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EVALUATING THE ENVIRONMENTAL FRIENDLINESS OF ESSENTIAL OIL BASED INSECTICIDES

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Abstract: This study investigates the environmental impact of various essential oil-based products by comparing their efficacy, formulation stability, pollinator-friendliness, operator and consumer safety profiles, and the release of active ingredients into the environment. Our findings reveal significant differences across these factors, highlighting the need for more sustainable product development. The importance of this research is particularly relevant to the pest management market, especially for mosquito management. Essential oil products offer a promising alternative to conventional chemical pesticides, which are often stigmatized in the public perception. By focusing on naturally derived and easily registerable essential oil formulations, the pest management industry can address the growing demand for more sustainable solutions. This research aims to encourage the industry to prioritize eco-friendly practices in the creation of essential oil-based products, ultimately contributing to a healthier and more sustainable environment.

Key words: Essential oils, pollinator safety, mosquitoes, *Aedes aegypti*

INTRODUCTION

The environmental impact of pest control products is increasingly recognized by homeowners in recent years [Readex Research, 2024], particularly in urban settings where the demand for more sustainable solutions is growing. In several use cases, essential oil-based insecticides have emerged as promising alternatives to conventional chemical pesticides, offering a range of benefits including efficacy, formulation stability, pollinator-friendliness, and improved safety profiles for both operators and consumers.

As consumers look for alternatives to conventional pesticides, which often contain synthetic chemistries, products considered to be minimum risk pesticides, such as plant essential oil-based pesticides, are gaining popularity. The United States Environmental Protection Agency (EPA) has exempted such minimum risk pesticides from registration so long as they comply with the regulations in 40 C.F.R. § 152.25. Chemical products containing active and inert ingredients considered minimum risk are registered under Section 25(b) of the Federal Insecticide, Fungicide Rodenticide Act (FIFRA), and can be referred to as a FIFRA 25(b), or 25(b). chemical or pesticide. This offers an expedient and streamlined registration process for new natural based solutions and a quick path to market.

Many natural plant-derived essential oils have been recognized for their insecticidal properties and are considered environmentally friendly due to their natural origin and biodegradability [O'Neal *et al.*, 2019]. These essential oils are often composed of many individual biorational compounds like terpenes, alkaloids and fatty acids, which can result in multiple mode of action for a single oil [de Almeida, 2011]. Studies have demonstrated that essential oils such as sesame oil, cinnamon oil, clove oil, and thyme oil show biological activity against various mosquito species by acting as both repellents and insecticides [Luker *et al.,* 2023]. These compounds do not interact with the same target sites as conventional insecticides, making them effective against insecticide-resistant mosquito strains [Johnson *et al.*, 2023]. For instance, major constituents of Thyme oil are Eugenol and Thymol which have shown to impact the insect neural system [Yoon and Tak, 2022]. Similarly, trans-cinnamaldehyde is the most insect-repelling component in cinnamon bark [Choi *et al.,* 2022].

Conventional products in this minimum risk pesticide space are typically oil and water emulsions. These formulations have very high amounts of active ingredients, such as essential oils, required to achieve desired efficacy, and therefore rely heavily on essential oil content to control insects. Additionally, conventional essential oil concentrates and spray dilutions are not stable for extended periods of time. Phase separation occurs between aqueous and oils phases which heterogenizes the mixture causing uneven coverage. Volatilization of essential oil constituents can contribute to reduced efficacy. Further, 25(b) compliant formulations having high essential oil concentrations can have hazardous characteristics, as exemplified in many of the safety data sheets of these formulations, as essential oils can necessitate safety considerations when used in high amounts.

Finally, essential oil-based products are often perceived as generally safer and more environmentally friendly than conventional insecticides. Consequently, less tests are required to produce, market, register and sell products, especially under the "25(b)" exempt guidelines. Herein we present various aspects that should be considered when developing essential oil-based products highlighting formulation stability, reliable mosquito control, non-target species safety, low active ingredient output to the environment and overall safety to the applicator and customer. Specifically, we show that when used correctly, essential oils can effectively manage pest populations while posing minimal risks to non-target species, such as honey bees (e.g.: *Apis mellifera*) and butterflies (e.g.: *Vanessa cardui*).

