

TITLE

Results of the Honey Bee (*Apis mellifera*) Toxicity of
Residues on Foliage (RT25) Ring Study: Phase II

TEST GUIDELINE

NA

AUTHOR(S)

Stephen L. Clark

STUDY PUBLICATION DATE

June 9, 2022

PERFORMING LABORATORY

Pacific EcoRisk
2250 Cordelia Road
Fairfield, CA 94534

LABORATORY PROJECT ID

NA

SPONSOR

Pollinator Research Task Force, LLC
c/o Landis International, Inc.
3185 Madison Highway
Valdosta, GA 31601

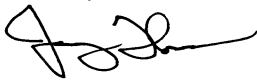
PAGE COUNT

282

STATEMENT OF NO DATA CONFIDENTIALITY CLAIM

No claim of confidentiality, on any basis whatsoever, is made for any information contained in this document. I acknowledge that information not designated as within the scope of FIFRA sec. 10(d)(1)(A), (B), or (C) and which pertains to a registered or previously registered pesticide is not entitled to confidential treatment and may be released to the public, subject to the provisions regarding disclosure to multinational entities under FIFRA 10(g).

Submitter Signature:



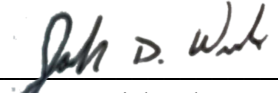
Jamey Thomas
Chair, PRTF Administrative Committee
Pollinator Research Task Force, LLC

Date: June 14, 2022

GOOD LABORATORY PRACTICE COMPLIANCE STATEMENT

This study was not conducted in accordance with 40 CFR 160.

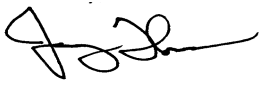
Sponsor Representative Signature:



Joseph D. Wisk, Ph.D., DABT
Senior Manager, Ecotoxicology
Pollinator Research Task Force, LLC

Date: June 13, 2022

Submitter Signature:



Jamey Thomas
Chair, PRTF Administrative Committee
Pollinator Research Task Force, LLC

Date: June 14, 2022

Results of the Honey Bee (*Apis mellifera*) Toxicity of Residues on Foliage (RT₂₅) Ring Study: Phase II

Prepared For:

Company Name: POLLINATOR RESEARCH TASK FORCE

Company Number: 92149

MRID# 51895501

Prepared By:

Pacific EcoRisk

2250 Cordelia Road

Fairfield, CA 94534

June 9, 2022



STUDY IDENTIFICATION AND APPROVAL

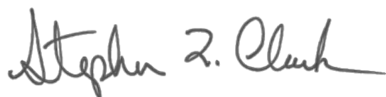
Sponsor: Pollinator Research Task Force, LLC
c/o Landis International, Inc.
3185 Madison Highway
Valdosta, GA 31601
U.S.A.

Sponsor Representative Joseph D. Wisk, Ph.D.
BASF Corporation

Independent Data Consultant: Pacific EcoRisk
2250 Cordelia Road
Fairfield, CA 94534

Study Personnel: Stephen L. Clark
Pacific EcoRisk

This report is approved and issued by:



Stephen L. Clark
Technical Manager
Pacific EcoRisk

6/9/22
Date

Executive Summary

Residual toxicity data for bees are generated through the Toxicity of Residues on Foliage Test (OCSPP Guideline 850.3030) and are referred to as RT₂₅ data. The RT₂₅ is the time required for pesticide residues to decline on the foliage of a treated crop such that mortality to adult honey bees (*Apis mellifera*) exposed to the treated foliage for 24 h is $\leq 25\%$. The RT₂₅ is intended to be a measure of the time that the pesticide formulated product is expected to remain toxic to bees in the field when sprayed at the maximum application rate. Traditionally, the residual toxicity (RT₂₅) information has been considered useful to growers and beekeepers to ensure bee safety, as it can help them determine the appropriate amount of time between pesticide application and increased bee activity. While compiling and reviewing the available RT₂₅ data, the United States Environmental Protection Agency (EPA) identified inconsistencies and variability in RT₂₅ values between formulated products of the same pesticide active ingredient. EPA also noticed that these data did not appear to be correlated with chemical/physical characteristics of the pesticide active ingredient. The Pollinator Research Task Force (PRTF), in collaboration with EPA, has taken the task to review the current test design (OCSPP Guideline 850.3030), work with different stakeholders to improve the method, and ensure the reliability and predictive nature of RT₂₅ data.

The PRTF formed a Ring Test Committee comprised of individuals from academia, government, and industry that reviewed RT₂₅ data from different products containing the same active ingredients and hypothesized that the major source of variability was related to the test design, since OCSPP 850.3030 does not adequately specify various test parameters which could influence exposure, leaving room for interpretation by the testing laboratories. Based on the PRTF Ring Test Committee review, a project was developed in two phases: short-term improvements (Phase I) and long-term improvements (Phase II). The initial Phase I effort focused on increased ‘standardization’ of the test guideline. The purpose of Phase I was to address potential short-term improvements and evaluate the agreed-upon methodology for a ring test in 2020, with a goal of both standardizing test conditions for the OCSPP 850.3030 protocol and evaluating whether more reliable and consistent data are produced. Results of the Phase I study were still unacceptably variable, and indications were that the applications in the field could be a major source of variability in the tests.

The Phase II study reported here was designed to control for two sources of variability, 1) differences in application equipment which could potentially lead to inconsistent distribution of the test substance over the treated plots, and 2) differences in environmental conditions which could result in different dissipation/degradation rates in the treated plots. Eurofins Agrosience Services and Smithers Viscient were selected for the Phase II study since they are within close geographic proximity to each other in North Carolina. The two facilities coordinated the timing of crop planting and dimethoate (selected as a reference chemical) applications to occur within a two-week window. The coordinated planting and applications at each test facility occurred at different times during the year to evaluate the impact of environmental conditions in the field on the bee bioassay results. The first coordinated event at each facility occurred in June when the

weather is typically hot with low humidity. The second coordinated event occurred in September when the weather is typically humid.

For each event, a single application of the test substance was applied by each facility using calibrated hand-held boom sprayers with standard nozzles at a rate of 0.5 lb active ingredient/acre in 200 L/ha of spray volume under natural field conditions to alfalfa (*Medicago sativa*, 20-40 cm in height). Similarly, control crop foliage was treated with water only. Sufficient alfalfa was obtained from the treatment and control plots and split to allow each laboratory to test both their own alfalfa and that from the other laboratory.

The bee exposures followed method standardizations implemented in the PRTF Phase I study. Mortality (i.e., when organism was completely immobile), appearance, and behavior were recorded at 4 ± 1 -h and 24 ± 1 -h post exposure for each specified weathering interval. When control-corrected honey bee mortality was greater than 25% at any timepoint (i.e., 6-h post weathered foliage exposure), weathered foliage samples continued to be harvested and tested for up to two days post application.

Both facilities submitted samples of tank mix solutions, treated alfalfa, and spray cards to EN-CAS (Winston-Salem, NC) for dimethoate residue analyses. Tank mix solutions were analyzed by high performance liquid chromatograph coupled to an ultraviolet light absorbance detector (HPLC-UV), while alfalfa and spray card analyses were conducted using gas chromatography coupled to a flame photometric detector (GC-FPD).

All laboratory data (biological observations, environmental conditions, and mortality observations) were submitted to Pacific EcoRisk (PER), who was contracted by the PRTF to anonymize the data (i.e., Lab A and Lab B), review the data for adherence to the study protocol and statistically analyze the data to generate RT_{25} values for each laboratory.

All field application conditions in the PRTF Ring Test Protocol were acceptable. All bee exposure environmental conditions were acceptable for Lab A. The bee exposures for Lab B met the study exposure environmental conditions, except that humidity in the June trial ranged from 36–53% rather than 50-80%.

The spray tank results for the two facilities were within 10% for the June samples and 24% for the September samples. The mean dimethoate concentration on facility A spray cards was 109% greater than facility B spray cards for the June application, but the mean concentration was identical for both facilities for the September application. The dimethoate concentration on alfalfa generally decreased over application intervals for four of five sets of alfalfa tested (including the duplicate analysis).

The RT_{25} values determined by both labs were similar for each facility's June applications alfalfa and for the facility B alfalfa September application (Table 13). The RT_{25} values determined by both labs for facility A September alfalfa were similar, but these values were ~3-5x lower than

all other RT₂₅ values. The tank mix solution, spray card, and alfalfa dimethoate analyses do not explain the reduced RT₂₅ values obtained by both labs for the facility A September application alfalfa.

This study demonstrates that consistent test results can be obtained for two labs testing the same alfalfa after controlling for application equipment and environmental conditions during dimethoate application.

Table of Contents

	Page
EXECUTIVE SUMMARY	ii
1. INTRODUCTION	1
2. PROCEDURES	3
2.1 Laboratory Data and Report Anonymization	4
2.2 Statistical Analysis Methods	4
3. RESULTS	5
3.1 Environmental Conditions.....	5
3.2 Dimethoate Chemical Analyses of Spray Tank Solutions, Spray Cards, and Treated Alfalfa	5
3.2.1 Dimethoate Analyses of Spray Tank Solutions.....	5
3.2.2 Dimethoate Analyses of Spray Cards.....	6
3.2.3 Dimethoate Analyses of Treated Alfalfa.....	7
3.3 Toxicity of Dimethoate to Adult Honey Bees.....	8
3.3.1 Facility A June Alfalfa Application Tested By Lab A	8
3.3.2 Facility A June Alfalfa Application Tested By Lab B	8
3.3.3 Facility B June Alfalfa Application Tested By Lab A	9
3.3.4 Facility B June Alfalfa Application Tested By Lab B	9
3.3.5 Facility A September Alfalfa Application Tested By Lab A	10
3.3.6 Facility A September Alfalfa Application Tested By Lab B	10
3.3.7 Facility B September Alfalfa Application Tested By Lab A	11
3.3.8 Facility B September Alfalfa Application Tested By Lab B.....	11
4. SUMMARY AND CONCLUSIONS	12

Appendices

Appendix A	Pollinator Research Task Force Ring Test Protocol
Appendix B	Anonymized Data Submittal - Lab A
Appendix C	Anonymized Data Submittal - Lab B
Appendix D	Dimethoate Analysis Report for Spray Tank Solutions, Spray Cards, and Treated Alfalfa
Appendix E	Summary of Statistics for the Toxicity of Facility A June Alfalfa Application Tested By Lab A
Appendix F	Summary of Statistics for the Toxicity of Facility A June Alfalfa Application Tested By Lab B
Appendix G	Summary of Statistics for the Toxicity of Facility B June Alfalfa Application Tested By Lab A
Appendix H	Summary of Statistics for the Toxicity of Facility B June Alfalfa Application Tested By Lab B
Appendix I	Summary of Statistics for the Toxicity of Facility A September Alfalfa Application Tested By Lab A
Appendix J	Summary of Statistics for the Toxicity of Facility A September Alfalfa Application Tested By Lab B
Appendix K	Summary of Statistics for the Toxicity of Facility B September Alfalfa Application Tested By Lab A
Appendix L	Summary of Statistics for the Toxicity of Facility B September Alfalfa Application Tested By Lab B

1. INTRODUCTION

Residual toxicity data for bees are generated through the Toxicity of Residues on Foliage Test (OCSPP Guideline 850.3030) and are referred to as RT₂₅ data. The RT₂₅ is the time needed for pesticide residues to decline on the foliage of a treated crop, such that adult honey bee (*Apis mellifera*) mortality is $\leq 25\%$ following exposure to the treated foliage for 24 h. The RT₂₅ is intended to be a measure of the time that the pesticide formulated product is expected to remain toxic to bees in the field when sprayed at the maximum application rate. Based on the United States Environmental Protection Agency (EPA) regulations for requiring ecological effects data related to impacts on non-target organisms (40 CFR 158.630), the EPA has typically determined whether RT₂₅ data are needed based on the results of the adult honey bee acute contact toxicity test (OCSPP Guideline 850.3020); the toxicity of residues on foliage study is triggered if one or more active ingredients within the formulation have a median lethal dose to 50% of the bees tested (LD₅₀) of less than 11 µg/bee and the use pattern(s) indicate(s) that honey bees may be exposed to the pesticide. This study is conditionally required in South Korea and an RT₂₅ study is required in Brazil for products applied by spray and whose contact LD₅₀ is <11 µg active ingredient/bee (IBAMA Bee Normative).

Traditionally, the RT₂₅ value has been considered useful to growers and beekeepers to ensure bee safety, as it can help them determine the appropriate amount of time between pesticide application and increased bee activity. While compiling and reviewing the available RT₂₅ data, EPA identified inconsistencies and variability in RT₂₅ values between formulated products of the same pesticide active ingredient. EPA also noticed that these data did not appear to be correlated with chemical/physical characteristics of the pesticide active ingredient. The Pollinator Research Task Force (PRTF), in collaboration with EPA, has taken the task to review the current test design (OCSPP Guideline 850.3030), work with different stakeholders to improve the method, and ensure the reliability and predictive nature of RT₂₅ data.

The PRTF formed a Ring Test Committee comprised of individuals from academia, government, and industry that reviewed RT₂₅ data from different products containing the same active ingredients and hypothesized that the major source of variability was related to the test design, since OCSPP 850.3030 does not adequately specify various test parameters which could influence exposure, leaving room for interpretation by the testing laboratories. As a result, different laboratories conducting these studies include different parameters in their study protocols. During the initial review of the current study design, the PRTF Ring Test Committee identified the potential sources of variability in the RT₂₅ data (Table 1).

Table 1. Sources of Variability in RT₂₅ data Identified by the PRTF Ring Test Committee	
Category	Description
Laboratory Test	Use of variable test cage sizes which can lead to inconsistent exposures
	Placement of treated foliage in cages
	Lack of a true positive control (reference toxicant)
	Current residue aging intervals (i.e., 3, 8, and 24 h post application) do not fit well with the EPA's Acute Risk Mitigation Policy. New protocols need to include 6 h as one of the weathering intervals
Field	Crop grown in field versus grown in flats in greenhouses
	Variable age of foliage used in the test
	The type of alfalfa used, including smooth vs. hairy types, and erect vs. creeping
	Product application in the field versus application in lab using a spray booth
	No recommendation for environmental parameters during weathering in the field
	No guidance on whether surfactants should or should not be used

Based on the PRTF Ring Test Committee review, a project was developed in two phases: short-term improvements (Phase I) and long-term improvements (Phase II). The initial Phase I effort focused on increased 'standardization' of the test guideline. The purpose of Phase I was to address potential short-term improvements and evaluate the agreed-upon methodology for a ring test in 2020, with a goal of both standardizing test conditions for the OSCPP 850.3030 protocol and evaluating whether more reliable and consistent data could be produced. Results of the Phase I study were still unacceptably variable, and indications were that the applications in the field could be a major source of variability in the tests.

The Phase II study reported here was designed to control for two sources of variability, 1) differences in application equipment which could potentially lead to inconsistent distribution of the test substance over the treated plots, and 2) differences in environmental conditions which could result in different dissipation/degradation rates in the treated plots. The Phase II Ring Test Protocol based upon the OSCPP 850.3030 protocol was titled "Standardization of Honey Bee Toxicity of Residues on Foliage (RT₂₅)". In addition to the Ring Test Committee, contributions to the Ring Test Protocol were provided by the PRTF members (Joseph Wisk, Daniel Schmel, Bibek Sharma, Timothy Joseph, Max Feken, and Verissimo Sa). Two contract research laboratories were selected to participate in the in-life phase of the ring test (i.e., field applications and bioassays): Eurofins Agrosience Services (Mebane, NC; John Porch) and Smithers Viscient (Snow Camp, NC; Alison Warmkessel). Both laboratories submitted samples of tank mix solutions, treated alfalfa, and spray cards to EN-CAS (Winston-Salem, NC) for dimethoate residue analysis.

The PRTF contracted Pacific EcoRisk (PER) to analyze and review data submitted by the laboratories that participated in the Phase II foliage residue ring test. This report details the laboratory methods, results from each laboratory (anonymized), and a statistical evaluation of the inter-laboratory results.

2. PROCEDURES

The test methods used in conducting this study followed OSCPP 850.3030 with modifications established by the PRTF's Ring Test Protocol (Appendix A), briefly described below. Dimethoate active ingredient (AI, 43.5% nominal purity), an organophosphate pesticide, served as the reference chemical/test substance for this study. The two test facilities are within close geographic proximity to each other in North Carolina and coordinated the timing of crop planting and dimethoate applications to occur within a two-week window. The coordinated planting and applications at each test facility occurred at two different times during the year to evaluate the impact of environmental conditions in the field on the bee bioassay results. The first coordinated event at each facility occurred in June (June) when the weather is typically hot with low humidity. The second coordinated event occurred in September (September) when the weather is typically humid.

For each event, a single application of the test substance was applied by each facility using calibrated hand-held boom sprayers with standard nozzles at a rate of 0.5 lb active ingredient/acre in 200 L/ha of spray volume under natural field conditions to alfalfa (*Medicago sativa*, 20-40 cm in height). Similarly, control crop foliage was treated with water only. The spray tank solutions were continuously stirred/circulated prior to and during use. Nozzle height above the crop was maintained consistent with the manufacturer recommendations and coordinated between the two facilities for consistency. Average wind speed was less than 3 m/sec during application, and dimethoate was applied on clear days with a maximum temperature of 20-40°C and <30% chance of precipitation.

At a minimum, nine test substance treatment plots were used to obtain three plots for harvesting at each time interval (i.e., 6±1-h and 24±1-h post application). Approximately 180 g fresh weight or 6,000 cm² total foliage was harvested from randomly selected control (i.e., untreated) and test substance treatment plots. Half of the harvested foliage was transported to each of the laboratories in bags placed in coolers held at 8–12°C. At the laboratories, the foliage was thoroughly mixed and then divided into approximately 15 g or 500 cm² portions cut into 12-15 cm lengths.

Test cages for this study were comprised of transparent 32-oz plastic containers (upper diameter = approx. 11 cm, base diameter = approx. 9 cm; height = approx. 14 cm) with a suitable opening for the introduction of foliage and bees, and another opening at the top for inserting the feeding syringe/tube. Six replicate cages per treatment were each loaded with 15 g of foliage placed upright/diagonally to maximize the exposure. Twenty-five young adult worker bees were

introduced to each cage, with bees being 3-5 days post emergence and acclimated in an incubator for approximately 24 hours before the introduction of foliage. The bees were fed *ad libitum* a 50% weight/volume (w/v) or weight/weight (w/w) solution of sugar/water (500 g/L) throughout the holding and test period.

Exposures were performed indoors in an incubator under controlled lighting and environmental conditions. Temperature and relative humidity during the exposure phase were maintained between 25 and 35°C and 50% and 80%, respectively, and in total darkness. Mortality (i.e., when organism was completely immobile), appearance, and behavior were recorded at 4±1-h and 24 ±1-h post exposure for each specified application interval.

Both facilities submitted samples of tank mix solutions, treated alfalfa, and spray cards to EN-CAS (Winston-Salem, NC) for dimethoate residue analyses. Tank mix solutions were analyzed by high performance liquid chromatograph coupled to an ultraviolet light absorbance detector (HPLC-UV), while alfalfa and spray card analyses were conducted using gas chromatography coupled to a flame photometric detector (GC-FPD).

2.1 Laboratory Data and Report Anonymization

Confidentiality is paramount in ring studies. Therefore, all laboratory submittals were anonymized by PER via the assignment of an alphabetic identifier (i.e., Lab A and Lab B); all identifying markers for each laboratory's data set and report were removed prior to review and analysis. No other participants or committee members were supplied with the laboratory anonymization codes. All subsequent communications regarding test or analytical data were confidential to prevent biased data review. PER's staff reviewed all raw data for adherence to the Ring Study Protocol; raw data included biological observations and environmental conditions.

2.2 Statistical Analysis Methods

Once all data were completely reviewed and in an anonymized format, the data were statistically analyzed using the Comprehensive Environmental Toxicity Information System (CETIS) software (TidePool Scientific Software, McKinleyville, CA). Linear interpolation was used to determine a point estimate of the residual time needed to reduce the activity of the test substance and bring honey bee mortality down to 25% (RT₂₅). As CETIS does not permit linear interpolation analysis of two data points (e.g., 6-h and 24-h), a t(0) data point assuming 100% mortality was added to generate the RT₂₅ upon approval of the PRTF study sponsor. A standard paired t-test was used to compare the mortality in the negative control(s) versus the treatment(s) at each time interval.

3. RESULTS

3.1 Environmental Conditions

Anonymized data submittals for laboratory A and B are provided in Appendix B and C, respectively.

Lab A

All field application conditions were acceptable. Bee exposure to foliage occurred in an environmental chamber under darkness and within the targeted temperature and humidity range using cages that met the protocol specifications.

Lab B

All field application conditions were acceptable. The bee exposure occurred in an environmental chamber under darkness and within the targeted temperature and humidity range and using cages that met the protocol specifications, except that the humidity for the June trial ranged from 36 – 53% rather than from 50-80%.

