WHO 1997 cited by Samuel et al. 2017).

Some operational key points to be considered for a HIP include mosquito susceptibility to insecticides, suitability of surfaces for spraying, community perception, and appropriate insecticides (Rozendaal 1997). Householders in Yucatan already spray insecticides over specific household surfaces, with formulations containing the pyrethroids cypermethrin, cyfluthrin, and imiprothrin dominating as residual surface sprays, even when none of these products is marketed as targeting mosquitoes (Gray et al. 2018). As HIPs include multiple insecticide molecules, any assessment of their efficacy (or lack thereof) is complicated by the fact that it is difficult to identify which molecule(s) mosquitoes have lost susceptibility to. Regardless, our study and that of Gray et al. (2018) show that surface spraying of pyrethroid-only formulations leads to lower mortality on natural *Aedes* populations in comparison to carbamate formulations.

Populations of *Ae. aegypti* in Yucatan and Mexico are resistant to pyrethroids but are mainly susceptible to carbamates (Kuri-Morales et al. 2018). The commercial product with the carbamate propoxur evaluated here showed to be highly effective compared to the pyrethroid-based HIPs. Propoxur kills on contact but also has an airborne effect without repelling, and surfaces considered in this study, such as painted cement or plywood, are very appropriate nonabsorbent surfaces for spraying. While commercial products with new (nonpyrethroid) molecules can have higher efficacy when used by the community, their misuse can lead to rapid evolution of resistance (Gray et al. 2018). Therefore, considering HIPs within insecticide resistance management plans may be needed to achieve maximum entomological impact without jeopardizing the efficacy of insecticide molecules to which *Ae. aegypti* is susceptible.

This study provides baseline data that can be used as a guide in the ongoing TIRS pilot studies in Mexico. Further research on the efficacy of this and/or other HIPs applied for TIRS as interventions against *Ae. aegypti* should be considered. Although HIPs are not yet included in the list of insecticides/products for IRS programs in Mexico (or elsewhere), they represent a useful and easy method for householders living in high-density communities in modern cities where *Ae. aegypti* are common. A DIY (do-it-yourself) Targeted Indoor Residual Spraying (DIY-TIRS) action, where householders deliver residual treatment in their own homes using commercially available residual insecticide formulations, is an entirely novel approach that has not been evaluated previously for any vector-borne disease.

ACKNOWLEDGMENTS

This study was funded by a grant from CONACYT Mexico (Project no. 255141). We thank the entire team at Unidad Colaborativa para Bioensayos Entomologicos, Universidad Autonoma de Yucatan for their assistance. Mention of trade names or commercial products in this publication is solely for the purpose of providing specific information and does not imply recommendation or endorsement by the institutions involved in this study.

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