MATERIAL AND METHODS

Spray dilution stability test. The spray dilution stability test compares Barricor Essential Mosquito Control (Barricor EMC) with two other "25(b)" product formulations containing conventional formulation ingredients comprising of only essential oils, emulsifiers, and water. Spray dilutions were prepared in 250mL glass cylinders by diluting the products in water at high label dilution rates. All dilutions were rigorously shaken equally before the beginning of the experiment. To record stability of the spray dilutions, pictures were taken 85min after preparation of the dilutions.

Barricor EMC contains: Sesame Oil 2.00%, Sodium Lauryl Sulfate 0.70%, Cinnamon Oil 0.67%, Clove Oil 0.67%, Thyme Oil 0.67%, and inert ingredients: Water, Acetyl Tributyl Citrate, Glycerin, Silica Gel, Tri-glycerol Monooleate, Xanthan Gum, Potassium Sorbate, Sodium Benzoate, Citric Acid. Compared product 1 contains: Sodium Lauryl Sulfate 11.0%, Geraniol 10.0%, Clove Oil 3.0%, Cornmint Oil 1.5%, and inert ingredients: Water, White Mineral Oil, Glyceryl Monooleate, and Sodium Alginate. Compared product 2 contains:

Rosemary 10% Geraniol 5% Peppermint Oil 2%, and inert ingredients: White Mineral Oil, Wintergreen Oil, Vanillin, Polyglyceryl Oleate.

Mosquito efficacy assay. This trial was performed by Snell Scientific. LLC in Meansville, GA. The experiment was performed in 6 replicates per treatment. Water was used as negative control. Ten mosquitoes (*Aedes aegypti*) were contained in 6" diameter cardboard rings covered with tulle mesh. Test substances, Barricor Essential Mosquito Control, was shaken and then mixed to the dilution rates of 2, and 4 fl. oz/gal water. A 2.5-gallon Stihl backpack sprayer was set on the 3rd flow rate setting with the misting screen on the applicator. Applications were made at a distance of 3' by spraying at an application rate of 1 gal/1000 ft^2. Mortality was observed 1 hr., 2 hrs., 4 hrs., and 24 hrs. after applications. The insects were transferred from the treatment arenas into the clean post-treatment arenas 1 hour after the applications.

Pollinator safety assays.Pollinator safety trials were performed by Eurofins Agroscience Services, LLC in Mebane, NC. Honey bees, (*Apis mellifera*) used in this study were sourced from Eurofins Agroscience Services, LLC. Cedar Grove Research Facility apiaries. This study was performed in 6 replicates per treatment, using between 20 and 26 honey bees per replicate. Honey bees were added to each test unit (32 oz container) which contained a single 90 mm diameter filter paper fixed to the bottom.