3.2 Chemical Analyses of Spray Tank Solutions, Spray Cards, and Treated Alfalfa

The final EN-CAS report for the dimethoate residue analyses performed on spray tank solutions, spray cards, and treated alfalfa submitted by Facility A and Facility B is provided in Appendix D.

3.2.1 Spray Tank Solutions

The results for the dimethoate analyses performed on spray tank solutions are presented in Table 2. No dimethoate was detected in the control tank samples submitted by either lab. The spray tank results for the two facilities were within 10% for the June samples and 24% for the September samples. The relative percent difference for the duplicate samples submitted by facility A was 2.9%.

Table 2. Dimethoate concentration (mg/L) in tank mix solutions.			
Sample Type	Application Timing	Facility A	Facility B
Control	June	NC	0
Control	September	0	0 ^a
Treated	June	2264	2488 (2562 ^b)
Treated	September	1958	2575

NC = not collected

a – freezer containing this sample thawed due to a power outage.

b – duplicate sample analysis.

3.2.2 Spray Cards

The results for the dimethoate analyses performed on spray cards are presented in Table 3. The mean dimethoate concentration on facility A spray cards was 109% greater than facility B spray cards for the June application, but the mean concentration was identical for both facilities for the September application.

Table 3. Dimethoate concentration (µg/card) on spray cards.			
Sample Type	Application Timing	Facility A	Facility B
Control	June	NR	0.0
Control	September	NR	0.0 ^a
Treated	June	422	275
Treated	June	863	257
Treated	June	440	293
Mean (S.D.)	June	575(±249)	275(±18)
Treated	September	347	347
Treated	September	510	510
Treated	September	537	537
Mean	September	465(±102)	465(±102)

NR – not reported

a – freezer containing this sample thawed due to a power outage.

3.2.3 Treated Alfalfa

No dimethoate was reported on the control treatment alfalfa samples submitted by either facility (Table 4). The dimethoate concentration decreased over time in the facility B June application alfalfa. No decrease over time was observed in the initial analysis of samples from the facility A June applications. Retain samples analyzed in an effort to confirm these results returned much lower concentrations and did demonstrate a decrease in concentration over time. The relative percent difference between the duplicate samples submitted by facility A for the June application ranged from 42-123%. Except for the 6-h after application facility A September application alfalfa, the dimethoate concentration decreased over the application intervals for alfalfa submitted by both facilities.

Table 4. Dimethoate concentration (µg/g) on treated alfalfa samples.				
Time Interval from Application	Plot	Application Timing	Facility A	Facility B
1-HAA	Control	June	0.0	0.0
6-HAA	Control	June	0.0	0.0
24-HAA	Control	June	0.0	0.0
48-HAA	Control	June	NC	NC
1-HAA	Control	September	0.0	NR ^a
6-HAA	Control	September	0.0	NR ^a
24-HAA	Control	September	0.0	NR ^a
48-HAA	Control	September	0.0	NC
1-HAA	Treated	June	22.1 (14.5 ^b)	10.6
6-HAA	Treated	June	32.8 (7.8 ^b)	8.0
24-HAA	Treated	June	21.6 (7.3 ^b)	2.3
48-HAA	Treated	June	NC	NC
1-HAA	Treated	September	15.2	19.5
6-HAA	Treated	September	6.4	19.4
24-HAA	Treated	September	18.7	12.3
48-HAA	Treated	September	5.2	NC

HAA – hours after application.

NC – not collected

NR – not reported

a – freezer containing these samples thawed due to a power outage. Samples exhibited signs of mold growth.

b – duplicate sample analysis.

3.3 Toxicity of Dimethoate to Adult Honey Bees

3.3.1 Facility A June Alfalfa Application Tested By Lab A

The results of this test are summarized below in Table 5. The RT_{25} was 10.8 h. The summary of statistics is provided in Appendix E.

Table 5. Facility A June Alfalfa Application Tested By Lab A.								
Timepoint	Treatment	% Mortality						
		Rep A	Rep B	Rep C	Rep D	Rep E	Rep F	Mean
6-h	Control	0	0	0	0	0	0	0
	Treatment	100	100	100	100	100	92	98.7*
24-h	Control	0	0	4	0	0	0	0.7
	Treatment	0	4	4	56	4	0	10.7 ^a
Summary of Statistics								
RT_{25} =					10.8 h			

* Statistically significant increase in mortality relative to the Control at $p < 0.05$.

a – Abbots Correction performed due to Control mortality.

3.3.2 Facility A June Alfalfa Application Tested By Lab B

The results of this test are summarized below in Table 6. The RT_{25} was 10.8 h. The summary of statistics is provided in Appendix F.

Table 6. Facility A June Alfalfa Application Tested By Lab B.								
Timepoint	Treatment	% Mortality						
		Rep A	Rep B	Rep C	Rep D	Rep E	Rep F	Mean
6-h	Control	0	0	0	0	0	0	0
	Treatment	100	100	100	100	100	100	100*
24-h	Control	16	16	4	4	0	12	8.7
	Treatment	0	4	0	16	0	12	5.3 ^a
Summary of Statistics								
RT_{25} =					10.8 h			

* Statistically significant increase in mortality relative to the Control at $p < 0.05$.

a – Mortality presented is not corrected for control mortality since Abbots Correction produced a negative number.

3.3.3 Facility B June Alfalfa Application Tested By Lab A

The results of this test are summarized below in Table 7. The RT_{25} was 10.2 h. The summary of statistics is provided in Appendix G.

Table 7. Facility B June Alfalfa Application Tested By Lab A.								
Timepoint	Treatment	% Mortality						
		Rep A	Rep B	Rep C	Rep D	Rep E	Rep F	Mean
6-h	Control	0	0	0	0	0	4	0
	Treatment	96	100	100	100	96	96	98.0^{*a}
24-h	Control	0	0	0	0	0	4	0.7
	Treatment	0	0	0	0	0	0	0
Summary of Statistics								
$RT_{25} =$					10.2 h			

* Statistically significant increase in mortality relative to the Control at $p < 0.05$.

a – Abbotts Correction performed due to Control mortality.

3.3.4 Facility B June Alfalfa Application Tested By Lab B

The results of this test are summarized below in Table 8. The RT_{25} was 10.8 h. The summary of statistics is provided in Appendix H.

Table 8. Facility B June Alfalfa Application Tested By Lab B.								
Timepoint	Treatment	% Mortality						
		Rep A	Rep B	Rep C	Rep D	Rep E	Rep F	Mean
6-h	Control	0	0	4	0	4	4	2.0
	Treatment	100	100	100	100	100	100	100^{*a}
24-h	Control	12	0	4	4	36	12	11.3
	Treatment	0	4	20	16	28	16	3.0 ^a
Summary of Statistics								
$RT_{25} =$					10.8 h			

* Statistically significant increase in mortality relative to the Control at $p < 0.05$.

a – Abbotts Correction performed due to Control mortality.

3.3.5 Facility A September Alfalfa Application Tested By Lab A

The results of this test are summarized below in Table 9. There was far less mortality in the 6-h post application sample relative to the mortality observed for both Facility A and Facility B June applications (Tables 4-7). These results likely skewed the linear interpolation resulting in a lower RT_{25} . The RT_{25} was 3.5 h. The facility A September dimethoate concentration results for the alfalfa are not proportionally different than facility B (Table 3), so the chemistry results do not explain the change in the RT_{25} for this date set. The summary of statistics is provided in Appendix I.

Table 9. Facility A September Alfalfa Application Tested By Lab A.								
Timepoint	Treatment	% Mortality						
		Rep A	Rep B	Rep C	Rep D	Rep E	Rep F	Mean
6-h	Control	0	4	8	0	4	4	3.3
	Treatment	8	40	8	0	8	0	7.6 ^a
24-h	Control	4	8	12	20	4	4	8.7
	Treatment	64	92	36	72	84	48	63.0^{*a}
48-h	Control	0	0	8	0	4	0	2.0
	Treatment	0	0	4	0	0	0	0.7 ^b
Summary of Statistics								
$RT_{25} =$					3.5 h			

* Statistically significant increase in mortality relative to the Control at $p < 0.05$.

a – Abbotts Correction performed due to Control mortality.

b – Mortality presented is not corrected for control mortality since Abbotts Correction produced a negative number.

3.3.6 Facility A September Alfalfa Application Tested By Lab B

The results of this test are summarized below in Table 10. Similar to the results reported by Lab A testing of this alfalfa (Table 8), there was far less mortality in the 6-h post application sample relative to the mortality observed for both Facility A and Facility B June applications (Tables 4-7). These results likely skewed the linear interpolation resulting in a lower RT_{25} . The RT_{25} was 2.3 h. The summary of statistics is provided in Appendix J.

Table 10. Facility A September Alfalfa Application Tested By Lab B.								
Timepoint	Treatment	% Mortality						
		Rep A	Rep B	Rep C	Rep D	Rep E	Rep F	Mean
6-h	Control	0	0	8	4	0	4	2.7
	Treatment	56	28	32	48	24	36	35.6^{*a}
24-h	Control	12	4	0	4	8	4	5.3
	Treatment	28	8	20	12	24	32	16.2^{*a}
Summary of Statistics								
$RT_{25} =$					2.3 h			

* Statistically significant increase in mortality relative to the Control at $p < 0.05$.

a – Abbotts Correction performed due to Control mortality

3.3.7 Facility B September Alfalfa Application Tested By Lab A

The results of this test are summarized below in Table 11. The RT_{25} was 10.7 h. The summary of statistics is provided in Appendix K.

Table 11. Facility B September Alfalfa Application Tested By Lab A.								
Timepoint	Treatment	% Mortality						
		Rep A	Rep B	Rep C	Rep D	Rep E	Rep F	Mean
6-h	Control	8	20	36	28	20	20	22 ^b
	Treatment	100	100	100	100	100	100	100*
24-h	Control	8	6	8	4	0	16	8.7
	Treatment	0	4	8	4	4	8	4.7 ^a
Summary of Statistics								
$RT_{25} =$					10.7 h			

* Statistically significant increase in mortality relative to the Control at $p < 0.05$.

a – Mortality presented is not corrected for control mortality since Abbotts Correction produced a negative number.

b – Control performance for the 6-h post application sample did not meet the EPA test validity criteria of $< 20\%$ mortality. Regardless, the data for this test are reported here to allow for comparison of RT_{25} results between labs since this study is not associated with a product registration.

3.3.8 Facility B September Alfalfa Application Tested By Lab B

The results of this test are summarized below in Table 12. The RT_{25} was 11.3 h. The summary of statistics is provided in Appendix L.

Table 12. Facility B September Alfalfa Application Tested By Lab B.								
Timepoint	Treatment	% Mortality						
		Rep A	Rep B	Rep C	Rep D	Rep E	Rep F	Mean
6-h	Control	0	0	0	0	0	0	0
	Treatment	100	100	100	100	100	96	99.3*
24-h	Control	4	0	0	4	8	8	4.0
	Treatment	20	16	32	12	12	24	16.0*^b
Summary of Statistics								
$RT_{25} =$					11.3 h			

* Statistically significant increase in mortality relative to the Control at $p < 0.05$.

a – Abbotts Correction performed due to Control mortality

4. SUMMARY AND CONCLUSIONS

The PRTF designed a Phase II ring study to control for two sources of variability in the honey bee toxicity testing of residues on foliage (RT₂₅): differences in application equipment, potentially leading to inconsistent distribution of the test substance over the treated plots, and/different environmental conditions resulting in different dissipation/degradation rates in the treated plots. The study involved Eurofins and Smithers performing RT₂₅ honey bee testing of alfalfa treated with dimethoate in June and September of 2021. The two facilities coordinated the timing of crop planting and dimethoate applications to occur within a two-week window to allow each laboratory to test both their own alfalfa as well as the alfalfa from the other lab. In addition to the honey bee tests, analytical chemistry samples were collected from tank mix solutions, and spray cards, and treated alfalfa.

The spray tank results for the two facilities were within 10% for the June samples and 24% for the September samples. The mean dimethoate concentration on facility A spray cards was 109% greater than facility B spray cards for the June application, but the mean concentration was identical for both facilities for the September application. The dimethoate concentration on alfalfa generally decreased over application intervals for four of five sets of alfalfa tested (including the duplicate analysis).

The RT₂₅ obtained by both labs was similar for both facility's June applications alfalfa and for the facility B alfalfa September application (Table 12). Although the RT₂₅ was similar for both labs testing of facility A September application alfalfa, they were ~3-5x lower than those obtained for the other pair-wise lab comparisons. The tank mix solution, spray card, and alfalfa dimethoate analyses do not explain the reduced RT₂₅ values obtained by both labs for the facility A September application alfalfa.

Table 12. Dimethoate RT₂₅ for Two Alfalfa Applications Tested by Two Labs				
Lab Identifier	June Application		September Application	
	Facility A Alfalfa	Facility B Alfalfa	Facility A Alfalfa	Facility B Alfalfa
Lab A	10.8 h	10.2 h	3.5 h	10.7 h
Lab B	10.8 h	10.8 h	2.3 h	11.3 h

This study demonstrates that consistent test results can be obtained for two labs testing the same alfalfa after controlling for application equipment and environmental conditions during dimethoate application.

Appendix A

Pollinator Research Task Force Ring Test Protocol



Protocol for Phase II: Standardization of Honey Bee Toxicity of Residues on Foliage (RT₂₅) Study Design

Based on EPA's Ecological Effects Test Guideline OCSP 850.3030, dated January 2012, with modifications

1. **Purpose:** This guideline is intended for use in developing data on the residual toxicity to honey bees of chemical substances and mixtures ("test chemicals" or "test substances") subject to environmental effects testing requirements. This guideline describes a toxicity test in which honey bees are exposed to weathered residues of a test substance on treated foliage.
2. **Definitions:**
 - a) Acute Residual Toxicity is the adverse effects occurring over a period of time (hours or days) from a single dose of the test substance to foliage.
 - b) Dose is the amount of test substance applied. Dose is expressed as a mass, pounds of test substance per acre (lbs/A) and for a pesticide, pound(s) of active ingredient applied per acre (lbs a.i./A). The dose used in this test should be the maximum, single application dose allowable according to the end-use product labeling.
 - c) Mortality: an animal is recorded as dead when it is completely immobile (e.g., no movement within 5 seconds).
 - d) RT₂₅ is the residual time needed to reduce the activity of the test substance and bring bee mortality down to 25% in cage test exposures to field-weathered spray deposits (see paragraph (e)(2) of this guideline). The time period represented by this toxicity value (RT) is considered to be the length of time (in hours) that the test substance is expected to remain toxic by contact to bees in the field, when bees are exposed to weathered residues of the test substance on vegetation at an expressed rate of application (lb a.i./A). Exposure to weathered residues in the laboratory are a surrogate for field conditions.
3. **Summary of test:** The honey bee (*Apis mellifera*) foliar residue study is a laboratory test designed to determine the length of time over which field-weathered foliar residues remain toxic by contact to honey bees. The test substance (e.g., a typical end-use product) is applied to crop foliage, the foliage is harvested at predetermined intervals post-application, and test bees are caged on the treated foliage. Results are expressed in terms of the length of time (observed time interval) following application, during which residues continue to cause 25% mortality (RT₂₅) in test populations at an expressed rate of application (lb a.i./A).
4. **General test guidance:** Based on EPA's Ecological Effects Test Guideline OCSP 850.3030, dated January 2012, with some modifications.
5. **Definitive test:** The goal of the definitive test is to determine the 24-h RT₂₅, length of time post-application that residues of the test substance on foliage are toxic to honey bees. For this determination, one treatment level, the maximum rate on the label and at least three different time intervals between application and harvest are typically used. The test substance will be evaluated at the labeled maximum, single application rate. A summary of test conditions is provided in Table 1, and validity elements for an acceptable definitive test are listed in section 11 of this protocol.

6. Test specifications:

6.1. Test organism:

- a) **Species:** Honey bee, *Apis mellifera*, is the test species.
- b) **Source:** Bees may be obtained from on-site colonies or from a commercial apiary. All control and treatment bees used in a test should be from the same source and breeding lineage. Bees are emerged from brood frames taken from the source colonies in an incubator (34-35 °C, 45-90% humidity) and reared for three to five days with "bee bread" (pollen that is already stored on the brood frame) supplemented with pollen patty and 50% w/v sucrose in water solution. In order to obtain a sufficient number of bees with known age (3-5 days post-emergence), brood frames can be collected from multiple colonies within the same apiary. Collection in early spring or late autumn should be avoided, as the bees have a changed physiology during this time.
- c) **Age:** The test should be conducted using young adult worker bees that are of a similar age (three to five days post-emergence) and feeding status.
- d) **Health status:** Bees used in the test should be in apparent good health. Only bees from apparently disease-free colonies should be used, and they should be kept in conditions conforming to proper culture practices. Bees treated with chemical substances, such as antibiotics, anti-varroa, etc., should not be used for toxicity tests for four weeks from the time of the end of the last treatment.
- e) **Care and handling:** During holding and testing, bees should be shielded from excessive activity, handling stress or other disturbances and kept in the dark. Bees should be handled only as much as is necessary to conform to test procedures.
- f) **Diet and feeding:** A 50% weight/volume (w/v) or weight/weight (w/w) solution of sugar/water (500 grams/liter) is provided *ad libitum* throughout the holding and test periods. Purified or distilled water should be used for preparation of the sugar solution. Top feeding is preferred, so for the ring test, the feeding syringe/tube should be inserted through an opening in the top of the test cage. Attention should be paid to avoid any contact between the feeders and the treated foliage.

6.2. **Test crop:** The test crop will be alfalfa (*Medicago sativa*). Alfalfa should be grown in an unshielded open outdoor field location. Foliar applications of the test substance should be performed when the alfalfa crop is between **20-40 centimeters** in height. To ensure harvest is not impeded by excessive weed growth, pre-emergence and early post-emergence herbicide applications may be made to the cropped area. Applications of any maintenance pesticides (herbicides, fungicides, insecticides) must not be made within 4 weeks of the start of the study. Fertilizer and irrigation treatments may be made as needed consistent with good agronomic practices up to 24 hours before start of the study but must not be made during the study. All agronomic practices, variety of alfalfa, the seeding rate, date of planting, fertilizer, irrigation and pesticide treatment history for the three years prior to the start of the study, should be reported. If seeds treated with seed-applied pesticides are used to establish the crop, the field should not be used for RT₂₅ studies for 1 year from planting.

6.3. **Test duration:** The test starts with the placement of weathered treated foliage into cages with bees, followed by a 24-h observation period during which mortality and clinical signs of toxicity are recorded at 4±1 and 24±1 h post-exposure.

- a) **Post-treatment weathering intervals:** The treated foliage should be harvested at minimum two mandatory intervals 6 ± 1 and 24 ± 1 h post-application, and placed in cages to expose young adult honey bees to the weathered residues of test substance. Based on the results from Phase I of this project, the 3-hour harvest interval will be excluded since 100% mortality was observed in both facilities with the same test substance at the 6-hour harvest interval. The two labs will coordinate test initiation so that the bioassay phases begin within 1 hour of each other. If mortality of bees exposed to the foliage harvested 24 h after the application is greater than 25% (control-corrected), weathered, treated foliage samples should continue to be collected and tested at 24-h intervals until the mortality is $\leq 25\%$ (control-corrected), up to five days post-application. For the ring test, the treated foliage will be harvested at 6 ± 1 and 24 ± 1 h post-application intervals, with option of 48 h, 72 h, 96 h and 120 h intervals if mortality stays $>25\%$.
- 6.4. **Observation period:** Bees will be observed for **24 h** after the bees and treated foliage are placed onto the cages.
- 6.5. **Test facilities:** Test substance application and weathering should occur outdoors under natural field conditions. The bee exposure portion of the test should be conducted indoors to control lighting and other environmental variables, while bees are being maintained in small test cages. The cages containing honeybees should be placed in an environmental chamber to control temperature and relative humidity.
- 6.6. **Sample Sharing:** At each harvest interval, the PRTF will arrange for samples of treated alfalfa to be transported from one of the facilities to another. Due to the close proximity of the facilities in the State of North Carolina, the transportation of samples should take less than 1 hour. Both facilities will conduct bioassays on subsamples from the same harvested foliage. The laboratories will coordinate the start time of the bioassays so that they begin within 1 hour of each other.
- 6.7. **Test cages:** Use of test cages with different dimensions could potentially lead to inconsistent exposure. So, for the ring test, each CRO will use a standard cage to remove this as a source of variability. Determining an optimum cage design was part of Phase I of the ring test. The test cages should have a suitable opening for the introduction of treated foliage and bees, and another opening at the top for inserting the feeding syringe/tube. Cages should be cleaned thoroughly between uses or new cages are used for every trial. For this ring test, transparent 32 oz plastic containers (upper diameter = approx. 11 cm, base diameter = approx. 9 cm; height = approx. 14 cm) will be used as test cages (see Fig .1). The top of the test cage will be covered with a screened lid to allow ventilation and has an opening for inserting a feeding syringe.
- 6.8. **Collection of bees:** The day prior to exposure, young bees should be collected from frames kept in the incubator and acclimated for approximately 24 hours. The bees can be acclimated in bulk or acclimated in the actual test cages. If the acclimation occurs in the test units/cages dead and impaired bees should be removed and, if needed, replaced by healthy bees from the same pool of newly emerged bees prior to the introduction of the test foliage. If acclimation occurs in the test cages, it is recommended that excess bees be acclimated in excess test cages in case there is a need to replace dead or impaired bees prior to test initiation. Introduction of bees into the test cages shall be done in an indiscriminate manner. During transfer to the exposure cages, immobilization of bees with cold temperatures, carbon dioxide gas (CO_2) or nitrogen gas (N_2), may be necessary but should be kept to the minimum.

