



NON-GLP STUDY REPORT

Date: July 2019

Sponsor: Shanghai Daisy, LLC.
Study: Fountain Mosquito Repellency 19-2
Trial: AEDSAE
Sponsor Code: N/A
Test Method: 414

REPORT TITLE

Repellency of the Aquaflame Color Changing Citronella Fountain With High Purity Citronella Oil
Against Yellow Fever Mosquitoes (*Aedes aegypti*)

STUDY

Fountain Mosquito Repellency 19-2

TRIAL

AEDSAE

SPONSOR CODE

N/A

EXPERIMENTAL START DATE

June 7, 2019

EXPERIMENTAL COMPLETION DATE

June 12, 2019

REPORT DATE

July 23, 2019

TEST FACILITY/AUTHORS

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SPONSOR

Shanghai Daisy, LLC.



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STUDY OBJECTIVE(S):

To evaluate the repellency associated with the test substances against yellow fever mosquitoes (*Aedes aegypti*).

TEST SUBSTANCE INFORMATION:

#	Test Substance	Active Ingredient	Lot and/or Batch #	Snell Receipt Code
1	Control-Untreated	N/A	N/A	N/A
2	Aquaflame Color Changing Citronella Fountain	Citronella Oil (High Purity)	N/A	031419-1-A-SHA & 060319-1-A-SHA

TEST SYSTEM INFORMATION:

Trial	Test System	Strain	Stage/Age	Source
AEDSAE	Yellow Fever Mosquito (<i>Aedes aegypti</i>)	Lab	Adults Females (4+ days old)	Lab Reared

TEST DESIGN:

# Reps per Substance	# Systems per Rep	# Systems per Substance
4	135	540

This work was conducted under sound scientific principles but does not comply with GLP standards under 40 CFR 160



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MATERIALS AND METHODS:

The following is the Snell Scientifics Standardized Testing Method for measuring area-wide mosquito repellency when using a human surrogate as an attractant in an indoor setting.

Materials:

- Test Building: 12'W x 20'L.
- Mosquito Cage: 6'W x 12'L x 6'H cage.
- Bait Cage Area: 4.5'W x 4.5'L x 3.25'H cage. The bait cage is used for housing the mosquito trap/attractant.
- Mosquito Attractant: A CO₂ generating mosquito attractant (Mosquito Magnet® Patriot Plus and propane tank) was used as a surrogate human to attract the mosquitoes.

Methods:

- One hundred and thirty-five (135) female mosquitoes were initially released inside the cage (prior to test start).
- The windows and door of the test building were closed during testing.
- The mosquitoes received a 15-minute acclimation period before starting each test replicate.
- The mosquito trap was activated and allowed to warm up for at least 15-minutes prior to the start of each replicate.
- The trap was positioned inside the bait cage area and a cloth was draped in place between the trap and the researcher.
- The cloth was equipped with a clear acrylic viewing window, which allowed for the researcher to monitor the number of mosquitoes on the screen areas on the bait cage during the test.
- Prior to the start of the pre-treatment period, fresh distilled water was added to an empty fountain. 10 drops of the supplied citronella oil were added to the water contained in the fountain. Ten minutes into the pre-treatment period, the fountain was activated (lights left inactive) outside of the building to allow for a 15-minute pre-use activation period.
- Observations of the number of mosquitoes that are attracted to the trap were recorded every 5-minutes for 25-minutes during the control portion of the study. This was done by recording the total mosquitoes on the left, right, and front sides of the bait cage area.
- Following the untreated control portion, the activated fountain was placed in the cage on the floor under the front panel.
- The total mosquitoes on the left, right, and front sides of the bait cage were then recorded every 5 minutes for 60-minutes.
- After the treatment replicate was terminated, the trap was turned off and removed from the bait cage and the test substance was removed from the cage.

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TABLES:

Table 1.

% Reduction in Activity- Yellow Fever Mosquitoes (<i>Aedes aegypti</i>)													
Test Substance:	Minutes After Exposure												0-60 min AVG
	5	10	15	20	25	30	35	40	45	50	55	60	
Untreated Control	9%	13%	14%	23%	9%	18%	8%	9%	15%	17%	21%	23%	15%
Aquaflame Color Changing Water Fountain + 10 drops of High Purity Citronella Oil	19%	7%	8%	18%	21%	32%	35%	38%	49%	46%	36%	42%	29%

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APPENDIX A: PHOTOGRAPHS

Photograph 1. Aquaflame Color Changing Fountain



Photograph 2. Citronella Oil (High Purity)



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Photograph 3. Mosquito Magnet in Mesh Bait Cage



Photograph 4. Test Systems on Bait Cage Screen





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Photograph 5. Fountain During Treatment Period