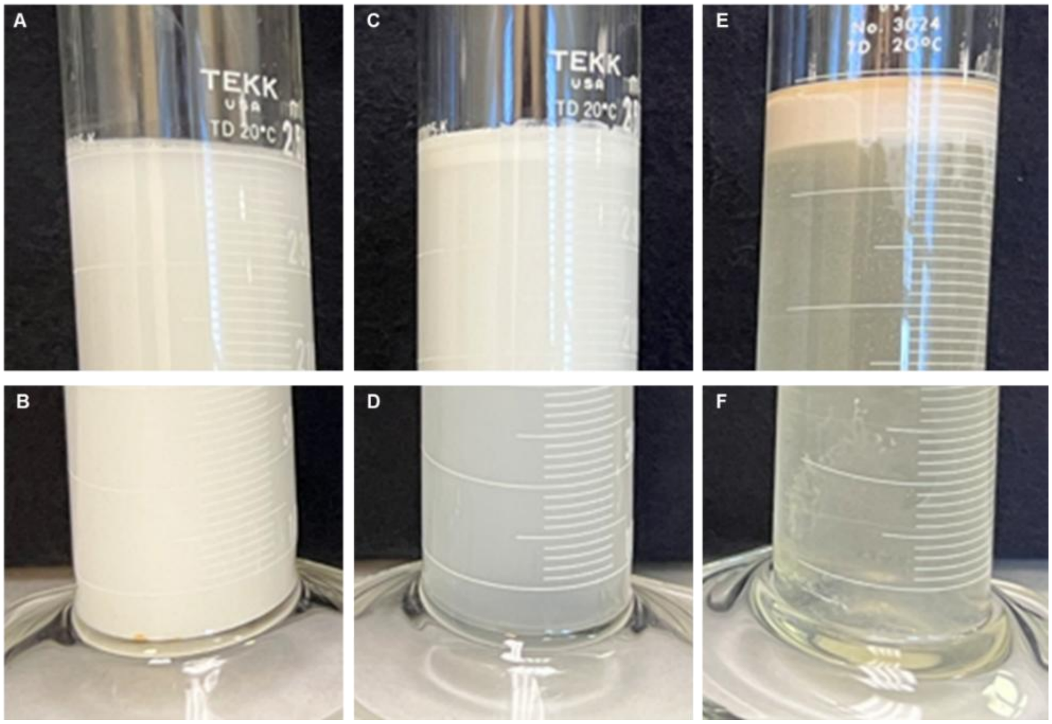
All stages of painted lady butterflies (*Vanessa cardui*) were obtained from Carolina Biological Supply Co. Bioassays were performed in 3 replicates per treatment. For larval bioassays each replicate included 6 healthy painted lady butterfly larvae. Adult bioassays each replication included 5 healthy painted lady butterfly adults. In bioassays, 32 oz containers with a single 90 mm diameter filter paper fixed to the bottom, was used to contain butterflies (adults and larvae) during application.

Direct spray bioassays were conducted to demonstrate the effects products may have when applied to non-target pollinators. Applications were done using 30 mL handheld sprayer. The application was adjusted to apply a rate of 1 gal/1000 ft^2 of product to the test unit area. The test solution was applied at 2, and 4 fl. oz/gal. Sprayed test units were left undisturbed 5 minutes before the insects were transferred to a new clean container. A control group was treated under the same parameters except water only was used. Honey bees were provided a syringe containing a 50% sucrose-water solution (w/v) for the duration of the experiment. Adult butterflies were provided a Petri dish containing a 25% sucrose-water solution (w/v) for the duration of the experiment. Larvae were returned to original containers, containing artificial larval diet. All test subjects were scored for mortality every 24 hours post-treatment up to 96 hours.

Chronic feeding exposure bioassays were conducted to demonstrate survivorship from continuous exposure through dietary consumption. The test solution was added to an artificial diet mix at 2 fl. oz/gal and 4 fl. oz/gal. Approximately 75 mL of diet was added to Petri dished and allowed to cool before larvae were introduced. The control group consisted of diet mixed with water only. Larvae were scored for mortality every 24 hours post-exposure up to 96 hours.

RESULTS

Spray dilution stability test. The spray dilution of Barricor EMC shows high level of stability and homogeneity over 85 min. No phase separation was visible in upper section (Figure 1A), as well as no discoloration at the lower section of the cylinder (Figure 1B). In contrast, the spray dilution of compared product 1 exhibit phase separation in upper section and discoloration in lower section of the cylinder (Figure 1C, 1D). The strongest phase separation was visible in the spray dilution of compared product 2 (Figure 1E, 1F).



Picture 1. Stability of spray dilutions in 250 mL glass cylinder. Homogeneity of spray dilutions (high label rates) of 3 essential oil-based products were evaluated after 85min. A) Upper and (B) lower section of Barricor EMC spray dilution, magnified. C) Upper and (D) lower section of compared product 1 spray dilution, magnified. E) Upper and (B) lower section of compared product 2 spray dilution, magnified.

Bioassay Results:

Barricor EMC exposure to mosquitoes resulted in 100% mortality after 24 hours in both 2 fl. oz/gal and 4 fl. oz/gal treatment concentrations at a volume of 1 gal/1000 ft^2 (Table 1).