- 6.9. **Controls:** Paired negative (untreated) controls are included in the test. Control crop foliage is treated with water only and identically to treatment plots, except for applications of the test substance. Control and test bees are kept under the same environmental conditions.
- 6.10. **Number of test organisms and replicates:** Six replicates should be assigned to each treatment and control group at each post-application interval, with a minimum of 25 bees for each replicate. Test organisms should be impartially assigned to different treatment groups.
- 6.11. **Test substance:** The substance to be tested will be Dimethoate 400 EC
- 6.12. **Application of test substance:** The test substance will be applied at the maximum single application rate of 0.5 lbs. a.i./acre (spray coverage = 200 L mix/ha). A single application should be made in the morning after the dew has dried and when alfalfa crop is between 20-40 centimeters in height. Application should be made in the field with a tractor mounted or hand-held boom sprayer, using standard nozzles in accordance with regionally accepted practices. The sprayer should be calibrated on the day of, or a day prior, to the spraying of the plants. Spray tank solutions should be continuously stirred or circulated prior to and during use. Nozzle height above the crop during application should be maintained consistent with manufacturer recommendations and will be coordinated between the two facilities for consistency. Wind speed should be less than 3 m/sec during application. Spray equipment should produce a wide enough swath so that the alfalfa plots can be treated in single-pass spray. Detailed aspects of the application shall be reported including nozzle type, spacing, height above crop canopy, flow rate, pressure, application speed and pass times, nominal and actual volumes applied, results of equipment calibration, volumes and concentrations of spray solutions prepared. Environmental conditions during application shall be recorded including air temperature, relative humidity, soil moisture, presence/absence of dew or moisture on the crop, cloud cover, wind speed, application time of day (beginning and end of spraying), time of sunrise and sunset and any other relevant observations that may affect the interpretation of the results.
- 6.13. **Application timing:** Phase II of this project will likely consist of two separate coordinated applications at each test facility at different times during the year in order to evaluate the impact of environmental conditions in the field on the test results. The first application at each facility will be targeted for late May/early June, during a period of time when it is generally hot and dry in North Carolina. The two facilities will coordinate the planting and treatment of alfalfa so that the applications will occur within two weeks of one another, under similar environmental conditions. Applications on the exact same day will be avoided so that sample shipment and bioassay conduct will more easily be coordinated within each individual facility. A second application at each facility will be targeted for late July/early August, during a period of time when it is generally very humid in North Carolina. Once again, the two facilities will coordinate the planting and treatment of alfalfa so that the applications will occur within two weeks of one another, under similar environmental conditions.
- 6.14. **Field plots and harvest of foliage:** Plots should be at least 1 m² (10.8 square feet) in alfalfa grown according to standard agricultural practices. Applications of any maintenance pesticides (herbicides, fungicides, insecticides) must not be made within 4 weeks of the start of the study. At a minimum, nine test substance treatment plots are used to obtain three plots for harvesting at each time interval (6±1 and 24±1 h post-application). After test substance residues have aged (weathered) for the appropriate

time period, alfalfa foliage sufficient to place in six treatment cages at each facility (approximately 180 g fresh weight or 6,000 cm³ total), should be harvested from three treated test plots using hand equipment, placed individually in labeled bags and returned immediately to the laboratory for processing and placement in test cages or transport to the other test facility. Foliage should be collected, using a random sampling scheme, from the top 15 cm of the canopy. Minimum distance of 10 m should be kept between treatment and control plots to avoid potential contamination of control plots due to drift. At each of the minimum time intervals, three alfalfa samples are harvested from the control plot using a random sampling scheme, to obtain sufficient foliage to place in six control cages at each test facility. If additional harvest intervals are required beyond the minimum two, control samples must be collected and tested also at each harvest interval.

- 6.15. **Preparation of treated foliage:** Samples of foliage are returned to the laboratory in bags and transported in coolers that should be held between 8 and 12 °C once the coolers are filled and closed. Temperature data loggers should be included in the coolers. The samples are mixed thoroughly and then divided into approximately 15 g or 500 cm³ portions. The current guideline recommends chopping the foliage into smaller (2.5 cm) lengths and loosely placing 15 g portions at the bottom of each test cage, but after the discussions with the project team it was concluded that this step is not necessary and should be avoided. For the ring test, leave the foliage in 12-15 cm lengths and loosely place 15 g portions upright/diagonally in each test cage to maximize the exposure.
- 6.16. **Introduction of the bees to the treated foliage in the cages:** Bees should then be released on the top of the foliage or the treated foliage added directly into the test cages if the bees are being acclimated in the test cages. Special attention should be paid to avoid any direct contact between the sugar solution feeders and the treated foliage.
- 6.17. **Sampling for residue analysis:** An approximately 15 g sample of the treated and untreated control foliage immediately after the spray has dried (approximately 1 hour \pm 30 minutes) and at each harvest interval will be collected to confirm test substance concentration. If the study extends past 24 hours, then samples of foliage will continue to be taken at each 24-h interval thereafter, to correspond with the exposure, up to 5 days post-application. Fresh sample weights should be recorded before freezing the samples. In addition, for the ring test, analytical evaluations will also be conducted on spray solution (*i.e.*, tank mix) and three spray cards (preferably glass fiber discs) placed randomly in the test plots for the application. The spray solution sample should be collected after completion of the application. The spray cards should be held in a horizontal position at the top height of the crop canopy so that it gets the full rate of the spray without interception by the crop. At the time of collection, the spray cards should be folded and placed into plastic bags similar to those used for foliage collection. Two tank mix samples, 50 ml each, will be collected upon completion of the application and labeled A and B. Tank mix sample A will be analyzed for rate verification, and sample B will be retained for further analysis if needed. Samples are to be transported from the field and subsequently deep frozen until shipment to the designated analytical laboratory. Samples should be shipped to the designated analytical laboratory deep frozen.
- 6.18. **Environmental conditions:**

a) **Environmental conditions during application and weathering in the field:**

Sunlight, precipitation and temperature are three extremely important factors in the dissipation of pesticide residues. Test substance application should be made preferably on clear days with maximum temperatures ranging between 20-40 °C and <30% chance of precipitation. Application should happen in the morning after dew or moisture from any overnight rains has dried off. Test plots should be protected from direct precipitation for at least 3 h (up to 6 h) following the application. If rainfall should occur, the test plots should be sheltered from direct rainfall using a tarp or other suitable canopy. If a canopy is used, it should be removed 3 h (up to 6 h) after application to allow full effect of natural weathering to take place (*i.e.*, direct sunlight). Also, application should be avoided in windy conditions (*i.e.*, average wind speed >3 m/s) to avoid contamination of untreated control plots. Treated test crop should be allowed to weather outdoors under natural field conditions.

b) **Environmental conditions during exposure phase:** Environmental parameters in the laboratory during the bioassays should be maintained as follows:

- I. Temperature and humidity. Temperature should be maintained between 25 and 35°C, with relative humidity between 50% and 80%.
- II. Lighting and photoperiod. It is recommended that test bees be maintained in the dark except during transfer to test cages and observations.
- III. Test cages, including treated and control cages, are placed within the incubator in a randomized pattern which is also recorded.

7. **Observations:**

7.1. **Analysis for test substance concentrations:** Test substance residues on treated foliage are expressed in parts per million (ppm; mg ai/kg foliage) fresh weight. For the ring test, analytical evaluations will also be conducted on spray solution (*i.e.*, tank mix; mg a.i./L) and three spray cards placed randomly in test plots during the application (analyzed as mg a.i./cm² and also reported in units of lb a.i./acre). The residue analyses for the trials will be conducted at one designated lab to avoid inter-lab variability.

7.2. **Field site conditions:** Environmental conditions should be monitored at the field site at the time of test substance application and during weathering period. Environmental information to be collected should include daily minimum and maximum air temperature, precipitation, and relative humidity. Wind speed and estimated cloud cover should be recorded at least at the time of application. A data-logging weather station shall be placed on site, within 1 km of the application area, to collect environmental data.

7.3. **Conditions during exposure in the lab:** Temperature and relative humidity should be recorded during the bee exposure in laboratory test cages.

8. **Measures of Effects:**

8.1. **Mortality:** For a given weathered residue treatment or control, bees should be observed for mortality at least once at 4±1 h after exposure and at exposure termination at 24 h. Dead bees should not be removed from the test cages until the test is terminated.

8.2. **Appearance and behavior:** For a given weathered residue treatment or control, bees should be observed for all clinical signs of intoxication and any other abnormal behavior once during the first 4±1 h after exposure and at test termination (24 h). Observations should be recorded by treatment level and by time of occurrence. Signs of intoxication

are those behaviors apparently due to the test substance and may include a wide variety of behaviors, such as ataxia, lethargy, excessive cleaning, tremors, convulsions and hypersensitivity (agitation). Prior to the evaluation at test termination, observations should be made without disturbing or removing bees from the test chambers; for these observations, estimates of mortality and effects are sufficient.

9. Treatment of results:

9.1. Descriptive summary statistics:

- a) Environmental conditions: Data should be summarized in tabular form, showing the range and mean temperature, precipitation, relative humidity, and wind speed.
- b) Mortality. Data should be summarized in tabular form, showing for each weathered age of foliage treatment and control the number of bees initially exposed, mortality at each observation time, and the percent mortality. Average mortality in the controls, if any, will be used to correct the mortality observed in the treatments using Abbott's formula.
- c) Appearance and behavior. Data should be summarized in tabular form, showing for each weathered age of foliage, appearance and behavior at each observation time. Statistical analysis of sublethal effects are not conducted.

9.2. Residual Time (RT_{25}): A test for comparing two paired populations (*e.g.*, paired t-test) should be performed to detect significant ($p < 0.05$) difference of treatments from controls. Abbott's correction should be used in the event of control mortality. Additional discussion about measurement endpoints and statistical procedures is found in OCSP 850.3000.

10. Tabular summary of test conditions: Table 1 lists the important conditions that should prevail during the definitive test. Meeting these conditions will increase the likelihood that the completed test will be acceptable or valid.

Table 1. Summary of Test Conditions for Honey Bee Toxicity of Residues on Foliage Test

Test type	Toxicity of residues on foliage
Test duration	24 h observation period for each aged residue interval (6±1 and 24±1 h aged residue intervals are tested; additional 24 h residue intervals may be appropriate).
Temperature during laboratory exposure	25 - 35°C
Relative humidity during laboratory exposure	50 – 80%
Lighting	Darkness, except during transfer of bees to treatment cages and observations
Test chamber	32 oz plastic cages with an upper diameter approximately 11 cm, base diameter of approximately 9 cm and height of approximately 14 cm will be used in the ring test
Foliage cutting length and placement	Foliage lengths of 12-15 cm; upright/diagonally placed in test cages
Test substance application	15-g or 500-cc portions of treated foliage placed in a test cage
Age of test bees	Young adult worker bees of similar age (1-5 days post-emergence) and feeding status
Number of bees per chamber	25 (minimum)
Number of bees per treatment and control	150 (minimum)
Number of treatments	Minimum of 2 treatment groups (6±1 and 24±1 h post-application of maximum single application rate) which includes the negative control(s). Additional intervals may be appropriate if mortality is >25% for the 24 h post- application treatment
Feeding	50% sugar/water (w/v) solution <i>ad libitum</i>
Measure of Effect or Measurement Endpoint	RT ₂₅ based upon mortality at 24 h after bees are exposed to foliage. If mortality of bees exposed to the foliage harvested 24 h after the application is greater than 25%, additional weathered, treated foliage samples will continue to be taken every 24 h.

11. **Test validity criteria:** The definitive test will be considered invalid if one or more of the following conditions occurred -

- a) Test bees were not of similar age and feeding status.
- b) More than 20% mortality averaged across control treatments.
- c) All bees in a test were not from the same source (apiary) and breeding lineage.
- d) Concurrent negative (untreated) controls were not included in the test.
- e) Environmental conditions (temperature, precipitation, relative humidity, wind speed and cloud cover) at the field site were not monitored/reported.
- f) Test organisms were not impartially assigned to test cages.
- g) Substances, other than the test pesticide were applied to the growing alfalfa within 4 weeks of test initiation.

12. **Reporting:**

12.1. **Protocol deviations:** Include a description of any deviations from the test protocol or any occurrences which may have influenced the results of the test.

12.2. **Test substance:**

- a) End-use product (name, state or form, source), its purity (for pesticides, the identity (common name, IUPAC and CAS names, CAS number) and concentration of active ingredient(s)) and known physical and chemical properties that are pertinent to the test.
- b) Storage conditions of the test substance.
- c) Methods of preparation of test substance for application onto foliage, the maximum label rate, and the actual application rate (lb a.i./A) with the finished spray volume per acre.
- d) Describe the stability of the test substance under storage conditions.

12.3. **Test organisms:**

- a) Scientific name, race, and source.
- b) Culture method and conditions.
- c) Health status of colonies used for collection of test bees (e.g., any adult diseases, use and application date(s) of any prophylactic or preventative treatments).
- d) Collection method and date of collection.
- e) Holding period.
- f) Age at initiation of exposure to an aged residue treatment.

12.4. **Test system and conditions:**

- a) Description of housing conditions: type, size, and material of test cages.
- b) Description of any feeding during the test (if applicable), including: method, type of food, source, amount given and frequency.
- c) Common and scientific name of treated crop.
- d) Plot size, and method and time of administration of test pesticide on plots.

- e) Number of aging intervals tested.
- f) Time after application to plot of foliage collection (age intervals tested) and placement of foliage in test chambers.
- g) Plots per aging interval and negative control.
- h) Number of bees per test cage.
- i) Number of cages (replicates) per aging interval plot and negative control plot.
- j) Methods used for test cage and treatment randomization as well as methods for impartial assignment of bees to test cages.
- k) Exposure duration to a given aged residue and duration of the study.
- l) Methods and frequency of environmental monitoring performed on treated plots during administration of test substance and weathering period for temperature and precipitation, and any other known weather conditions that would impact initial concentration or stability of residue levels on treated plots.
- m) Methods and frequency of environmental monitoring performed during the definitive study or positive control study for test room temperature, humidity and lighting.
- n) For the definitive test, all analytical procedures and preservation methods should be described. The accuracy of the method, method detection limit, and limit of quantification should be given.

12.5. **Results:**

- a) Laboratory environmental monitoring data results (test room temperature, humidity and lighting) in tabular form (provide raw data for measurements not made on a continuous basis), and descriptive statistics (mean, standard deviation, minimum, maximum).
- b) Field site environmental monitoring data results (temperature, precipitation, wind speed, relative humidity, cloud cover) in tabular form (provide raw data for measurements not made on a continuous basis), and descriptive statistics (mean, standard deviation, minimum, maximum).
- c) For the bioassays, the number of dead bees which were observed at least once during the first 4 hours of exposure and at 24 h (provide the raw data).
- d) For the bioassays, a description of signs of intoxication and other abnormal behavior, including time of onset, duration, severity, and number affected at each aged residue treatment and control(s) (provide the raw data).
- e) Provide 24-h RT_{25} values.
- f) Description of method used, including software package, for determining the 24-h RT_{25} value.
- g) Results of analysis of variance (ANOVA) to detect significant differences of treatment groups from the controls.

13. **References:** The references in this paragraph should be consulted for additional background material on this test guideline.

- a) Abbott, W.S., 1925. A method of computing the effectiveness of an insecticide. *Journal of Economic Entomology* 18:265-267.

- b) Johansen, C. *et al.*, 1977. Bee Research Investigations. Dept. of Entomology, Washington State University, unpublished, 22 pp.
- c) Lagier, R.F. *et al.*, 1974. Adjuvants Decrease Insecticide Hazard to Honey Bees. College of Agriculture Research Center, Washington State University Bulletin 801, 7 pp.
- d) Mayer, D. and C. Johansen, 1990. Pollinator Protection: A Bee & Pesticide Handbook. Wicwas Press. Cheshire, CT.
- e) Mayer, D. (approved by), 1996. Standard Operating Procedure (SOPs) – Residue Bioassay. The Bee Group-Irrigated Agriculture Research and Extension Center. Prosser, WA.
- f) U.S. Environmental Protection Agency, 1982. Pesticide Assessment Guidelines Subdivision L Hazard Evaluation: Nontarget Insects. Office of Pesticides and Toxic Substances, Washington, D.C., EPA-540/9-82-019.
- g) U.S. Environmental Protection Agency, 1985. Hazard Evaluation Division Standard Evaluation Procedure, Honey Bee—Toxicity of Residues on Foliage. Office of Pesticides Programs, Washington, D.C., EPA-540/9-85-003.
- h) USEPA 2012. Ecological Effects Test Guidelines OCSPP 850.3030: Honey Bee Toxicity of Residues on Foliage. Office of Chemical Safety and Pollution Prevention (7101). EPA 712-C-018. January 2012.
- i) USEPA. 2012. Ecological Effects Test Guidelines OCSPP 850.3020: Honey Bee Acute Contact Toxicity Test. Office of Chemical Safety and Pollution Prevention (7101). EPA-712-C-019. January 2012.
- j) EPA. 2017. U.S. Environmental Protection Agency's policy to mitigate the acute risk to bees from pesticide products. Office of Pesticide Programs. January 12, 2017. EPA-HQ-OPP-2014-0818-0477.

Next steps:

- Finalization of phase II ring test protocol: May 2021
- Experimental phase(s) of ring test: May-June, 2021, and July-August, 2021
- Transfer of residue samples to designated analytics lab: End June 2021 and end of August 2021
- Data submission by participating CROs to designated 3rd party: September, 2021
- Data evaluation and analytical analysis of residue samples: October, 2021
- Draft report: December, 2021
- Decision on phase III improvements: 1Q 2022
- Phase III improvements: 2022 onwards

Fig 1. Test cage



Transparent 32 oz plastic containers (upper diameter = approx. 11 cm, base diameter = approx. 9 cm; height = approx. 14 cm) will be used as test cages. The top of the test cage will be covered with a screened lid to allow ventilation and has an opening for inserting feeding syringe.

Appendix B

Anonymized Data Submittal – Lab A





**PRTF RT25 Ring Test 2021
DATA PACKAGE**

Pages: 125

PROTOCOL

0001

Standardization of Honey Bee Toxicity of Residues on Foliage (RT₂₅) Study Design – Phase II

Pollinator Research Task Force: *Joseph Wisk, Daniel Schmehl, Bibek Sharma, Timothy Joseph, Max Feken and Verissimo Sa*

Preface:

Residual toxicity data for bees are generated through the Toxicity of Residues on Foliage Test (OCSPP Guideline 850.3030)¹ and are referred to as RT₂₅ data. The RT₂₅ is the time needed for pesticide residues to decline on the foliage of a treated crop, such that when adult honey bees (*Apis mellifera*) are exposed to the treated foliage for 24 h, mortality is below or equal to 25%. The RT₂₅ is intended to be a measure of the time that the pesticide formulated product is expected to remain toxic to bees in the field when sprayed at the maximum application rate. Based on the EPA's regulations for requiring ecological effects data related to impacts on non-target organisms (40 CFR 158.630), the EPA has typically determined whether RT₂₅ data are needed based on the results of the adult bee acute contact toxicity test (OCSPP Guideline 850.3020)²; the toxicity of residues on foliage study is triggered if one or more active ingredients within the formulation has a median lethal dose to 50% of the bees tested (LD₅₀) of less than 11 µg/bee and the use pattern(s) indicate(s) that honey bees may be exposed to the pesticide. This study is also conditionally required in South Korea and will be required in the future in Brazil (IBAMA Bee Normative).

Traditionally, the residual toxicity (RT₂₅) information has been considered useful to growers and beekeepers to ensure bee safety, as it can help them determine the appropriate amount of time between pesticide application and increased bee activity. As per US EPA's recently released policy to mitigate the acute risk to bees from pesticide products (2017)³, "if acceptable product-specific toxicity of residues on foliage data (OCSPP 850.3030) are submitted and indicate an RT₂₅ value of ≤6 h, then the EPA will generally allow the acute risk mitigation language to be amended to indicate that the subject product may be applied during bloom if it is applied between 2 h prior to sunset but not less than 8 h prior to sunrise at the application site."

While compiling and reviewing the available RT₂₅ data, EPA identified inconsistencies and variability in RT₂₅ values between formulated products of the same pesticide active ingredient. EPA also noticed that these data did not appear to be correlated with chemical/physical characteristics of the pesticide active ingredient. The Pollinator Research Task Force (PRTF), in collaboration with EPA, has taken the task to review the current test design (OCSPP 850.3030) and work with different stakeholders to improve the method, and ensure the reliability and predictive nature of RT₂₅ data. The PRTF was formed in January 2016 and is comprised of eight pesticide registrants, namely BASF Corp., Bayer Crop Science LP, Corteva Agrosciences, FMC Corp., Mitsui Chemicals Agro. Inc., Syngenta Crop Protection LLC, UPL NA, Inc. and Valent USA Corp. with the focus of mining and generating data to refine and improve pollinator risk assessments in North America and globally, where applicable.

Summary of OCSPP 850.3030 test design:

The honey bee toxicity of residues on foliage study (OCSPP 850.3030) is a laboratory/field test designed to determine the length of time over which field-weathered foliar residues remain toxic to adult honey bees. The test substance (a typical end-use product; TEP) is applied to crop foliage (e.g., alfalfa); the foliage is then harvested at predetermined intervals post-application,

and test bees are caged along with the treated foliage for 24 h. The treated foliage, which has typically been “weathered” under ambient field conditions, is harvested 3, 8 and 24 h post application. If mortality of bees exposed to the foliage harvested 24 h after the application is greater than 25%, additional weathered, treated foliage samples continue to be taken every 24 h (*i.e.*, 48, 72, 96, 120 h, *etc.* post-application) and bees are then exposed to these additional samples for 24 h until mortality of bees exposed to the treated foliage is 25% or less. Results are expressed in terms of the length of time (in hours) required to reduce mortality in exposed bees to 25% or less following application at a specific rate of application (lb a.i./A).

Evaluation of current test design:

Some of the variability in RT₂₅ data from different TEP products containing the same active ingredient may be explained by the inert ingredients within formulated products which may affect the dissipation of the active ingredient and therefore the length of time that residues remain toxic to bees. However, the PRTF believes that the major sources of variability are inherent in the test design, since OCSPF 850.3030 does not adequately specify various test parameters which could influence exposure, leaving room for interpretation by the testing facility. As a result, different laboratories conducting these studies include different parameters in their study protocols.

During the initial review of the current study design, the PRTF has identified the following potential sources of variation in the RT₂₅ data:

- Use of variable test cage sizes which potentially lead to inconsistent exposure.
- Placement of treated foliage in cages.
- Inconsistencies in product application, crop condition, and ambient field conditions, including environmental parameters during weathering in the field. Examples of inconsistencies are listed below:
 - Crop grown in the field versus grown in flats in greenhouse.
 - Variable age of foliage used in the test. The type of alfalfa used, including smooth vs. hairy types, and erect vs. creeping.
 - Product application in the field versus application in lab using a spray booth.
 - No recommendation for environmental parameters during weathering in the field.
 - No guidance on whether surfactants should or should not be used.
- Lack of a true positive control (reference toxicant).
- Current residue aging intervals (*i.e.*, 3, 8 and 24 h post application) do not fit well with the EPA's Acute Risk Mitigation Policy. New protocols need to include 6 h as one of the weathering intervals.

The proposed project has been divided into phases: short-term improvements, and long-term improvements. The initial efforts focused on increased “standardization” of the test guideline. Results from Phase I of the project were still variable, and indications are that applications in the field were the source of variability in the test. There are two potential sources of variability; differences in application equipment, potentially leading to inconsistent distribution of the test material over the treated plots and/or different environmental conditions resulting in different dissipation/degradation rates in the treated plots.

Phase II of the project will involve two laboratories that are in close proximity to one another. The coordination of crop planting and application timing between the two facilities will allow for the impact of the environmental conditions to be evaluated. It will also allow for the facilities to share samples of alfalfa treated at each facility to confirm consistency of results during the in-life phase of the test.

The two participating laboratories will be:

- Smithers– Snow Camp, NC
- Eurofins US lab – Mebane, NC

Project goal: Standardize the study design to enhance the consistency, reliability, and utility of RT₂₅ data to pesticide regulatory agencies, registrants, and eventually pesticide users. The focus will be to “standardize” and evaluate sources of variability during the field portion of the test.

Protocol for Phase II of the RT25 Project: see the next page

Protocol for Phase II: Standardization of Honey Bee Toxicity of Residues on Foliage (RT₂₅) Study Design

Based on EPA's Ecological Effects Test Guideline OCSPP 850.3030, dated January 2012, with modifications

1. **Purpose:** This guideline is intended for use in developing data on the residual toxicity to honey bees of chemical substances and mixtures ("test chemicals" or "test substances") subject to environmental effects testing requirements. This guideline describes a toxicity test in which honey bees are exposed to weathered residues of a test substance on treated foliage.
2. **Definitions:**
 - a) Acute Residual Toxicity is the adverse effects occurring over a period of time (hours or days) from a single dose of the test substance to foliage.
 - b) Dose is the amount of test substance applied. Dose is expressed as a mass, pounds of test substance per acre (lbs/A) and for a pesticide, pound(s) of active ingredient applied per acre (lbs a.i./A). The dose used in this test should be the maximum, single application dose allowable according to the end-use product labeling.
 - c) Mortality: an animal is recorded as dead when it is completely immobile (e.g., no movement within 5 seconds).
 - d) RT₂₅ is the residual time needed to reduce the activity of the test substance and bring bee mortality down to 25% in cage test exposures to field-weathered spray deposits (see paragraph (e)(2) of this guideline). The time period represented by this toxicity value (RT) is considered to be the length of time (in hours) that the test substance is expected to remain toxic by contact to bees in the field, when bees are exposed to weathered residues of the test substance on vegetation at an expressed rate of application (lb a.i./A). Exposure to weathered residues in the laboratory are a surrogate for field conditions.
3. **Summary of test:** The honey bee (*Apis mellifera*) foliar residue study is a laboratory test designed to determine the length of time over which field-weathered foliar residues remain toxic by contact to honey bees. The test substance (e.g., a typical end-use product) is applied to crop foliage, the foliage is harvested at predetermined intervals post-application, and test bees are caged on the treated foliage. Results are expressed in terms of the length of time (observed time interval) following application, during which residues continue to cause 25% mortality (RT₂₅) in test populations at an expressed rate of application (lb a.i./A).
4. **General test guidance:** Based on EPA's Ecological Effects Test Guideline OCSPP 850.3030, dated January 2012, with some modifications.
5. **Definitive test:** The goal of the definitive test is to determine the 24-h RT₂₅, length of time post-application that residues of the test substance on foliage are toxic to honey bees. For this determination, one treatment level, the maximum rate on the label and at least three different time intervals between application and harvest are typically used. The test substance will be evaluated at the labeled maximum, single application rate. A summary of test conditions is provided in Table 1, and validity elements for an acceptable definitive test are listed in section 11 of this protocol.

6. Test specifications:

6.1. Test organism:

- a) **Species:** Honey bee, *Apis mellifera*, is the test species.
- b) **Source:** Bees may be obtained from on-site colonies or from a commercial apiary. All control and treatment bees used in a test should be from the same source and breeding lineage. Bees are emerged from brood frames taken from the source colonies in an incubator (34-35 °C, 45-90% humidity) and reared for three to five days with "bee bread" (pollen that is already stored on the brood frame) supplemented with pollen patty and 50% w/v sucrose in water solution. In order to obtain a sufficient number of bees with known age (3-5 days post-emergence), brood frames can be collected from multiple colonies within the same apiary. Collection in early spring or late autumn should be avoided, as the bees have a changed physiology during this time.
- c) **Age:** The test should be conducted using young adult worker bees that are of a similar age (three to five days post-emergence) and feeding status.
- d) **Health status:** Bees used in the test should be in apparent good health. Only bees from apparently disease-free colonies should be used, and they should be kept in conditions conforming to proper culture practices. Bees treated with chemical substances, such as antibiotics, anti-varroa, *etc.*, should not be used for toxicity tests for four weeks from the time of the end of the last treatment.
- e) **Care and handling:** During holding and testing, bees should be shielded from excessive activity, handling stress or other disturbances and kept in the dark. Bees should be handled only as much as is necessary to conform to test procedures.
- f) **Diet and feeding:** A 50% weight/volume (w/v) or weight/weight (w/w) solution of sugar/water (500 grams/liter) is provided *ad libitum* throughout the holding and test periods. Purified or distilled water should be used for preparation of the sugar solution. Top feeding is preferred, so for the ring test, the feeding syringe/tube should be inserted through an opening in the top of the test cage. Attention should be paid to avoid any contact between the feeders and the treated foliage.

6.2. **Test crop:** The test crop will be alfalfa (*Medicago sativa*). Alfalfa should be grown in an unshielded open outdoor field location. Foliar applications of the test substance should be performed when the alfalfa crop is between **20-40 centimeters** in height. To ensure harvest is not impeded by excessive weed growth, pre-emergence and early post-emergence herbicide applications may be made to the cropped area. Applications of any maintenance pesticides (herbicides, fungicides, insecticides) must not be made within 4 weeks of the start of the study. Fertilizer and irrigation treatments may be made as needed consistent with good agronomic practices up to 24 hours before start of the study but must not be made during the study. All agronomic practices, variety of alfalfa, the seeding rate, date of planting, fertilizer, irrigation and pesticide treatment history for the three years prior to the start of the study, should be reported. If seeds treated with seed-applied pesticides are used to establish the crop, the field should not be used for RT₂₅ studies for 1 year from planting.

6.3. **Test duration:** The test starts with the placement of weathered treated foliage into cages with bees, followed by a 24-h observation period during which mortality and clinical signs of toxicity are recorded at 4±1 and 24±1 h post-exposure.

- a) **Post-treatment weathering intervals:** The treated foliage should be harvested at minimum two mandatory intervals 6 ± 1 and 24 ± 1 h post-application, and placed in cages to expose young adult honey bees to the weathered residues of test substance. Based on the results from Phase I of this project, the 3-hour harvest interval will be excluded since 100% mortality was observed in both facilities with the same test substance at the 6-hour harvest interval. The two labs will coordinate test initiation so that the bioassay phases begin within 1 hour of each other. If mortality of bees exposed to the foliage harvested 24 h after the application is greater than 25% (control-corrected), weathered, treated foliage samples should continue to be collected and tested at 24-h intervals until the mortality is $\leq 25\%$ (control-corrected), up to five days post-application. For the ring test, the treated foliage will be harvested at 6 ± 1 and 24 ± 1 h post-application intervals, with option of 48 h, 72 h, 96 h and 120 h intervals if mortality stays $>25\%$.
- 6.4. **Observation period:** Bees will be observed for **24 h** after the bees and treated foliage are placed onto the cages.
- 6.5. **Test facilities:** Test substance application and weathering should occur outdoors under natural field conditions. The bee exposure portion of the test should be conducted indoors to control lighting and other environmental variables, while bees are being maintained in small test cages. The cages containing honeybees should be placed in an environmental chamber to control temperature and relative humidity.
- 6.6. **Sample Sharing:** At each harvest interval, the PRTF will arrange for samples of treated alfalfa to be transported from one of the facilities to another. Due to the close proximity of the facilities in the State of North Carolina, the transportation of samples should take less than 1 hour. Both facilities will conduct bioassays on subsamples from the same harvested foliage. The laboratories will coordinate the start time of the bioassays so that they begin within 1 hour of each other.
- 6.7. **Test cages:** Use of test cages with different dimensions could potentially lead to inconsistent exposure. So, for the ring test, each CRO will use a standard cage to remove this as a source of variability. Determining an optimum cage design was part of Phase I of the ring test. The test cages should have a suitable opening for the introduction of treated foliage and bees, and another opening at the top for inserting the feeding syringe/tube. Cages should be cleaned thoroughly between uses or new cages are used for every trial. For this ring test, transparent 32 oz plastic containers (upper diameter = approx. 11 cm, base diameter = approx. 9 cm; height = approx. 14 cm) will be used as test cages (see Fig .1). The top of the test cage will be covered with a screened lid to allow ventilation and has an opening for inserting a feeding syringe.
- 6.8. **Collection of bees:** The day prior to exposure, young bees should be collected from frames kept in the incubator and acclimated for approximately 24 hours. The bees can be acclimated in bulk or acclimated in the actual test cages. If the acclimation occurs in the test units/cages dead and impaired bees should be removed and, if needed, replaced by healthy bees from the same pool of newly emerged bees prior to the introduction of the test foliage. If acclimation occurs in the test cages, it is recommended that excess bees be acclimated in excess test cages in case there is a need to replace dead or impaired bees prior to test initiation. Introduction of bees into the test cages shall be done in an indiscriminate manner. During transfer to the exposure cages, immobilization of bees with cold temperatures, carbon dioxide gas (CO_2) or nitrogen gas (N_2), may be necessary but should be kept to the minimum.

- 6.9. **Controls:** Paired negative (untreated) controls are included in the test. Control crop foliage is treated with water only and identically to treatment plots, except for applications of the test substance. Control and test bees are kept under the same environmental conditions.
- 6.10. **Number of test organisms and replicates:** Six replicates should be assigned to each treatment and control group at each post-application interval, with a minimum of 25 bees for each replicate. Test organisms should be impartially assigned to different treatment groups.
- 6.11. **Test substance:** The substance to be tested will be Dimethoate 400 EC
- 6.12. **Application of test substance:** The test substance will be applied at the maximum single application rate of 0.5 lbs. a.i./acre (spray coverage = 200 L mix/ha). A single application should be made in the morning after the dew has dried and when alfalfa crop is between 20-40 centimeters in height. Application should be made in the field with a tractor mounted or hand-held boom sprayer, using standard nozzles in accordance with regionally accepted practices. The sprayer should be calibrated on the day of, or a day prior, to the spraying of the plants. Spray tank solutions should be continuously stirred or circulated prior to and during use. Nozzle height above the crop during application should be maintained consistent with manufacturer recommendations and will be coordinated between the two facilities for consistency. Wind speed should be less than 3 m/sec during application. Spray equipment should produce a wide enough swath so that the alfalfa plots can be treated in single-pass spray. Detailed aspects of the application shall be reported including nozzle type, spacing, height above crop canopy, flow rate, pressure, application speed and pass times, nominal and actual volumes applied, results of equipment calibration, volumes and concentrations of spray solutions prepared. Environmental conditions during application shall be recorded including air temperature, relative humidity, soil moisture, presence/absence of dew or moisture on the crop, cloud cover, wind speed, application time of day (beginning and end of spraying), time of sunrise and sunset and any other relevant observations that may affect the interpretation of the results.
- 6.13. **Application timing:** Phase II of this project will likely consist of two separate coordinated applications at each test facility at different times during the year in order to evaluate the impact of environmental conditions in the field on the test results. The first application at each facility will be targeted for late May/early June, during a period of time when it is generally hot and dry in North Carolina. The two facilities will coordinate the planting and treatment of alfalfa so that the applications will occur within two weeks of one another, under similar environmental conditions. Applications on the exact same day will be avoided so that sample shipment and bioassay conduct will more easily be coordinated within each individual facility. A second application at each facility will be targeted for late July/early August, during a period of time when it is generally very humid in North Carolina. Once again, the two facilities will coordinate the planting and treatment of alfalfa so that the applications will occur within two weeks of one another, under similar environmental conditions.
- 6.14. **Field plots and harvest of foliage:** Plots should be at least 1 m² (10.8 square feet) in alfalfa grown according to standard agricultural practices. Applications of any maintenance pesticides (herbicides, fungicides, insecticides) must not be made within 4 weeks of the start of the study. At a minimum, nine test substance treatment plots are used to obtain three plots for harvesting at each time interval (6±1 and 24±1 h post-application). After test substance residues have aged (weathered) for the appropriate

time period, alfalfa foliage sufficient to place in six treatment cages at each facility (approximately 180 g fresh weight or 6,000 cm³ total), should be harvested from three treated test plots using hand equipment, placed individually in labeled bags and returned immediately to the laboratory for processing and placement in test cages or transport to the other test facility. Foliage should be collected, using a random sampling scheme, from the top 15 cm of the canopy. Minimum distance of 10 m should be kept between treatment and control plots to avoid potential contamination of control plots due to drift. At each of the minimum time intervals, three alfalfa samples are harvested from the control plot using a random sampling scheme, to obtain sufficient foliage to place in six control cages at each test facility. If additional harvest intervals are required beyond the minimum two, control samples must be collected and tested also at each harvest interval.

- 6.15. **Preparation of treated foliage:** Samples of foliage are returned to the laboratory in bags and transported in coolers that should be held between 8 and 12 °C once the coolers are filled and closed. Temperature data loggers should be included in the coolers. The samples are mixed thoroughly and then divided into approximately 15 g or 500 cm³ portions. The current guideline recommends chopping the foliage into smaller (2.5 cm) lengths and loosely placing 15 g portions at the bottom of each test cage, but after the discussions with the project team it was concluded that this step is not necessary and should be avoided. For the ring test, leave the foliage in 12-15 cm lengths and loosely place 15 g portions upright/diagonally in each test cage to maximize the exposure.
- 6.16. **Introduction of the bees to the treated foliage in the cages:** Bees should then be released on the top of the foliage or the treated foliage added directly into the test cages if the bees are being acclimated in the test cages. Special attention should be paid to avoid any direct contact between the sugar solution feeders and the treated foliage.
- 6.17. **Sampling for residue analysis:** An approximately 15 g sample of the treated and untreated control foliage immediately after the spray has dried (approximately 1 hour \pm 30 minutes) and at each harvest interval will be collected to confirm test substance concentration. If the study extends past 24 hours, then samples of foliage will continue to be taken at each 24-h interval thereafter, to correspond with the exposure, up to 5 days post-application. Fresh sample weights should be recorded before freezing the samples. In addition, for the ring test, analytical evaluations will also be conducted on spray solution (*i.e.*, tank mix) and three spray cards (preferably glass fiber discs) placed randomly in the test plots for the application. The spray solution sample should be collected after completion of the application. The spray cards should be held in a horizontal position at the top height of the crop canopy so that it gets the full rate of the spray without interception by the crop. At the time of collection, the spray cards should be folded and placed into plastic bags similar to those used for foliage collection. Two tank mix samples, 50 ml each, will be collected upon completion of the application and labeled A and B. Tank mix sample A will be analyzed for rate verification, and sample B will be retained for further analysis if needed. Samples are to be transported from the field and subsequently deep frozen until shipment to the designated analytical laboratory. Samples should be shipped to the designated analytical laboratory deep frozen.
- 6.18. **Environmental conditions:**

a) **Environmental conditions during application and weathering in the field:**

Sunlight, precipitation and temperature are three extremely important factors in the dissipation of pesticide residues. Test substance application should be made preferably on clear days with maximum temperatures ranging between 20-40 °C and <30% chance of precipitation. Application should happen in the morning after dew or moisture from any overnight rains has dried off. Test plots should be protected from direct precipitation for at least 3 h (up to 6 h) following the application. If rainfall should occur, the test plots should be sheltered from direct rainfall using a tarp or other suitable canopy. If a canopy is used, it should be removed 3 h (up to 6 h) after application to allow full effect of natural weathering to take place (*i.e.*, direct sunlight). Also, application should be avoided in windy conditions (*i.e.*, average wind speed >3 m/s) to avoid contamination of untreated control plots. Treated test crop should be allowed to weather outdoors under natural field conditions.

b) **Environmental conditions during exposure phase:** Environmental parameters in the laboratory during the bioassays should be maintained as follows:

- I. Temperature and humidity. Temperature should be maintained between 25 and 35°C, with relative humidity between 50% and 80%.
- II. Lighting and photoperiod. It is recommended that test bees be maintained in the dark except during transfer to test cages and observations.
- III. Test cages, including treated and control cages, are placed within the incubator in a randomized pattern which is also recorded.

7. **Observations:**

7.1. **Analysis for test substance concentrations:** Test substance residues on treated foliage are expressed in parts per million (ppm; mg ai/kg foliage) fresh weight. For the ring test, analytical evaluations will also be conducted on spray solution (*i.e.*, tank mix; mg a.i./L) and three spray cards placed randomly in test plots during the application (analyzed as mg a.i./cm² and also reported in units of lb a.i./acre). The residue analyses for the trials will be conducted at one designated lab to avoid inter-lab variability.

7.2. **Field site conditions:** Environmental conditions should be monitored at the field site at the time of test substance application and during weathering period. Environmental information to be collected should include daily minimum and maximum air temperature, precipitation, and relative humidity. Wind speed and estimated cloud cover should be recorded at least at the time of application. A data-logging weather station shall be placed on site, within 1 km of the application area, to collect environmental data.

7.3. **Conditions during exposure in the lab:** Temperature and relative humidity should be recorded during the bee exposure in laboratory test cages.

8. **Measures of Effects:**

8.1. **Mortality:** For a given weathered residue treatment or control, bees should be observed for mortality at least once at 4±1 h after exposure and at exposure termination at 24 h. Dead bees should not be removed from the test cages until the test is terminated.

8.2. **Appearance and behavior:** For a given weathered residue treatment or control, bees should be observed for all clinical signs of intoxication and any other abnormal behavior once during the first 4±1 h after exposure and at test termination (24 h). Observations should be recorded by treatment level and by time of occurrence. Signs of intoxication

are those behaviors apparently due to the test substance and may include a wide variety of behaviors, such as ataxia, lethargy, excessive cleaning, tremors, convulsions and hypersensitivity (agitation). Prior to the evaluation at test termination, observations should be made without disturbing or removing bees from the test chambers; for these observations, estimates of mortality and effects are sufficient.