Table 1: Average percent knock down (KD) and mortality of *Aedes aegypti* mosquitoes after direct spray application. The P-values, as a result of a student's t-test ($\alpha = 0.05$), for Barricor EMC (2 fl. oz/gal) treatments against the control in mosquitoes bioassays were 0.111, 0.175, 0.017, and $1x10^{-7}$ for 1, 2, 4, and 24 hours post application respectively. Additionally, Barricor EMC (4 fl. oz/gal) treatments against the control in mosquitoes bioassays resulted in P-values of 0.006, 0.012, 0.006, and 0. $1x10^{-7}$ for 1, 2, 4, and 24 hours post application respectively.

	1-hr	2-hr	4-hr	24-hr
Application at 1 gal/1000 ft^2	**% KD/Mortality** StDev	**% KD/Mortality** StDev	**% KD/Mortality** StDev	**% KD/Mortality** StDev
UTC	**0.0** 0.0	**0.0** 0.0	**0.0** 0.0	**5.0** 0.5
Barricor EMC (2 fl. oz/gal)	**10.0** 1.3	**7.0** 1.0	**15.0** 1.0	**100.0** 0.0
Barricor EMC (4 fl. oz/gal)	**22.0** 1.2	**10.0** 0.6	**18.0** 1.0	**100.0** 0.0

Survival of honey bees and painted lady butterflies (adults and larvae) treated with Barricor EMC was not significantly reduced compared to their respective control groups.

Direct spray of Barricor EMC at label rates to honey bees did not result in significantly different levels of mortality compared to the control for all evaluations (Table2).

Table 2. Average percent mortality of *Apis mellifera* honey bees after direct spray application. Student's t-test (α = 0.05) for Barricor EMC (2 fl. oz/gal) treatments against the control group in honeybee direct spray bioassays resulted in P-values of 0.45, 0.76, and 0.76 for 48, 72, and 96 hours post application respectively (Table 2). Student's t-test (α = 0.05) for Barricor EMC (4 fl. oz/gal) treatments against the control group in honeybee direct spray bioassays resulted in P-values of 1.0, 0.53, and 0.31, for 48, 72, and 96 hours post application respectively.

	24-hr	48-hr	72-hr	96-hr
Application at 1 gal/1000 ft^2	**% Survival** StDev	**% Survival** StDev	**% Survival** StDev	**% Survival** StDev
UTC	**100.0** 0.0	**96.9** 2.4	**94.6** 3.5	**89.9** 9.7
Barricor EMC (2 fl. oz/gal)	**100.0** 0.0	**95.2** 4.3	**93.5** 5.1	**91.1** 8.0
Barricor EMC (4 fl. oz/gal)	**100.0** 0.0	**96.7** 5.2	**95.9** 4.9	**94.2** 4.9

Direct spray of Barricor EMC at label rates to *Vanessa cardui* painted lady butterfly larva (Table 3) did not result in mortality.

Table 3. Efficacy of Barricor EMC at label rates to *Vanessa cardui* painted lady larva as a direct spray application. The P-values, as a result of a student's t-test (α = 0.05), for Barricor EMC (2 fl. oz/gal) treatments against the control in painted lady butterfly larval direct spray bioassays were 0.45, 0.76, and 0.76 for 48, 72, and 96 hours post application respectively. The P-values, as a result of a student's t-test (α = 0.05), for Barricor EMC (4 fl. oz/gal) treatments against the control in painted lady butterfly larval direct spray bioassays were 1.0, 0.53, and 0.31, for 48, 72, and 96 hours post application respectively.

	24-hr	48-hr	72-hr	96-hr
Application at 1 gal/1000 ft^2	**% Survival** StDev	**% Survival** StDev	**% Survival** StDev	**% Survival** StDev
UTC	**100.0** 0.0	**100.0** 0.0	**100.0** 0.0	**100.0** 0.0
Barricor EMC (2 fl. oz/gal)	**100.0** 0.0	**100.0** 0.0	**100.0** 0.0	**100.0** 0.0
Barricor EMC (4 fl. oz/gal)	**100.0** 0.0	**100.0** 0.0	**100.0** 0.0	**100.0** 0.0

Direct spray of Barricor EMC at label rates to *Vanessa cardui* painted lady adults (Table 4) did not result in high levels of mortality compared to the control for all evaluations.