9. Treatment of results:

9.1. Descriptive summary statistics:

- a) Environmental conditions: Data should be summarized in tabular form, showing the range and mean temperature, precipitation, relative humidity, and wind speed.
- b) Mortality. Data should be summarized in tabular form, showing for each weathered age of foliage treatment and control the number of bees initially exposed, mortality at each observation time, and the percent mortality. Average mortality in the controls, if any, will be used to correct the mortality observed in the treatments using Abbott's formula.
- c) Appearance and behavior. Data should be summarized in tabular form, showing for each weathered age of foliage, appearance and behavior at each observation time. Statistical analysis of sublethal effects are not conducted.

9.2. Residual Time (RT_{25}): A test for comparing two paired populations (*e.g.*, paired t-test) should be performed to detect significant ($p < 0.05$) difference of treatments from controls. Abbott's correction should be used in the event of control mortality. Additional discussion about measurement endpoints and statistical procedures is found in OCSP 850.3000.

10. Tabular summary of test conditions: Table 1 lists the important conditions that should prevail during the definitive test. Meeting these conditions will increase the likelihood that the completed test will be acceptable or valid.

Table 1. Summary of Test Conditions for Honey Bee Toxicity of Residues on Foliage Test

Test type	Toxicity of residues on foliage
Test duration	24 h observation period for each aged residue interval (6±1 and 24±1 h aged residue intervals are tested; additional 24 h residue intervals may be appropriate).
Temperature during laboratory exposure	25 - 35°C
Relative humidity during laboratory exposure	50 – 80%
Lighting	Darkness, except during transfer of bees to treatment cages and observations
Test chamber	32 oz plastic cages with an upper diameter approximately 11 cm, base diameter of approximately 9 cm and height of approximately 14 cm will be used in the ring test
Foliage cutting length and placement	Foliage lengths of 12-15 cm; upright/diagonally placed in test cages
Test substance application	15-g or 500-cc portions of treated foliage placed in a test cage
Age of test bees	Young adult worker bees of similar age (1-5 days post-emergence) and feeding status
Number of bees per chamber	25 (minimum)
Number of bees per treatment and control	150 (minimum)
Number of treatments	Minimum of 2 treatment groups (6±1 and 24±1 h post-application of maximum single application rate) which includes the negative control(s). Additional intervals may be appropriate if mortality is >25% for the 24 h post- application treatment
Feeding	50% sugar/water (w/v) solution <i>ad libitum</i>
Measure of Effect or Measurement Endpoint	RT ₂₅ based upon mortality at 24 h after bees are exposed to foliage. If mortality of bees exposed to the foliage harvested 24 h after the application is greater than 25%, additional weathered, treated foliage samples will continue to be taken every 24 h.

11. **Test validity criteria:** The definitive test will be considered invalid if one or more of the following conditions occurred -

- a) Test bees were not of similar age and feeding status.
- b) More than 20% mortality averaged across control treatments.
- c) All bees in a test were not from the same source (apiary) and breeding lineage.
- d) Concurrent negative (untreated) controls were not included in the test.
- e) Environmental conditions (temperature, precipitation, relative humidity, wind speed and cloud cover) at the field site were not monitored/reported.
- f) Test organisms were not impartially assigned to test cages.
- g) Substances, other than the test pesticide were applied to the growing alfalfa within 4 weeks of test initiation.

12. **Reporting:**

12.1. **Protocol deviations:** Include a description of any deviations from the test protocol or any occurrences which may have influenced the results of the test.

12.2. **Test substance:**

- a) End-use product (name, state or form, source), its purity (for pesticides, the identity (common name, IUPAC and CAS names, CAS number) and concentration of active ingredient(s)) and known physical and chemical properties that are pertinent to the test.
- b) Storage conditions of the test substance.
- c) Methods of preparation of test substance for application onto foliage, the maximum label rate, and the actual application rate (lb a.i./A) with the finished spray volume per acre.
- d) Describe the stability of the test substance under storage conditions.

12.3. **Test organisms:**

- a) Scientific name, race, and source.
- b) Culture method and conditions.
- c) Health status of colonies used for collection of test bees (e.g., any adult diseases, use and application date(s) of any prophylactic or preventative treatments).
- d) Collection method and date of collection.
- e) Holding period.
- f) Age at initiation of exposure to an aged residue treatment.

12.4. **Test system and conditions:**

- a) Description of housing conditions: type, size, and material of test cages.
- b) Description of any feeding during the test (if applicable), including: method, type of food, source, amount given and frequency.
- c) Common and scientific name of treated crop.
- d) Plot size, and method and time of administration of test pesticide on plots.

- e) Number of aging intervals tested.
- f) Time after application to plot of foliage collection (age intervals tested) and placement of foliage in test chambers.
- g) Plots per aging interval and negative control.
- h) Number of bees per test cage.
- i) Number of cages (replicates) per aging interval plot and negative control plot.
- j) Methods used for test cage and treatment randomization as well as methods for impartial assignment of bees to test cages.
- k) Exposure duration to a given aged residue and duration of the study.
- l) Methods and frequency of environmental monitoring performed on treated plots during administration of test substance and weathering period for temperature and precipitation, and any other known weather conditions that would impact initial concentration or stability of residue levels on treated plots.
- m) Methods and frequency of environmental monitoring performed during the definitive study or positive control study for test room temperature, humidity and lighting.
- n) For the definitive test, all analytical procedures and preservation methods should be described. The accuracy of the method, method detection limit, and limit of quantification should be given.

12.5. **Results:**

- a) Laboratory environmental monitoring data results (test room temperature, humidity and lighting) in tabular form (provide raw data for measurements not made on a continuous basis), and descriptive statistics (mean, standard deviation, minimum, maximum).
- b) Field site environmental monitoring data results (temperature, precipitation, wind speed, relative humidity, cloud cover) in tabular form (provide raw data for measurements not made on a continuous basis), and descriptive statistics (mean, standard deviation, minimum, maximum).
- c) For the bioassays, the number of dead bees which were observed at least once during the first 4 hours of exposure and at 24 h (provide the raw data).
- d) For the bioassays, a description of signs of intoxication and other abnormal behavior, including time of onset, duration, severity, and number affected at each aged residue treatment and control(s) (provide the raw data).
- e) Provide 24-h RT_{25} values.
- f) Description of method used, including software package, for determining the 24-h RT_{25} value.
- g) Results of analysis of variance (ANOVA) to detect significant differences of treatment groups from the controls.

13. **References:** The references in this paragraph should be consulted for additional background material on this test guideline.

- a) Abbott, W.S., 1925. A method of computing the effectiveness of an insecticide. *Journal of Economic Entomology* 18:265-267.

- b) Johansen, C. *et al.*, 1977. Bee Research Investigations. Dept. of Entomology, Washington State University, unpublished, 22 pp.
- c) Lagier, R.F. *et al.*, 1974. Adjuvants Decrease Insecticide Hazard to Honey Bees. College of Agriculture Research Center, Washington State University Bulletin 801, 7 pp.
- d) Mayer, D. and C. Johansen, 1990. Pollinator Protection: A Bee & Pesticide Handbook. Wicwas Press. Cheshire, CT.
- e) Mayer, D. (approved by), 1996. Standard Operating Procedure (SOPs) – Residue Bioassay. The Bee Group-Irrigated Agriculture Research and Extension Center. Prosser, WA.
- f) U.S. Environmental Protection Agency, 1982. Pesticide Assessment Guidelines Subdivision L Hazard Evaluation: Nontarget Insects. Office of Pesticides and Toxic Substances, Washington, D.C., EPA-540/9-82-019.
- g) U.S. Environmental Protection Agency, 1985. Hazard Evaluation Division Standard Evaluation Procedure, Honey Bee—Toxicity of Residues on Foliage. Office of Pesticides Programs, Washington, D.C., EPA-540/9-85-003.
- h) USEPA 2012. Ecological Effects Test Guidelines OCSPP 850.3030: Honey Bee Toxicity of Residues on Foliage. Office of Chemical Safety and Pollution Prevention (7101). EPA 712-C-018. January 2012.
- i) USEPA. 2012. Ecological Effects Test Guidelines OCSPP 850.3020: Honey Bee Acute Contact Toxicity Test. Office of Chemical Safety and Pollution Prevention (7101). EPA-712-C-019. January 2012.
- j) EPA. 2017. U.S. Environmental Protection Agency's policy to mitigate the acute risk to bees from pesticide products. Office of Pesticide Programs. January 12, 2017. EPA-HQ-OPP-2014-0818-0477.

Next steps:

- Finalization of phase II ring test protocol: May 2021
- Experimental phase(s) of ring test: May-June, 2021, and July-August, 2021
- Transfer of residue samples to designated analytics lab: End June 2021 and end of August 2021
- Data submission by participating CROs to designated 3rd party: September, 2021
- Data evaluation and analytical analysis of residue samples: October, 2021
- Draft report: December, 2021
- Decision on phase III improvements: 1Q 2022
- Phase III improvements: 2022 onwards

Fig 1. Test cage



Transparent 32 oz plastic containers (upper diameter = approx. 11 cm, base diameter = approx. 9 cm; height = approx. 14 cm) will be used as test cages. The top of the test cage will be covered with a screened lid to allow ventilation and has an opening for inserting feeding syringe.

TEST SYSTEM RECEIPT

Daily Log (Date & Initial each entry)
--

Frame removed from hive 100 containing capped brood, pollen, nectar, and honey on 02 June 2021. No miles or signs of disease observed in hive. Frame placed into ^{WEA 07 June 2021} ~~that~~ the emergence chamber inside ECO6. ECO6 provides complete darkness when door is closed. ACO 02 June 2021

03 June 2021: no bees seen emerged from brood frame yet AW 03 June 2021

07 June 2021: bees indiscriminately placed into cages for Smithers Flehr foliage exposure. 13 cages were populated with 25 bees each and provided with a 2mL syringe of 50% sugar syrup Lot: 07Jun21A-50. Also 07 June 2021

08 June 2021: bees indiscriminately placed into cages for Smither's + 24 hr exposure, EuroFins + 6 hr exposure, 13 cages each and provided with a 2mL syringe of 50% sugar syrup Lot: 075np21A-50. AW 08 June 2021

09 June 2021: bees indiscriminately placed into cages for Eurofins
+24 hr sample exposure, 13 cages populated with 25 bees
each and provided with a 2mL syringe of 50% sugar
syrup lot: 07Jun21A-50. AW09Jun2021

NA [REDACTED] 09 Nov 2011

Daily Log (Date & Initial each entry)

12 Sep 2021: 1 frame removed from hive 107 and placed into the environmental chamber ECO6. No mites or signs of disease observed ECO6 provides complete darkness 10 Sep 21 AW

13 Sep 2021: 1 additional frame removed from hive 105 and placed into ECO6, no signs of pests or diseases seen 13 Sep 21 AW

13 Sep 2021: 25 bees placed into 13 cages (6 control 6 treatment, 1 extra replicate cage for [REDACTED] 1hr time interval exposure 13 Sep 2021 AW

14 Sep 2021: 25 bees placed into 13 cages (6 control 6 treatment, 1 extra replicate) for [REDACTED] 24hr time interval exposure 14 Sep 2021 AW

15 Sep 2021: 25 bees placed into 13 cages (6 control, 6 treatment, 1 extra replicate) for [REDACTED] 1hr time interval exposure AW 15 Sep 2021

16 Sep 2021: 25 bees placed into 13 cages (6 control, 6 treatment, 1 extra replicate) for [REDACTED] 24hr time interval exposure AW 16 Sep 2021

17 Sep 2021: 25 bees placed into 13 cages (6 control 6 treatment, 1 extra replicate) for [REDACTED] 48hr time interval exposure. AW 17 Sep 2021

NH [REDACTED] 09 Nov 2021

NA [REDACTED] 09 Nov 2021

NOTE TO FILE

colony 20-A-10
AW 11 Apr 2020

Study Number or Logbook Name: 14198.4100 Animal Receipt

Describe situation or observation:

4, 3lb packages of bees picked up on 11 Apr 2020 from The Carolina Honey Bee Company located in Traveler's Rest, SC. The packages were healthy with healthy live queens. The packages were placed in hives: 100, 102, 105, 107. Hives 100, 102, 107 have queens marked with a blue dot, Hive 105 has an unmarked queen. Hives installed by [redacted] from 16:00 - 17:00. Hives ^{IF AW 11 Apr 20} fed approx 5L of 1:1 sugar syrup. Hives closed, with plastic barriers blocking screened bottom boards because temperatures expected to be cool. Hives will be opened and checked for health/status in 2-3 days depending on weather.

This will be designated colony 20-A-10 for all four hives.

Date and initials of recorder: AW 11 Apr 2020

Study director or management assessment (if needed):

Date and initials of study director/management:

colony 20-A-10



10 S. Main St
Travelers Rest, SC 29690
864-610-2337



QTY	Description	PAID
4	3 lb Package Bees – MARKED	
	Extra Queen	
	Extra Un Marked Queen	
		\$500.00

All of our packages are put together with the utmost care. In the event you find a problem with your queen at the time of installation, DO NOT remove the queen, cork or candy. Please call 864-610-2337. Please leave a message if no answer and we will get back to you as soon as we can.

Please inspect your packages before leaving with them. We are, sorry, but we cannot be responsible for them once they leave. Please make sure when traveling with them that they have ample ventilation and keep them out of direct sunlight. We will not be able to replace packages that leave here in good condition.

021

Test System/Colony Receipt Log

Species: Apis melliferaDate Shipped: 08 May 2021Date Received: 08 May 2021Birth Date: N/AAge upon receipt: N/A weeks 08 May 21 daysAnimals were assigned to Room No (s): CRC IE 08 May 2021
CRS Apiary upon receiptAnimals were assigned to Study No (s): 13049-4115
PRIF Ring test upon receipt

Supplier information: Name, Address, Phone

Check if birds raised at CRC: ☐Carolina Bee Company
14 Center St. Traveler's Rest, SC
29690Vet check required? N

Check tests conducted:

☐ Mycoplasma synoviae
☐ Mycoplasma gallisepticum
☐ Salmonella pullorum
☐ Salmonella Group D / Enteritidis
☐ Avian Influenza

Test results maintained in SMV CRC Animal Receipt Log

Documents included with shipment:

Package Bee Pickup Slip

Number of Animals Upon Receipt:

Males Females N/A TotalAlive Alive 08 May 2021Dead Dead DeadQuarantine Information: Start Date: N/AEnd Date: 08 May 21

Describe housing at beginning of quarantine:

2 empty hives, old equipment
but cleaned well and verified
to be secure

Health Observations and Comments: (note date and initials for any observations or comments. A health observation must be made and noted on this form during quarantine. Use additional pages as necessary and attach to this form)

2 3-lb packages of honey bees picked up from
Carolina Bee Company. Each package had 1 healthy
marked (white) queen. Opened package, removed candy cork
on queen cage (cork only, candy plugs remaining), placed queen cage
between 2 frames. Placed package into hive so they can
crawl out towards queen, packages will be removed and 5
frames that were removed to fit the package will be placed back into
hives in 1-2 days.Form completed by (initial and date): 08 May 2021Form (045) completed (check): N/ANot needed
for this species
AW 08 May 2021

Package Bee Pickup Slip

5/08/21

Name:

Email:

Number of Packages: 2


Additional Queens: Marked

Unmarked

*** Please inspect your packages before leaving with them. We cannot be responsible for them once they leave our possession. Please make sure when traveling that they have ample ventilation and keep them out of direct sunlight. We will not be able to replace packages that leave here in good condition.

Our packages are put together with the utmost care. In the event you find a problem with your queen at the time of installation into your hive **DO NOT** remove the queen or the cork or the candy. **DO NOT** shake or install your package bees. Please call us ASAP **BEFORE 6 PM SUNDAY** if no answer leave a message and we will contact you as soon as possible.



Signed:

 OS May 2021

023

FEED INFORMATION

Sugar Solution Log

Sugar Lot	Water ID	Sugar Weight (g)	Water Volume (mL)	Scale	Syrup Lot ^a	Date Mixed	Date Expires	Initials
5015B-3-01: 06AM 002322082	distilled water	100g	200mL	E030	07JUN21A-50	07 June 2021	11 June 2021	
5015B-3-01: 06AM 002322082	distilled water	100g	200mL	E033	13Sep21A-50	13 Sep 2021	17 Sep 2021	
<div style="position: relative; height: 100px;"> NA 09 Nov 2021 </div>								

^aWhen creating lot numbers for sugar syrup use the format: date, unique letter, -concentration. For example, if two samples of 50% sucrose are prepared on 07 Sep 2018, they could have lot numbers of 07SEP18A-50 and 07SEP18B-50.

Basal Feed and Feed Ingredient Documentation*

21-10

Circle One: Basal Feed or Feed Ingredient

Basal Feed Type/Feed Ingredient:	<u>Granulated sugar</u>
Date of Receipt:	<u>15 Feb 2021</u>
Supplier:	<u>Teagues Farm and Market</u>
Lot Number:	<u>5015B-3-01:06AM 002322082</u>
Size of Container:	<u>20, 50 lb bags</u>
Number of Containers:	<u>20</u>
Gross weight received:	<u>1000 lbs sugar</u>
Expiration date:	<u>15 Feb 2023</u>
Feed placed under noted conditions :	<input checked="" type="checkbox"/> Ambient <input checked="" type="checkbox"/> Away from test articles and test diets <input type="checkbox"/> Refrigerated <input type="checkbox"/> Frozen <input checked="" type="checkbox"/> Free of rodent/insect contamination
Completed by (date/init.):	<u>15 Feb 2021</u>

Screen Required (circle one):	<u>Contaminant</u> Proximates None
Sample No.	<u>23518 CL</u>
Date Sample Taken:	<u>22 MAR 2021</u>
Date sample sent for analysis:	<u>22 MAR 2021</u>
Analytical lab used:	<u>Columbia Laboratories</u>
Completed by (date/init.):	<u>22 MAR 2021</u>
Note: If Diet Ingredient, the Contaminant Screen described in SOP 7.10, is not required, but may be for study-specific purposes (check individual protocols). A Contaminant Screen may not be required for all Diet Feeds (see provisions in SOP 7.10).	

026



Photo taken from one bag in shipment of 20 bags
of granulated sugar. Sugar received 15 Feb 2021

15 Feb 2021



12423 NE Whitaker Way
Portland, OR 97230
503-254-1794

Report Number: 21-003245/D02.R00
Report Date: 04/07/2021
Purchase Order: GSugar21-10
Received: 03/25/21 11:25 AM



Cover Letter



United States of America (USA)

Dear Alison Warmkessel,

Enclosed please find Columbia Laboratories analytical report for samples received as order number 21-003245 on 03/25/2021 at 11:25. Should you have any questions about this report or any other matter, please do not hesitate to contact us. We are here to help you.

Thank you for allowing Columbia Laboratories to be of service to you, we appreciate your business.

Sincerely,

Derrick Tanner
General Manager

Reviewed by  24 May 2021



12423 NE Whitaker Way
Portland, OR 97230
503-254-1794

Report Number: 21-003245/D02.R00
Report Date: 04/07/2021
Purchase Order: GSugar21-10
Received: 03/25/21 11:25 AM



Customer:



United States of America (USA)

Sample ID: 23518 Lot SO15B-3-01:06AM002322082
Sample Matrix: Sugar
Laboratory ID: 21-003245-0001-00
Evidence of Cooling: No
Temp: 18 °C
Relinquished by: UPS

Sample Results

Metals

Smithers Vincent Metals Profile

Analyte	Result	Units	LOQ	Analyzed	Method	Notes
Arsenic	< LOQ	mg/kg	0.00794	04/02/21	AOAC 2013.06 (mod.) ₁	
Cadmium	< LOQ	mg/kg	0.00794	04/02/21	AOAC 2013.06 (mod.) ₁	
Copper	< LOQ	mg/kg	0.0159	04/02/21	AOAC 2013.06 (mod.) ₁	
Lead	< LOQ	mg/kg	0.00794	04/02/21	AOAC 2013.06 (mod.) ₁	
Mercury	< LOQ	mg/kg	0.00397	04/02/21	AOAC 2013.06 (mod.) ₁	
Molybdenum	< LOQ	mg/kg	0.0159	04/02/21	AOAC 2013.06 (mod.) ₁	
Selenium	< LOQ	mg/kg	0.0397	04/06/21	AOAC 2013.06 (mod.) ₁	

1) trace metals in food by Inductively Coupled Mass Spectrometry

Nutrition

Analyte	Result	Units	LOQ	Analyzed	Method	Notes
Acid Insoluble Ash ¹	< LOQ	g/100g	0.10	03/30/21	AOAC 941.12 (mod.)	
pH	3.43	N/A		03/30/21	AOAC 981.12 (mod.)	

Smithers Vincent Metals Profile

Analyte	Result	Units	LOQ	Analyzed	Method	Notes
Aluminum	< LOQ	mg/kg	1.59	04/02/21	AOAC 2011.14 (mod) ₂	
Calcium	30.7	mg/kg	3.97	04/05/21	AOAC 2011.14 (mod) ₂	
Iron	< LOQ	mg/kg	1.59	04/02/21	AOAC 2011.14 (mod) ₂	
Magnesium	< LOQ	mg/kg	3.97	04/05/21	AOAC 2011.14 (mod) ₂	
Manganese	< LOQ	mg/kg	0.794	04/02/21	AOAC 2011.14 (mod) ₂	
Phosphorus	< LOQ	mg/kg	79.4	04/05/21	AOAC 2011.14 (mod) ₂	
Potassium	< LOQ	mg/kg	79.4	04/02/21	AOAC 2011.14 (mod) ₂	
Zinc	< LOQ	mg/kg	0.794	04/02/21	AOAC 2011.14 (mod) ₂	

2) Minerals in food by inductively coupled atomic emission spectroscopy

Pesticides

Multi-Residue Pesticide Profile

Analyte	Result	Units	Analyzed	Method	Notes
---------	--------	-------	----------	--------	-------

www.columbialaboratories.com

Page 2 of 8

Test results relate only to the parameters tested and to the samples as received by the laboratory. Test results meet all requirements of Columbia Laboratories quality assurance plan unless otherwise noted. This report shall not be reproduced, except in full, without the written consent of this laboratory. Samples will be retained for a maximum of 30 days from the receipt date unless prior arrangements have been made.

029



12423 NE Whitaker Way
Portland, OR 97230
503-254-1794

Report Number: 21-003245/D02.R00
Report Date: 04/07/2021
Purchase Order: GSugar21-10
Received: 03/25/21 11:25 AM



Pesticides

Multi-Residue Pesticide Profile

Analyte	Result	Units	Analyzed	Method	Notes
Multi-Residue Pesticide Profile	< LOQ for all analytes	mg/kg	04/02/21	AOAC 2007.01 & EN 15662 (mod)	

Abbreviations

Limit(s) of Quantitation (LOQ): The minimum levels, concentrations, or quantities of a target variable (e.g., target analyte) that can be reported with a specified degree of confidence.

† = Analyte not ISO accredited.

Units of Measure

g/100g = Grams per 100 Grams

mg/kg = Milligram per kilogram = parts per million (ppm)

N/A = Not Applicable

Approved Signatory

Derrick Tanner
General Manager



Columbia Laboratories, Inc
P2220 Multi-Residue Profile, Limits of Quantitation (MDL Sheet)

Compound	LOQ (mg/kg)	Compound	LOQ (mg/kg)	Compound	LOQ (mg/kg)
2,4,5-T	0.010	Butachlor	0.010	Cymoxanil	0.010
2,4,5-TP	0.010	Butralin	0.020	Cypermethrin	0.010
2,4-D	0.010	Butylate	0.010	Cyprodinil	0.010
2,4-DB	0.010	Cadusafos	0.010	Cyromazine	0.010
2,4-DP (Dichlorprop)	0.010	Captafol	0.100	DCPMU	0.010
Abamectin (Avermectin)	0.010	Captan	0.020	DDD, o,p'-	0.010
Acephate	0.020	Carbaryl	0.010	DDD, p,p'-	0.010
Acequinocyl	0.010	Carbendazim	0.010	DDE, o,p'-	0.010
Acetamiprid	0.010	Carbofuran	0.010	DDE, p,p'-	0.010
Acetochlor	0.020	Carbofuran, 3-hydroxy	0.010	DDT, o,p'-	0.010
Acifluorfen	0.010	Carbophenothion	0.010	DDT, p,p'-	0.010
Acrinathrin	0.010	Carbophenothion methyl	0.010	DEF (Tribufos)	0.010
Alachlor	0.020	Carboxin	0.010	Deltamethrin	0.010
Aldicarb	0.010	Carfentrazone-ethyl	0.010	Demeton-S	0.020
Aldicarb sulfone (Aldoxycarb)	0.010	Chlorantraniliprole	0.010	Demeton-S methyl-sulfone	0.020
Aldicarb-sulfoxide	0.010	Chlordane, cis-	0.010	Demeton-s-methyl	0.020
Aldrin	0.010	Chlordane, trans-	0.010	Desmedipham	0.010
Ametoctradin	0.010	Chlordimeform	0.010	Diallate	0.010
Ametryn	0.010	Chlorfenapyr	0.020	Diazinon	0.010
Aminocyclopyrachlor	0.010	Chlorfenson (Ovex)	0.010	Diazoxon	0.010
Anilazine	0.030	Chlorfenvinphos	0.010	Dicamba (Banvel)	0.010
Aspon	0.010	Chlorimuron-ethyl	0.010	Dichlobenil	0.010
Asulam	0.010	Chlormitrofen (CNP)	0.020	Dichlofenthion	0.010
Atrazine	0.010	Chlorobenzilate	0.010	Dichlofluanid	0.010
Atrazine-desethyl	0.010	Chloroneb	0.010	Dichlorobenzamide	0.010
Azinphos-ethyl	0.010	Chlorothalonil	0.040	Dichlorvos	0.010
Azinphos-methyl	0.010	Chlorpropham (CIPC)	0.010	Diclobutrazol	0.010
Azoxystrobin	0.010	Chlorpyrifos (ethyl)	0.010	Diclofop (acid)	0.010
Benalaxyl	0.010	Chlorpyrifos-methyl	0.010	Diclofop-methyl	0.010
Bendiocarb	0.010	Chlorsulfuron	0.010	Dicloran	0.040
Benfluralin	0.010	Chlorthal-dimethyl (Dacthal)	0.010	Dicofol, p,p'-/o,p'-	0.020
Benoxacor	0.010	Chlorthion	0.020	Dicrotophos	0.010
Bensulide	0.010	Chlorthiophos	0.010	Dieldrin	0.010
Bentazon	0.010	Clethodim	0.010	Diethofencarb	0.010
BHC alpha isomer	0.010	Clethodim sulfone	0.010	Diethyltoluamide (DEET)	0.010
BHC beta isomer	0.010	Clethodim sulfoxide	0.010	Difenoconazole	0.010
BHC delta isomer	0.010	Clofentezine	0.010	Diffubenzuron	0.010
Bifenazate	0.010	Clomazone	0.010	Diffufenzopyr	0.010
Bifenox	0.010	Clopyralid	0.010	Dimethenamid	0.010
Bifenthrin	0.010	Clothianidin	0.010	Dimethoate	0.010
Binapacryl	0.040	Coumaphos	0.010	Dimethomorph	0.010
Bitertanol	0.020	Crotoxyphos	0.010	Diniconazole	0.010
Boscalid	0.010	Cyanazine	0.010	Dinocap	0.010
Bromacil	0.020	Cyanofenphos	0.010	Dinoseb (Dinitro)	0.010
Bromophos-methyl	0.010	Cyanophos	0.040	Dinotefuran	0.010
Bromophos-ethyl	0.020	Cyantraniliprole	0.010	Dioxathion	0.010
Bromopropylate	0.010	Cyazofamid	0.010	Diphenamid	0.010
Bromoxynil	0.010	Cycloate	0.010	Diphenylamine (DPA)	0.010
Bromuconazole	0.010	Cycloxydim	0.010	Disulfoton	0.020
Bupirimate	0.010	Cyfluthrin	0.030	Disulfoton sulfone	0.010
Buprofezin	0.010	Cyhalothrin, lambda	0.010	Disulfoton sulfoxide	0.010

LOQ = Limit of Quantitation, mg/kg: If an amount below this level is detected (and the identity confirmed), it may be reported as "Trace".
MDL = Method Detection Limit = LOQ

1 of 3



Columbia Laboratories, Inc
P2220 Multi-Residue Profile, Limits of Quantitation (MDL Sheet)

Compound	LOQ (mg/kg)	Compound	LOQ (mg/kg)	Compound	LOQ (mg/kg)
Dithianon	0.010	Flufenacet	0.010	Isoxaflutole	0.010
Diuron	0.010	Flumioxazin	0.010	Kresoxim-methyl	0.010
DNOC	0.010	Fluometuron	0.010	Lactofen	0.020
Edifenphos	0.010	Fluopicolide	0.010	Lenacil	0.010
Endosulfan (α Isomer)	0.020	Fluopyram	0.010	Lindane	0.010
Endosulfan (β Isomer)	0.020	Fluoxastrobin	0.010	Linuron	0.010
Endosulfan sulfate	0.010	Flupyradifurone	0.010	Malaoxon (Malathion-o-analog)	0.010
Endrin	0.020	Fluridone	0.010	Malathion	0.010
Endrin aldehyde	0.020	Fluroxypyr (free acid)	0.010	Mandipropamid	0.010
EPN	0.010	Flusilazol	0.010	MCPA	0.010
EPTC	0.010	Fluthiacet Methyl	0.010	MCPB	0.010
Esfenvalerate/Fenvalerate	0.020	Flutolanil	0.010	MCPP (Mecoprop)	0.010
Etacozazole	0.010	Flutriafol	0.010	Mecarbam	0.010
Ethalfuralin	0.010	Fluvalinate -tau	0.010	Mepanipyrim	0.010
Ethiofencarb	0.010	Fluxapyroxad	0.010	Mesosulfuron Methyl	0.010
Ethion	0.010	Folpet	0.020	Mesotrione	0.010
Ethirimol	0.010	Fomesafen	0.010	Metalaxyl/Mefenoxam	0.010
Ethofumesate	0.010	Fonofos	0.010	Metconazole	0.010
Ethoprophos	0.010	Foramsulfuron	0.010	Methacrifos	0.010
Ethoxyquin	0.010	Forchlorfenuron	0.010	Methamidophos	0.010
Etofenprox	0.010	Formetanate	0.010	Methidathion	0.010
Etokazole	0.010	Furathiocarb	0.010	Methiocarb	0.010
Etridiazole	0.010	Halosulfuron-methyl	0.010	Methiocarb sulfone	0.010
Etrifos	0.010	Haloxypol (free acid)	0.010	Methiocarb sulfoxide	0.010
Famoxadone	0.020	Heptachlor & Heptachlor epoxide	0.010	Methomyl	0.010
Famphur	0.010	Hexachlorobenzene (HCB)	0.010	Methoxychlor	0.010
Fenamidone	0.010	Hexaconazole	0.010	Methoxyfenozide	0.010
Fenamiphos	0.010	Hexazinone (Velpar)	0.010	Metobromuron	0.010
Fenamiphos Sulfone	0.010	Hexythiazox	0.010	Metolachlor	0.010
Fenamiphos Sulfoxide	0.010	Hydroprene	0.010	Metolcarb	0.010
Fenarimol	0.010	Imazalil	0.010	Metrafenone	0.010
Fenazaquin	0.010	Imazamox	0.010	Metribuzin	0.010
Fenbuconazole	0.010	Imazapic	0.010	Metsulfuron-methyl	0.010
Fenbutatin oxide	0.010	Imazapyr	0.010	Mevinphos	0.010
Fenchlorphos	0.010	Imazaquin	0.010	Mexacarbate	0.010
Fenhexamid	0.010	Imazethapyr	0.010	MGK-264	0.010
Fenitrothion	0.010	Imidacloprid	0.010	Mirex	0.010
Fenobucarb (Baycarb)	0.010	Imidoxone (Phosmet-Oxon)	0.010	Molinate	0.010
Fenoxaprop-P-Ethyl	0.010	Indaziflam	0.010	Monocrotophos	0.010
Fenoxycarb	0.010	Indoxacarb	0.010	Monolinuron	0.010
Fenpropathrin	0.010	Iprobenfos	0.010	Myclobutanil	0.010
Fenpyroximate	0.010	Iprodione	0.020	Naled	0.010
Fenson	0.020	Isazophos	0.010	Napropamide	0.010
Fensulfothion	0.010	Isobenzan	0.010	Neburon	0.010
Fenthion	0.010	Isocarbophos	0.010	Nicosulfuron	0.010
Fenuron	0.010	Isodrin	0.010	Nitrapyrin	0.020
Fipronil	0.010	Isufenphos	0.010	Nitrofen	0.020
Flonicamid	0.010	Isufenphos-methyl/ OA	0.010	Norflurazon	0.010
Fluazifop	0.010	Isoprocarb	0.010	Novaluron	0.010
Fluazinam	0.010	Isopropalin	0.010	Nuarimol	0.020
Fluchloralin	0.010	Isoprotolane	0.010	Omethoate	0.010
Flucythrinate	0.030	Isoproturon	0.010	O-Phenylphenol	0.010
Fludioxonil	0.010	Isoxaben	0.010	Oryzalin	0.010

LOQ = Limit of Quantitation, mg/kg: If an amount below this level is detected (and the identity confirmed), it may be reported as "Trace".
MDL = Method Detection Limit = LOQ



Columbia Laboratories, Inc
P2220 Multi-Residue Profile, Limits of Quantitation (MDL Sheet)

Compound	LOQ (mg/kg)	Compound	LOQ (mg/kg)	Compound	LOQ (mg/kg)
Compound	0.010	Propanil	0.010	Terbutriene	0.010
Oxadiazon	0.010	Propargite	0.010	Terbacil	0.040
Oxadixyl	0.010	Propazine	0.010	Terbufos	0.010
Oxamyl	0.010	Propetamphos	0.010	Terbufos sulfone	0.010
Oxamyl-oxime	0.010	Propham	0.010	Terbufos sulfoxide	0.010
Oxychlorane	0.010	Propiconazole	0.010	Terbuthylazine	0.010
Oxydemeton-Methyl	0.010	Propoxur	0.010	Terbutryn	0.010
Oxyfluorfen	0.010	Propoxycarbazon sodium	0.010	Tertrachlorvinphos	0.010
Oxythioquinox	0.020	Prosulfuron	0.010	Tetraconazole	0.010
Paclobutrazol	0.010	Prothioconazole	0.010	Tetradifon	0.010
Paraoxon-methyl/ethyl	0.010	Prothiofos	0.010	Tetramethrin	0.010
Parathion-ethyl	0.010	Pymetrozine	0.010	Tetrasul	0.010
Parathion-methyl	0.030	Pyraclostrobin	0.010	Thiabendazole	0.010
PCP (Pentachlorophenol)	0.010	Pyraflufen-ethyl	0.010	Thiabendazole, 5-hydroxy	0.010
Penconazole	0.010	Pyrazophos	0.010	Thiacloprid	0.010
Pendimethalin	0.010	Pyrethrins	0.010	Thiamethoxam	0.010
Penflufen	0.010	Pyridaben	0.010	Thifensulfuron-methyl	0.010
Pentachloroaniline (PCA)	0.010	Pyridate	0.010	Thiobencarb (benthocarb)	0.010
Pentachloroanisole	0.010	Pyrimethanil	0.010	Thiodicarb	0.010
Pentachlorobenzene (PCB)	0.010	Pyriproxifen	0.010	Thiometon	0.020
Pentachlorothioanisole (PCTA)	0.030	Pyroxasulfone	0.010	Thionazin	0.010
Penthiopyrad	0.010	Pyroxulam	0.010	Thiophanate-methyl	0.010
Permethrin	0.010	Quinalphos	0.010	Tolclofos-methyl	0.010
Perthane	0.010	Quinclorac	0.010	Tolfenpyrad	0.010
Phenmedipham	0.010	Quinoxifen	0.010	Tolylfluanid	0.010
Phenothrin	0.010	Quintozene (PCNB)	0.010	Topramezone	0.010
Phenthoate	0.010	Quizalofop (free acid)	0.010	Tralkoxydim	0.010
Phorate	0.010	Resmethrin	0.010	Triadimefon	0.010
Phorate OA	0.010	Rimsulfuron	0.010	Triadimenol	0.010
Phorate Sulfone	0.010	Rotenone	0.010	Tri-allate	0.010
Phorate Sulfoxide	0.010	S-421	0.010	Triasulfuron	0.010
Phosalone	0.010	Saflufenacil	0.010	Triazophos	0.010
Phosmet	0.010	Sebuthylazine	0.010	Tribenuron-methyl	0.010
Phosphamidon	0.010	Sethoxydim	0.010	Trichlorfon	0.010
Phoxim	0.010	Simazine	0.010	Triclopyr	0.020
Phthalimide	0.020	Simetryn	0.010	Trifloxystrobin	0.010
Picloram	0.010	Splinetoram	0.010	Trifloxysulfuron -sodium	0.010
Pinoxaden	0.010	Spinosad (α, β Isomers)	0.010	Triflumizole	0.010
Piperonyl Butoxide	0.010	Spirodiclofen	0.010	Trifluralin	0.010
Pirimicarb	0.010	Spiromesifen	0.010	Triflusaluron-methyl	0.010
Pirimiphos-Ethyl	0.010	Spirotetramat	0.010	Triforin	0.010
Pirimiphos-Methyl	0.010	Spirotetramat-enol	0.010	Trinexapac (acid)	0.010
Pirimisulfuron-Methyl	0.010	Spiroxamine	0.010	Trinexapac Ethyl	0.010
Prallethrin	0.010	Sulfallate	0.010	Triticonazole	0.010
Prochloraz	0.010	Sulfentrazone	0.030	Vinclozolin	0.010
Procyimdone	0.010	Sulfometuron-methyl	0.010	Zoxamide	0.010
Prodiamine	0.010	Sulfosulfuron	0.010		
Profenofos	0.010	Sulfotep	0.010		
Profluralin	0.010	Sulfoxalor	0.010		
Promecarb	0.010	Sulprofos	0.010		
Prometon	0.010	Tebuconazole	0.010		
Prometryne	0.010	Tebufenozide	0.010		
Pronamide (Propyzamide)	0.010	Tebuthiuron	0.010		
Propachlor	0.010	Tecnazene	0.010		
Propamocarb	0.010	Tefluthrin	0.010		

mg/kg = Parts per Million (ppm)

LOQ = Limit of Quantitation, mg/kg:

If an amount below this level is detected (and the identity confirmed),
it may be reported as "Trace".

MDL = Method Detection Limit = LOQ

LOQs above are typical of most analyses. Factors affecting the LOQ include
instrumentation sensitivity for a particular analyte, sample size, moisture content
(percent solids) of the sample, effectiveness of the cleanup on the sample extract,
and especially the type of sample matrix.

3 of 3



12423 NE Whitaker Way
Portland, OR 97230
503-254-1794



Report Number: 21-003245/D02.R00
Report Date: 04/07/2021
Purchase Order: GSugar21-10
Received: 03/25/21 11:25 AM



Columbia Laboratories
Sample Receipt Form

Revision: 2.00 Document Control: CF015
Revised: 01/11/2021 Effective: 03/16/2021

Job Number: _____ Search Name: _____

Package/Cooler opened on (if different than received date/time) Date: 3/16/21 Time: 11:25

Received By (Initials): LV Logged in by (Initials): _____ Date: _____ Time: _____

1) Were custody seals on outside of the package/cooler? YES NO NA
If YES, how many and where? _____

Does date match collection date on COC? _____ YES NO NA

2) Was Chain of Custody (COC) included in the package/cooler? YES NO NA

3) Was COC signed when relinquished and received? (time, date)? YES NO NA

4) How was the package/cooler delivered?

UPS FEDEX USPS CLIENT COURIER OTHER: _____

Tracking Number (written in or copy of shipping label): 1Z 6AT 1DU 12 SCH9
6723

5) Was packing material used? YES NO NA

Peanuts Bubble Wrap Foam Paper Other: _____

6) Was temperature upon receipt 4°C+ 2°C (if appropriate)? YES NO NA
If not, client contacted: _____
Proceed? YES NO

7) Was there evidence of cooling? YES NO NA
What kind?