Table 4. Efficacy of Barricor EMC at label rates to *Vanessa cardui* painted lady adults as a direct spray application. Barricor EMC (4 fl. oz/gal) treatment on adult painted lady butterflies resulted in P-values of 0.42, 0.42, and 0.42 for hours 48, 72, and 96 respectively according to a student's t-test ($\alpha = 0.05$).

	24-hr	48-hr	72-hr	96-hr
Application at 1 gal/1000 ft^2	**% Survival** StDev	**% Survival** StDev	**% Survival** StDev	**% Survival** StDev
UTC	**100.0** 0.0	**100.0** 0.0	**100.0** 0.0	**100.0** 0.0
Barricor EMC (2 fl. oz/gal)	**100.0** 0.0	**100.0** 0.0	**100.0** 0.0	**100.0** 0.0
Barricor EMC (4 fl. oz/gal)	**100.0** 0.0	**93.3** 11.5	**93.3** 11.5	**93.3** 11.5

In the chronic feeding exposure bioassays (Table 5) control larval survival was reduced to 94% after 48 hours, while larval survival in both treatment groups remained 100%. This reduction in control survival was not a limitation in the data due to the insignificance of this value.

Table 5. Efficacy of Barricor EMC at label rates to *Vanessa cardui* painted lady larva in continuous oral uptake assay. Oral uptake (continuous exposure *ad libitum*) of Barricor EMC at label rates by painted lady butterfly larva (Table 5) did not result in increased levels of mortality.

	24-hr	48-hr	72-hr	96-hr
Application	**% Survival** StDev	**% Survival** StDev	**% Survival** StDev	**% Survival** StDev
UTC	**100.0** 0.0	**94.4** 9.6	**94.4** 9.6	**94.4** 9.6
Barricor EMC (2 fl. oz/gal)	**100.0** 0.0	**100.0** 0.0	**100.0** 0.0	**100.0** 0.0
Barricor EMC (4 fl. oz/gal)	**100.0** 0.0	**100.0** 0.0	**100.0** 0.0	**100.0** 0.0

Table 6: List of products targeting mosquitoes. Information regarding signal words, dilutions rates and composition derived from product labels and safety data sheets (SDS). *Sodium Lauryl Sulfate.

Product name	Barricor Essential Mosquito Control	Compare product 3	Compare product 4	Compare product 5
Signal word	Label: no signal word SDS: no signal word	Label: Caution SDS: Warning	Label: Caution SDS: Danger	Label: Caution SDS: Danger
Dilution rates	2-4 fl. oz / gal	1-8 fl. oz / gal	0.33-0.66 fl. oz / gal	6-16 fl. oz / gal
Essential oils in product (%)	Sesame Oil: 2.00% SLS*: 0.70% Cinnamon Oil: 0.67% Clove Oil: 0.67% Thyme Oil: 0.67%	Geraniol: 10.0% Cinnamon Oil: 5.0% SLS*: 3.3% Cornmint Oil: 2.0%	Soybean oil: 26.3% Clove oil: 13.2% Citronella oil: 10.5% 2-Phenylethyl propionate: 10.5% Lemongrass oil: 7.9%	Lemongrass oil: 5.0% Geraniol: 4.5% Castor oil: 6.0% Cedarwood oil: 3.0% SLS*: 3.0% Corn oil: 0.5%
Amount % of AI in spray dilution	0.07% to 0.15%	0.32% to 1.27%	0.18% to 0.35%	0.31% to 0.83%

This novel formulation concept ensures a stable spray dilution over time, which is essential for consistent and reliable product performance, as phase separation within the spray dilution can result in diminished product efficacy. Furthermore, this formulation concept allows for a substantial reduction in active ingredient levels compared to other "25(b)" products (Table 6), thereby aiming to minimize environmental impact by limiting the quantity of active ingredients released into the environment. Barricor EMC was classified as “not a hazardous substance or mixture” when tested for product safety. Consequently, no signal words are required on the product label or safety data sheet (SDS), exceeding market safety standards. In contrast, the labels and SDS of comparable products (Table 6) necessitate signal words such as caution, warning, or danger.

In conclusion, Barricor EMC demonstrates excellent efficacy in direct spray application against mosquitoes while showing no significant negative impact on the survival of pollinators, such as adult honey bees and butterfly larva and adults, when used as directed at label rates.

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