Blue Ice Ice Cooler Packs Dry Ice

8) Were all sample containers sealed in separate plastic bags? YES NO NA

9) Did all sample containers arrive in good condition? YES NO NA

10) Were all sample container labels complete? YES NO NA

11) Did all sample container labels and tags agree with the COC? YES NO NA

12) Were correct sample containers used for the tests indicated? YES NO NA

13) Were VOA vials checked for absence of air bubbles (note if found)? YES NO NA

14) Was a sufficient amount of sample sent in each sample container? YES NO NA

16) Sample location prior to login: R99 R39 R44 F44 Ambient Shelf Cannabis Table Other: _____

Explain any discrepancies: _____

Page ____ of ____

035

Page 73 of 282

MAINTENANCE

Date:	Syrup Lot	2 mL Sugar Syrup Refreshed for all Test Organism Cages:	Initials
07 June 2021 08 June 2021 07	07 June 2021 07 Jun 21A-50 wf 07 Jun 2021	<input checked="" type="checkbox"/>	
08 June 2021	07 Jun 21A-50	<input checked="" type="checkbox"/>	
09 June 2021	07 Jun 21A-50	<input checked="" type="checkbox"/>	
10 June 2021	07 Jun 21A-50	<input checked="" type="checkbox"/>	

Date:	Syrup Lot	2 mL Sugar Syrup Refreshed for all Test Organism Cages:	Initials
13 Sep 2021	13 Sep 21A-S0	<input checked="" type="checkbox"/>	
14 Sep 2021	13 Sep 21A-S0	<input checked="" type="checkbox"/>	
15 Sep 2021	13 Sep 21A-S0	<input checked="" type="checkbox"/>	
16 Sep 2021	13 Sep 21A-S0	<input checked="" type="checkbox"/>	

① 17 Sep 2021

13 Sep 21A-S0

✓

① 18 Sep 2021

13 Sep 21A-S0

✓

① Sugar syrup lot 13 Sep 21A-S0 expiration date of 17 Sep 2021 extended through end of trial 2 exposure (19 Sep 2021). because no signs of mold or spoilage present. 19 Sep 2021

TEST MATERIAL INFORMATION

Test Substance Receipt- non-GMO, non-Cortex [REDACTED] TMC #: 21-310

Test Substance: Dimethoate 400 EC Synonym: NA
Sponsor: NA City/State: NA
Received from: Horizon Company/Hart Agland City/State: Boulder, CO
Container: 2.5 gal jug
Storage Location: Pollinator building enclosed garage storage
Gross Weight (g): 10.8 kg Scale used: E028
Date Received: 04 June 2020 By: [REDACTED]

LABEL INFORMATION ONLY

Test Substance: Dimethoate 400 EC
Lot, Batch, Code, Ref No.: lot 004-17 B463 Net Weight/Amount: 2.5 gallons
Expiration Date: 7/10 Purity: 48.5%
Other: "manufactured" 7/10/17

SPONSOR INFORMATION

By: _____ Date: _____

Source(s): _____
Test Substance: _____
Lot, Batch, Code, Ref. No.: NA [REDACTED] Storage Requirements: _____
CAS #: _____ Purity: _____
Other: _____

Hazard Rating: _____ By: NA (C [REDACTED] rson IHC) Date: 09 Nov 2021
_____ With Conditions _____

* Original maintained in [REDACTED] MC Log, unless otherwise specified.

CRC Form 179

041

Test Substance Receipt – non-GMO, non-Cortex

TMC #: 21-36

PHYSICAL CHARACTERIZATION

Color: amber

Performed By: [REDACTED]

Date: 04 June 2020

Solid: _____

Liquid: ✓

Gas: _____

Powder: _____

Viscous: _____

Crystal: _____

Pellet: _____

Other: _____

SHIPPING INFORMATION

Hazardous: _____

Non-Hazardous: _____

Proper Shipping Name: _____

Classification: _____

UN #: 09 Nov 2021

Packing Group: _____

DISPOSITION OF TEST SUBSTANCE

Final Weight (g): _____ Scale used: _____ Date: _____ By: _____

Returned To (date/init): _____

Disposal Status: _____

Disposal holding bins (date/init): _____

Disposal pick up/Removed for destruction (date/init): _____

Comments: _____

* Original maintained in Smithers CRC TMC Log, unless otherwise specified.

Smithers CRC 179

Test Substance Receipt and Chain of Custody

Courier Name: UPS	Date Delivered: 04 June 2020	Time Delivered: 1300
Tracking #: 12 752 A09 03 9702 8839		
Received by: [REDACTED]	Date Received/Inspected: 04 June 2020	Time received/Inspected: 1330
Condition of outer packaging: good		

TMC Receipt Record

Test Substance: Dimethoate 400 EC		
Assigned Smithers CRC TMC #: 21-36		
Relinquished by (receiver above): [REDACTED]	Date: 04 June 2020	Time: 1332
Received into the TMC by: [REDACTED]	Date: 04 June 2020	Time: 1332
Condition of primary container: good		
Test substance arrived: <input type="checkbox"/> Frozen <input checked="" type="checkbox"/> Room temperature <input type="checkbox"/> Other (please specify):		

Original to be maintained in the [REDACTED] TMC Logbook unless otherwise specified.

CRC Form No. 178

043

21-36

Test Substance Receipt and Chain of Custody Form

Received By: [redacted] Date: 04 June 2020 Time: 1332
(Initial)

Shipped From (company name and address):

Hart Agland
1025 Delaware Ave
Unit C
Longmont CO 80501

Courier: UPS

Tracking #: 1Z 752 A09 03 9702 8839

Conditions of Outer
Packaging:

good

Comments: stored in pollinator building garage

Relinquished by (receiver above): [redacted] Date: 04 June 2020 Time: 1332
(Initial)

Received into TMC by: [redacted] Date: 04 June 2020 Time: 1332
(Initial)

Test Substance	Identification (Lot#, Batch#, Sample #)	Assigned Smithers CRC TMC #	Amount
<u>Dimethoate 400 EC</u>	<u>01707-006</u>		<u>2.5 gal</u>
Total # of Items: <u>1</u>			

I certify that all contents have been reviewed and are present as listed: [redacted] 04 June 2020 1335
(Initial/Date) (Time)

Conditions of Primary Container: good Test substance arrived: ambient
(Frozen, Ambient, etc.)

Comments: NA

Original to be maintained in the [redacted] TMC Logbook unless otherwise specified.

21-36

DIMETHOATE 400 EC

Group **1B** Insecticide

Organophosphate Insecticide
SYSTEMIC INSECTICIDE-MITICIDE

ACTIVE INGREDIENT:

Dimethoate (O,O-dimethyl-S-[(methylcarbamoyl)methyl] phosphorodithioate) 43.5%

OTHER INGREDIENTS: 56.5%**TOTAL:** 100.0%

*This product contains petroleum distillates.
(1 Gallon contains 4.0 pounds of Dimethoate)

KEEP OUT OF REACH OF CHILDREN WARNING/AVISO

Si usted no entiende la etiqueta, busque a alguien para que se la explique a usted en detalle.
(If you do not understand the label, find someone to explain it to you in detail.)

IN CASE OF A MEDICAL EMERGENCY INVOLVING THIS PRODUCT, CALL TOLL FREE,
DAY OR NIGHT 1-800-331-3148

See Inside For Additional Precautionary Statements

DO NOT STORE BELOW 45 °F.

EPA Reg. No. 34704-207-279

EPA Est. No. 5905-GA-1



00035832546778

Sold By

FMC

FMC Corporation
2929 Walnut Street, Philadelphia, PA 19104

NET CONTENTS: 2.5 GALLONS

copy of label from jug/container [redacted] 04 June 2020

045

21-36

SAFETY DATA SHEET

Dimethoate 400 EC

SDS #: FO004182-A
Revision date: 2017-12-20
Format: NA
Version 1.01

**1. PRODUCT AND COMPANY IDENTIFICATION****Product Identifier**

Product Name Dimethoate 400 EC

Other means of identification

Product Code(s) FO004182-A

Synonyms DIMETHOATE: O,O-dimethyl S-[2-(methylamino)-2-oxoethyl] phosphorodithioate (CAS name); 2-dimethoxyphosphinothioylthio-N-methylacetamide (IUPAC name)

Active Ingredient(s) Dimethoate

Chemical Family Organophosphate

Recommended use of the chemical and restrictions on use

Recommended Use: Insecticide

Restrictions on Use: Use as recommended by the label

Supplier Address

FMC Corporation
2929 Walnut Street
Philadelphia, PA 19104
(215) 299-6000 (General Information)
msdsinfo@fmc.com (E-Mail General Information)

Emergency telephone number

For leak, fire, spill or accident emergencies, call:
1 800 / 424 9300 (CHEMTREC - U.S.A.)
1 703 / 741-5970 (CHEMTREC - International)
1 703 / 527 3887 (CHEMTREC - Alternate)

Medical Emergencies:
1 800 / 331-3148 (ProPharma Group - U.S.A. & Canada)
1 651 / 632-6793 (ProPharma Group - All Other Countries - Collect)

2. HAZARDS IDENTIFICATION**Classification****OSHA Regulatory Status**

This material is considered hazardous by the OSHA Hazard Communication Standard (29 CFR 1910.1200)

Acute toxicity - Oral	Category 4
Acute toxicity - Inhalation (Dusts/Mists)	Category 4
Aspiration toxicity	Category 1
Flammable liquids	Category 3

GHS Label elements, including precautionary statements

EMERGENCY OVERVIEW

Danger

Hazard Statements

H302 - Harmful if swallowed
H332 - Harmful if inhaled
H304 - May be fatal if swallowed and enters airways
H401 - Toxic to aquatic life
H411 - Toxic to aquatic life with long lasting effects

Physical Hazards

H226 - Flammable liquid and vapor



Precautionary Statements - Prevention

P264 - Wash hands thoroughly after handling
P270 - Do not eat, drink or smoke when using this product
P261 - Avoid breathing dust/fume/gas/mist/vapors/spray
P271 - Use only outdoors or in a well-ventilated area
P210 - Keep away from heat/sparks/open flames/hot surfaces. No smoking
P233 - Keep container tightly closed
P241 - Use explosion-proof electrical/ventilating/lighting equipment
P242 - Use only non-sparking tools
P243 - Take precautionary measures against static discharge
P280 - Wear protective gloves/protective clothing/eye protection/face protection
P273 - Avoid release to the environment

Precautionary Statements - Response

P303 + P361 + P353 - IF ON SKIN (or hair): Take off immediately all contaminated clothing. Rinse skin with water/shower
P312 - Call a POISON CENTER or doctor if you feel unwell
P304 + P340 - IF INHALED: Remove person to fresh air and keep comfortable for breathing
P301 + P310 - IF SWALLOWED: Immediately call a POISON CENTER or doctor
P330 - Rinse mouth
P391 - Collect spillage

Precautionary Statements - Storage

P405 - Store locked up
P403 + P233 - Store in a well-ventilated place. Keep container tightly closed

Precautionary Statements - Disposal

P501 - Dispose of contents/container according to label directions

Hazards not otherwise classified (HNOC)

No hazards not otherwise classified were identified.

Other Information

Harmful to aquatic life with long lasting effects. Toxic to aquatic life.

3. COMPOSITION/INFORMATION ON INGREDIENTS

Chemical Family

Organophosphate.

Chemical name	CAS-No	Weight %
Dimethoate	60-51-5	43.5
Cyclohexanone	108-94-1	30-40
Naphtha (petroleum), heavy aromatic	64742-94-5	5-15
Xylenes	1330-20-7	1-5
Trimethylbenzene	25551-13-7	1-5

Synonyms are provided in Section 1.

4. FIRST AID MEASURES

Eye Contact	Hold eyes open and rinse slowly and gently with water for 15 to 20 minutes. Remove contact lenses, if present, after the first 5 minutes, then continue rinsing eye. Call a poison control center or doctor for further treatment advice.
Skin Contact	Take off contaminated clothing. Rinse skin immediately with plenty of water for 15-20 minutes. Call a poison control center or doctor for further treatment advice. Wash contaminated clothing before reuse.
Inhalation	Move to fresh air. If person is not breathing, call 911 or an ambulance, then give artificial respiration, preferably mouth-to-mouth if possible. Call a poison control center or doctor for further treatment advice.
Ingestion	Induce vomiting ONLY under the direct supervision of qualified medical personnel or a poison control center. Never give anything by mouth to an unconscious person. Immediate medical attention is required.
Most important symptoms and effects, both acute and delayed	<p>Symptoms of poisoning may include headache, nausea, vomiting, blurred vision, tightness in chest, drooling, frothing of mouth and nose, convulsions, coma and death. This product may present an aspiration hazard. Aspiration into the lungs during swallowing or subsequent vomiting may cause chemical pneumonitis, which can be fatal.</p> <p>Prolonged or repeated overexposure may cause behavioral changes. Prolonged or repeated skin exposure may cause redness, a burning sensation, drying and cracking of the skin (dermatitis). Prolonged or repeated overexposure may cause liver, kidney and blood system effects.</p>
Indication of immediate medical attention and special treatment needed, if necessary	<p>This product contains a cholinesterase inhibitor affecting the central and peripheral nervous systems and producing respiratory depression. Decontamination procedures such as whole body washing, gastric lavage and administration of activated charcoal are often required. If symptoms are present, administer atropine sulphate in large doses. Two to four mg intravenously or intramuscularly, as soon as possible. Repeat at 5 to 10 minute intervals until signs of atropinization appear. Maintain full atropinization until all organophosphate is metabolized. Obidoxime chloride (Toxogonin), alternatively pralidoxime chloride (2-PAM), may be administered as an adjunct to, but not a substitute for atropine, which is a symptomatic and often life-saving antidote. Treatment with oxime should be maintained as long as atropine sulphate is administered. At first sign of pulmonary edema, the patient should be given supplemental oxygen and treated symptomatically. Continued absorption may occur and relapse may occur after initial improvement. VERY CLOSE SUPERVISION OF THE PATIENT IS INDICATED FOR AT LEAST 48 HOURS, DEPENDING ON THE SEVERITY OF POISONING.</p>

5. FIRE-FIGHTING MEASURES

Suitable Extinguishing Media	Dry chemical, carbon dioxide, water spray or regular foam. Avoid heavy hose streams.
Specific Hazards Arising from the Chemical	Flammable liquid and vapor. This material will ignite when exposed to heat, sparks, flames, or other sources of ignition (e.g. static electricity, pilot lights, or mechanical/electrical equipment). Material may decompose rapidly when exposed to heat and flame. Heat of decomposition may cause closed containers to build up pressure and explode.

Hazardous Combustion Products Carbon oxides (COx), nitrogen oxides (NOx), Phosphorus oxides, sulfur oxides.**Explosion data****Sensitivity to Mechanical Impact**

Not sensitive.

Sensitivity to Static Discharge

Yes, May be ignited by friction, heat, sparks or flames.

Protective equipment and precautions for firefighters

As in any fire, wear self-contained breathing apparatus and full protective gear. Move containers from fire area if you can do it without risk. Use water spray to cool fire exposed surfaces and protect personnel. Approach fire from upwind to avoid hazardous vapours and toxic decomposition products.

6. ACCIDENTAL RELEASE MEASURES**Personal Precautions**

In case of spill, avoid contact. Isolate area and keep out animals and unprotected persons. Isolate and post spill area. Ensure clean-up is conducted by trained personnel only. Remove all sources of ignition. Wear suitable protective clothing, gloves and eye/face protection. Always wear a self-contained breathing apparatus or full-face airline respirator when using this chemical. For personal protection see section 8.

Other

For further clean-up instructions, call FMC Emergency Hotline number listed in Section 1 "Product and Company Identification" above.

Environmental Precautions

Prevent entry into waterways, sewers, basements or confined areas. Keep people and animals away from and upwind of spill/leak. Keep material out of lakes, streams, ponds, and sewer drains.

Methods for Containment

Remove all sources of ignition. Ventilate area of release. Stop the spill at source if it is safe to do so. Contain and absorb spilled material with inert, non-combustible absorbent material, such as sand. Sweep up and shovel into suitable containers for disposal. For a water spill, confine the spill immediately with booms. Large spills that soak into the ground should be dug up, placed into suitable containers and disposed of appropriately (see Section 13). Notify the appropriate authorities as required.

Methods for cleaning up

Pick up and transfer to properly labeled containers.

7. HANDLING AND STORAGE**Handling**

This material is a toxic liquid. Wear chemically resistant protective equipment during handling. Use only in well-ventilated areas. Avoid contact with eyes, skin and clothing. Do not breathe vapors or spray mist. Keep away from children and all unprotected persons. Do not use near sources of heat, flame or direct sunlight. Dimethoate should never be heated above 35°C. Heat only indirectly and with solvent present. Local heating with, for example, electric heating equipment or steam, may significantly increase the risk of explosion and should never take place. Keep away from incompatibles. Use caution when opening cap. Keep containers tightly closed when not in use. Wash thoroughly after handling.

Storage

Store in a well-ventilated place. Keep cool. Keep away from heat and sources of ignition i.e., steam pipes, radiant heaters, hot air vents or welding sparks. Avoid storage above 77°F / 25°C for prolonged period of time. Keep away from incompatible materials. Storage area should be clearly identified, clear of obstruction and accessible only to trained and authorized personnel. Containers should be visually inspected on a regular basis to detect any abnormalities (swollen drums, increases in temperature, etc.).

Incompatible products

Strong oxidizing agents, Strong acids, strong bases.

8. EXPOSURE CONTROLS/PERSONAL PROTECTION**Control parameters**

Chemical name	ACGIH TLV	OSHA PEL	NIOSH	Mexico
Cyclohexanone	STEL 50 ppm	TWA: 50 ppm	IDLH: 700 ppm	Mexico: TWA 50 ppm

(108-94-1)	TWA: 20 ppm	TWA: 200 mg/m ³	TWA: 25 ppm TWA: 100 mg/m ³	Mexico: TWA 200 mg/m ³ Mexico: STEL 100 ppm Mexico: STEL 400 mg/m ³
Xylenes (1330-20-7)	STEL 150 ppm TWA: 100 ppm	TWA: 100 ppm TWA: 435 mg/m ³	-	Mexico: TWA 100 ppm Mexico: TWA 435 mg/m ³ Mexico: STEL 150 ppm Mexico: STEL 655 mg/m ³
Trimethylbenzene (25551-13-7)	TWA: 25 ppm	-	-	Mexico: TWA 25 ppm Mexico: TWA 125 mg/m ³ Mexico: STEL 35 ppm Mexico: STEL 170 mg/m ³
Chemical name	British Columbia	Quebec	Ontario TWA EV	Alberta
Cyclohexanone (108-94-1)	TWA: 20 ppm STEL: 50 ppm Skin	TWA: 25 ppm TWA: 100 mg/m ³ Skin	TWA: 20 ppm STEL: 50 ppm Skin	TWA: 20 ppm TWA: 80 mg/m ³ STEL: 50 ppm STEL: 200 mg/m ³ Skin
Xylenes (1330-20-7)	TWA: 100 ppm STEL: 150 ppm	TWA: 100 ppm TWA: 434 mg/m ³ STEL: 150 ppm STEL: 651 mg/m ³	TWA: 100 ppm STEL: 150 ppm	TWA: 100 ppm TWA: 434 mg/m ³ STEL: 150 ppm STEL: 651 mg/m ³
Trimethylbenzene (25551-13-7)	TWA: 25 ppm	TWA: 25 ppm TWA: 123 mg/m ³	TWA: 25 ppm	TWA: 25 ppm TWA: 123 mg/m ³

Appropriate engineering controls**Engineering measures**

Apply technical measures to comply with the occupational exposure limits (if listed above). When working in confined spaces (tanks, containers, etc.), make sure there is an adequate source of air for breathing and wear the recommended equipment. Ventilate all transport vehicles prior to discharge.

Individual protection measures, such as personal protective equipment**Eye/Face Protection**

Chemical resistant goggles must be worn. Maintain eye wash fountain and quick-drench facilities in work area.

Skin and Body Protection

Wear impervious protective clothing, including boots, gloves, lab coat, apron or coveralls, as appropriate, to prevent skin contact.

Hand Protection

Impervious gloves. Wear long chemical resistant gloves, such as barrier laminate, butyl rubber or nitrile rubber. The breakthrough times of these materials for the product are unknown. Generally, however, the use of protective gloves will give only partial protection against dermal exposure. Small tears in the gloves and cross-contamination can easily occur. It is recommended to limit the work to be done manually and to change the gloves frequently. Be careful not to touch anything with contaminated gloves. Used gloves should be thrown out and not be reused.

Respiratory Protection

For splash, spray or mist exposure wear, as a minimum, a properly fitted half-face or full-face respirator with dust/mists/fume cartridges (approved by U.S. NIOSH/MSHA, EU CEN or comparably certified organization). Respirator use and selection must be based on airborne concentrations.

Hygiene measures

Avoid breathing vapors, mist or gas. Avoid contact with skin, eyes and clothing. Do not eat, drink or smoke when using this product. Remove and wash contaminated clothing and gloves, including the inside, before re-use. Wash hands and face before breaks and immediately after handling the product. Remove and wash contaminated clothing before re-use. Persons working with this product for a longer period should have frequent blood tests for cholinesterase levels. If the cholinesterase levels fall below a critical point, no further exposure should be allowed until it has been determined, by means of blood tests, that cholinesterase levels have returned to normal.

General information

If the product is used in mixtures, it is recommended that you contact the appropriate protective equipment suppliers.

9. PHYSICAL AND CHEMICAL PROPERTIES**Information on basic physical and chemical properties**

Appearance	Yellow liquid
Physical State	Liquid
Color	Colorless, Light yellow
Odor	Slight mercaptan
Odor threshold	No information available
pH	3.59 (5% solution)
Melting point/freezing point	< 5 °C / 41 °F
Boiling Point/Range	Decomposes at temperatures above >80°C.
Flash point	42 °C / 108 °F
Evaporation Rate	No information available
Flammability (solid, gas)	No information available
Flammability Limit in Air	
Upper flammability limit:	1.3-1.9
Lower flammability limit:	9.4-12.6
Vapor pressure	1.85 x 10 ⁻⁶ mmHg@25°C (Dimethoate)
Vapor density	No information available
Density	No information available
Specific gravity	1.09 - 1.11 @25°C
Water solubility	Emulsifies
Solubility in other solvents	No information available
Partition coefficient	Log Kow = 0.704 (Dimethoate)
Autoignition temperature	No information available
Decomposition temperature	176°F (80°C)
Viscosity, kinematic	No information available
Viscosity, dynamic	No information available
Explosive properties	Combustible liquid
Oxidizing properties	No information available
Molecular weight	No information available
Bulk density	8.94-9.10 lb/gal

10. STABILITY AND REACTIVITY

Reactivity	It is strongly advised not to heat this product above 95°F (35°C) and only heat indirectly with solvent present. Above 176°F (80°C) the product will decompose rapidly, significantly increasing the risk of inducing explosions. The released heat from decomposition can raise the temperature further and accelerate decomposition.
Chemical Stability	<p>Dimethoate is stable for a long period at temperatures not exceeding 25°C. At higher temperatures decomposition will take place and lower the quality of the product.</p> <p>The decomposition is dependent on time as well as temperature due to self-accelerating exothermic and autocatalytic reactions. The reactions involve rearrangements and polymerisation.</p> <p>At higher temperatures the released heat can raise the temperature further and accelerate the decomposition.</p> <p>Tests have shown that, if dimethoate is heated to and kept at 40°C for 2 weeks, the content of active ingredient will be lowered by 6% or more and after 20 weeks at 40°C the content of active ingredients halved.</p>
Possibility of Hazardous Reactions	None under normal processing.
Hazardous polymerization	Hazardous polymerization may occur. See "Chemical Stability" above.
Conditions to avoid	Heat (temperatures above flash point), sparks, ignition points, flames, static electricity. Keep away from open flames, hot surfaces and sources of ignition.
Incompatible materials	Strong oxidizing agents, Strong acids, strong bases.

Hazardous Decomposition Products Carbon oxides (COx), sulfur oxides, Phosphorous oxides, nitrogen oxides (NOx).

11. TOXICOLOGICAL INFORMATION

The below results are based on testing performed on representative samples of a mixture similar to this product.

Product Information

LD50 Oral 450 mg/kg (rat)
 LD50 Dermal > 2000 mg/kg (rat)
 LC50 Inhalation 2.5 mg/L (4-hr) (rat)

Serious eye damage/eye irritation Irritating to eyes.
 Skin corrosion/irritation Irritating to skin.
 Sensitization Non-sensitizer

Information on toxicological effects

Symptoms No information available.

Delayed and immediate effects as well as chronic effects from short and long-term exposure

Mutagenicity No known mutagenic or teratogenic effects.
 Carcinogenicity May cause cancer
 Reproductive toxicity Not expected to have reproductive effects.
 Teratogenicity Not expected to be a teratogen.
 STOT - single exposure No information available.
 STOT - repeated exposure No information available.
 Aspiration hazard This product presents an aspiration pneumonia hazard.

Chemical name	ACGIH	IARC	NTP	OSHA
Dimethoate 60-51-5		Group 2A		
Cyclohexanone 108-94-1	A3	Group 3		
Xylenes 1330-20-7		Group 3		

Legend:

ACGIH (American Conference of Governmental Industrial Hygienists)
 A3 - Animal Carcinogen
 IARC (International Agency for Research on Cancer)
 Group 2A - Probably Carcinogenic to Humans
 Group 3 - Not classifiable as to its carcinogenicity to humans

12. ECOLOGICAL INFORMATION

Ecotoxicity

Persistence and degradability Not readily biodegradable.
 Bioaccumulation Not expected to bioaccumulate.
 Mobility Moderately mobile, Absorption depends on soil pH and organic matter content.

13. DISPOSAL CONSIDERATIONS

Waste disposal methods Improper disposal of excess pesticide, spray mixture, or rinsate is prohibited. If these wastes cannot be disposed of by use according to label instructions, contact appropriate

disposal authorities for guidance. Proper personal protective equipment, as described in Sections 7 and 8, must be worn while handling materials for waste disposal.

Contaminated Packaging

Dispose of rinse water in accordance with local and national guidelines. Containers must be disposed of in accordance with local, state and federal regulations. Refer to the product label for container disposal instructions.

14. TRANSPORT INFORMATION

DOT

UN/ID no	NA1993
Proper Shipping Name	Combustible liquid, n.o.s
Hazard class	Combustible liquid
Packing Group	III
Reportable Quantity (RQ)	RQ
Description	NA1993, Combustible liquid, n.o.s. (Cyclohexanone, Aromatic hydrocarbons, Dimethoate), III, RQ

TDG

UN/ID no	NA1993
Proper Shipping Name	Combustible liquid, n.o.s
Hazard class	Combustible liquid
Packing Group	III
Description	NA1993, Combustible liquid, n.o.s. (Cyclohexanone, Aromatic hydrocarbons, Dimethoate), III, RQ

15. REGULATORY INFORMATION

U.S. Federal Regulations**SARA 313**

Section 313 of Title III of the Superfund Amendments and Reauthorization Act of 1986 (SARA). This product contains a chemical or chemicals which are subject to the reporting requirements of the Act and Title 40 of the Code of Federal Regulations, Part 372:

Chemical name	CAS-No	Weight %	SARA 313 - Threshold Values %
Dimethoate - 60-51-5	60-51-5	43.5	1.0
Xylenes - 1330-20-7	1330-20-7	1-5	1.0

SARA 311/312 Hazard Categories

Acute health hazard	Yes
Chronic health hazard	Yes
Fire hazard	Yes
Sudden release of pressure hazard	No
Reactive Hazard	No

Clean Water Act

This product contains the following substances which are regulated pollutants pursuant to the Clean Water Act (40 CFR 122.21 and 40 CFR 122.42):

Chemical name	CWA - Reportable Quantities	CWA - Toxic Pollutants	CWA - Priority Pollutants	CWA - Hazardous Substances
Xylenes 1330-20-7	100 lb			X

CERCLA

This material, as supplied, contains one or more substances regulated as a hazardous substance under the Comprehensive Environmental Response Compensation and Liability Act (CERCLA) (40 CFR 302):

Dimethoate 400 EC

SDS #: FO004182-A
Revision date: 2017-12-20
Version 1.01

Chemical name	Hazardous Substances RQs	Extremely Hazardous Substances RQs
Cyclohexanone 108-94-1	5000 lb 2270 kg	
Xylenes 1330-20-7	100 lb 45.4 kg	
Dimethoate 60-51-5	10 lb 4.54 kg	10 lb
Cumene 98-82-8	5000 lb 2270 kg	

FIFRA Information

This chemical is a pesticide product registered by the Environmental Protection Agency and is subject to certain labeling requirements under federal pesticide law. These requirements differ from the classification criteria and hazard information required for safety data sheets, and for workplace labels of non-pesticide chemicals. Following is the hazard information as required on the pesticide label:

WARNING

May be fatal if swallowed.

Causes substantial but temporary eye injury. Do not get on eyes or on clothing.

Harmful if absorbed through skin. Avoid contact with skin.

US State Regulations

California Proposition 65

This product does not contain any Proposition 65 chemicals.

U.S. State Right-to-Know Regulations

Chemical name	New Jersey	Massachusetts	Pennsylvania
Dimethoate 60-51-5	X	X	X
Cyclohexanone 108-94-1	X	X	X
Xylenes 1330-20-7	X	X	X
Trimethylbenzene 25551-13-7	X	X	X

International Inventories

Chemical name	TSCA (United States)	DSL (Canada)	EINECS/ELINCS (Europe)	ENCS (Japan)	China (IECSC)	KECL (Korea)	PICCS (Philippines)	AICS (Australia)
Dimethoate 60-51-5	X	X	X	X	X	X	X	X
Cyclohexanone 108-94-1	X	X	X	X	X	X	X	X
Naphtha (petroleum), heavy aromatic 64742-94-5	X	X	X		X	X	X	X
Xylenes 1330-20-7	X	X	X	X	X	X	X	X
Trimethylbenzene 25551-13-7	X	X	X	X	X	X	X	X

Chemical name	Carcinogen Status	Mexico
Cyclohexanone		Mexico: TWA 50 ppm

		Mexico: TWA 200 mg/m ³ Mexico: STEL 100 ppm Mexico: STEL 400 mg/m ³
Xylenes		Mexico: TWA 100 ppm Mexico: TWA 435 mg/m ³ Mexico: STEL 150 ppm Mexico: STEL 655 mg/m ³
Trimethylbenzene		Mexico: TWA 25 ppm Mexico: TWA 125 mg/m ³ Mexico: STEL 35 ppm Mexico: STEL 170 mg/m ³

Chemical name	Mexico - Pollutant Release and Transfer Register - Reporting Emissions for Fabrication, Process or Use -Threshold Quantities	Pollutant Release and Transfer Register - Reporting Emissions - Threshold Quantities
Xylenes	1000 5000 kg/yr	1000 kg/yr
Cumene	1000 5000 kg/yr	1000 kg/yr

CANADA

WHMIS Statement

This product has been classified in accordance with the Hazardous Products Regulations (HPR) and the SDS contains all the information required by the HPR.

WHMIS Hazard Class

D1A - Very toxic materials



16. OTHER INFORMATION

NFPA	Health Hazards 2	Flammability 2	Instability 2	Special Hazards -
HMIS	Health Hazards 2*	Flammability 2	Physical hazard 2	Personal Protection X

*Indicates a chronic health hazard.

NFPA/HMIS Ratings Legend

Severe = 4; Serious = 3; Moderate = 2; Slight = 1; Minimal = 0

Revision date:

2017-12-20

Reason for revision:

(M)SDS sections updated

Disclaimer

FMC Corporation believes that the information and recommendations contained herein (including data and statements) are accurate as of the date hereof. NO WARRANTY OF FITNESS FOR ANY PARTICULAR PURPOSE, WARRANTY OF MERCHANTABILITY OR ANY OTHER WARRANTY, EXPRESSED OR IMPLIED, IS MADE CONCERNING THE INFORMATION PROVIDED HEREIN. The information provided herein relates only to the specified product designated and may not be applicable where such product is used in combination with any other materials or in any process. Use of this product is regulated by the U.S. Environmental Protection Agency (EPA). It is a violation of Federal law to use this product in a manner inconsistent with its labeling. Further, since the conditions and methods of use are beyond the control of FMC Corporation, FMC corporation expressly disclaims any and all liability as to any results obtained or arising from any use of the products or reliance on such information.

Prepared By:

FMC Corporation

FMC Logo - Trademark of FMC Corporation

© 2017 FMC Corporation. All Rights Reserved.

Page 10 / 11

055

Dimethoate 400 EC

SDS # : FO004182-A
Revision date: 2017-12-20
Version 1.01

End of Safety Data Sheet

CAGE AND PLOT ASSIGNMENT

Randomization of Test Cages**Procedure:**

Cages are listed in order of time interval/treatment/rep. Each is assigned a random #.

The resulting random function will be pasted as a value to "hold" the value.

Cages will be sorted by random #, and each cage will be assigned a sequential number which will be the order in which bees are added to the cages. Bees will be indiscriminately selected from brood frames and placed into the cages (25 per cage).

Each day will have its own randomization.

The resulting randomized list is presented below.

This randomization will be used for both Smithers cages and Eurofins cages.

Day 0 treatment replicates

Time/TRT/Rep	Random No.	Cage order
6hr, Control, R5	0.057080708	1
6hr, TRT, R1	0.178158334	2
6hr, Control, R2	0.205326899	3
6hr, TRT, R2	0.297875144	4
6hr, TRT, R3	0.313983752	5
6hr, Control, R4	0.406918427	6
6hr, TRT, R5	0.576431535	7
6hr, Control, R3	0.594235462	8
6hr, TRT, R6	0.807859253	9
6hr, Control, R1	0.859024425	10
6hr, Control, R6	0.941946338	11
6hr, TRT, R4	0.969900676	12

Day 1 treatment replicates

TRT/Rep	Random No.	Cage order
24hr, Control, R3	0.093116439	13
24hr, Control, R4	0.236332674	14
24hr, Control, R1	0.246959721	15
24hr, Control, R2	0.387191858	16
24hr, TRT, R5	0.465418779	17
24hr, TRT, R1	0.474746284	18
24hr, Control, R6	0.568948965	19
24hr, TRT, R3	0.582232819	20
24hr, TRT, R4	0.73572655	21
24hr, TRT, R2	0.775510202	22
24hr, Control, R5	0.909050916	23
24hr, TRT, R6	0.989287205	24

Day 2 treatment replicates

TRT/Rep	Random No.	Cage order
48hr, Control, R6	0.094439075	25
48hr, Control, R3	0.193237875	26
48hr, Control, R2	0.319242397	27
48hr, TRT, R1	0.450281517	28
48hr, TRT, R5	0.477284463	29
48hr, TRT, R6	0.502733248	30
48hr, Control, R1	0.54325091	31
48hr, TRT, R3	0.545349839	32
48hr, TRT, R2	0.618490286	33
48hr, Control, R4	0.705644449	34
48hr, Control, R5	0.852452991	35
48hr, TRT, R4	0.990684066	36

Created in excel by:

[REDACTED] 04 June 2021

Randomization carried out by:

[REDACTED] 07 June 2021

Randomization of Plots

Procedure:

Plots are listed in order. Each is assigned a random #.

The resulting random function will be pasted as a value to "hold" the value. Plot will be sorted by random #, and each plot will be assigned to a time interval.

Treatment intervals require 3 plots, Control plots only require one plot.

The resulting randomized list is presented below.

Treatment Plot Assignment

Plot	Random No.	Time Interval
8	0.130168329	0hr
14	0.224145496	0hr
9	0.322380093	0hr
6	0.484306059	6hr
15	0.489479361	6hr
12	0.56826095	6hr
13	0.588578762	24hr
5	0.606725131	24hr
7	0.6821645	24hr
11	0.765415285	48hr
1	0.853763496	48hr
2	0.855613038	48hr
3	0.885523395	72hr
4	0.966297995	72hr
10	0.990038493	72hr

Control Plot Assignment

Plot	Random No.	Time Interval
2	0.344439169	0hr
4	0.519767978	6hr
3	0.595578381	24hr
5	0.693999057	48hr
1	0.948406151	72hr

Created in excel by [REDACTED] 04 June 2021

Plots marked out by: [REDACTED] 07 June 2021

Randomization of Plots

Trial 2 (MF) 13 Sep 2021

Procedure:

Plots are listed in order. Each is assigned a random #.

The resulting random function will be pasted as a value to "hold" the value. Plot will be sorted by random #, and each plot will be assigned to a time interval.

Treatment intervals require 3 plots, Control plots only require one plot.

The resulting randomized list is presented below.

Treatment Plot Assignment

Plot	Random No.	Time Interval
15	0.088393796	0hr
3	0.211282947	0hr
5	0.356923059	0hr
1	0.374241128	6hr
13	0.381898255	6hr
2	0.407668433	6hr
12	0.41222759	24hr
6	0.444087663	24hr
7	0.536827519	24hr
11	0.594245071	48hr
4	0.685486177	48hr
8	0.694748982	48hr
10	0.743786365	72hr
14	0.967299994	72hr
9	0.995698046	72hr

Control Plot Assignment

Plot	Random No.	Time Interval
3	0.208631636	0hr
1	0.41210003	6hr
4	0.698461145	24hr
2	0.720722341	48hr
5	0.898499391	72hr

Created in excel by: 13 Sep 2021

Plots marked out by:

Randomization of Test Cages**Procedure:**

Cages are listed in order of time interval/treatment/rep. Each is assigned a random #.

The resulting random function will be pasted as a value to "hold" the value.

Cages will be sorted by random #, and each cage will be assigned a sequential number which will be the order in which bees are added to the cages. Bees will be indiscriminately selected from brood frames and placed into the cages (25 per cage).

Each day will have its own randomization.

The resulting randomized list is presented below.

This randomization will be used for both Smithers cages and Eurofins cages.

Day 0 treatment replicates

Time/TRT/Rep	Random No.	Cage order
6hr, TRT, R6	0.035377047	1
6hr, TRT, R4	0.0783551	2
6hr, TRT, R2	0.093494225	3
6hr, Control, R1	0.31714218	4
6hr, TRT, R5	0.408344718	5
6hr, Control, R4	0.415131504	6
6hr, Control, R5	0.460001449	7
6hr, TRT, R3	0.651522696	8
6hr, Control, R3	0.654915553	9
6hr, Control, R6	0.74333583	10
6hr, TRT, R1	0.91673266	11
6hr, Control, R2	0.934413965	12

Day 1 treatment replicates

TRT/Rep	Random No.	Cage order
24hr, TRT, R2	0.075940298	13
24hr, TRT, R5	0.125895439	14
24hr, Control, R3	0.197519561	15
24hr, TRT, R3	0.21635416	16
24hr, Control, R4	0.377612763	17
24hr, Control, R1	0.417760129	18
24hr, Control, R2	0.488331445	19
24hr, TRT, R4	0.489359338	20
24hr, TRT, R1	0.655359811	21
24hr, Control, R6	0.704428751	22
24hr, Control, R5	0.709318038	23
24hr, TRT, R6	0.802188941	24

Created in excel by: [REDACTED]

13 Sep 2021

Randomization carried out by: _____

062

APPLICATION 2021 TRIAL 1

Study No.: PRTF RT25 Ring Test 2021

Date: 08 June 2021

Application Event: 1

Sprayer ID: SPR 07

Calibration Information

Nozzle output per 15 seconds

Output measured in (mL, L, gal, etc.): mL

Sprayer Pressure: LV 3 of 5

Sprayer Swath: 55 inches

Output (mL)				Total	Avg
	Run 1	Run 2	Run 3		
Time	15.40	15.03	15.22	45.65	15.22 seconds
Nozzle 1 (mL)	78	81	83	242	80.67 mL
Nozzle 2 (mL)	81	82	82	245	81.67 mL
Output (mL)/sec	10.32	10.84	10.84	Average mL/sec: 10.67	
Variance 95%	10.14				
Variance 105%	70.16 ^{WE} 11.20				

Date: 08 June 2021

Initials: [REDACTED]

Timer: TIM 110

064

Study No.: PRTF RT25 Ring test 2021

Date: 08 June 2021

Application Event: 1

Sprayer Description

Sprayer ID: SPR07

Description: backpack sprayer with internal circulation, modified with 2 nozzles

PSI: LV 30PS

Number of Nozzles: 2

Nozzle screen: Red screen, nozzles: yellow Teejet "turbo" 02 fan

Swath width: 55 inches

Nozzle distance to target: ~ 12 inches

Agitation type during mixing: shaken, poured into tank, circulation turned on

Sprayer Diagram or Photo:



Initial/Date: 08 June 2021

Application Environmental Conditions

Parameter	Reading	Instrument/Monitor
Temperature (°C)	23.8 (1)	Ambient WS-2902A (1)
Humidity	92% (1)	Ambient WS-2902A (1)
Wind Speed	2.0	Ambient WS-2902A (1)
Wind Direction	north east 08 JUN 21 (2)	NA (1)
Soil Conditions	Dry / Moist / Wet (1)	N/A
Crop Height	15-50 cm (1)	N/A
% Crop Coverage in Plot	90% (1)	N/A
Crop Type	Alfalfa	N/A
Species Name	medicago sativa	N/A
Distance between Control and Treatment Plots	85ft (1)	N/A

(1) 08 Jun 2021

Control Application Calculations

Application variables		
Description	Value	Units
Application rate	0.5	lbs a.i./acre
Plot size	80	sq ft.
Sprayer rate	10.67	mL/sec
spray coverage	80.93725339	L/acre

Conversions	
Sq ft per acre	43560
mL per L	1000
g per lb	453.592

Plot size (acres)	Total spray solution for plot (L)	Total spray solution for plot (mL)	Total spray time (sec)
0.001836547	0.148645093	148.6450935	13.93112404

Total application solution	
Overage	16.81858406
Total mL water	2500

067

Study No.: PRTF RT25 Ring Test 2021Initial/Date: (CD) 08 JUN 2021Application Event: 1**Application Information**Amount of Water: 2500 mLTime of Mixing: 0953Container No.: N/AAmount used: N/A

Application Pass Times (sec):	
Plot	Time (sec)
Control 1	<u>18</u>
Control 2	<u>16</u>

Used Time 16 08 JUN 21 (CD)

Application Start Time: <u>1002</u>
Application End Time: <u>1002</u>
Estimated Volume Remaining: <u>1750 mL</u>
Disposal Method: <u>Fallow field</u>
Total Pass Time: <u>18 sec</u> ^{WB. 08 JUN 21 (CD)} <u>34 sec</u>
Applied By: <u>[REDACTED] 08 June 2021</u>

Application Rate Verification Calculation:**Expected Passtime**
(Based on
$$\text{Time per pass} \times \text{No. of passes} = \text{Expected Total Passtime}$$
Actual Passtime:

$$\text{Total Pass time} \div \text{Expected Pass time} = \text{Percent Application Rate}$$
Actual a.i. output:

$$\text{Target FP application} \times \text{Percent Application Rate} = \text{Total FP Applied}$$

Treatment Application Calculations

Application variables		
Description	Value	Units
Application rate	0.5	lbs a.i./acre
Plot size	240	sq ft.
Sprayer rate	10.67	mL/sec
spray coverage	80.93725339	L/acre
Density	1.1	g/mL
Purity	0.47415	g a.i./mL
Purity	43.5	%

Conversions	
Sq ft per acre	43560
mL per L	1000
g per lb	453.592

690

Plot size (acres)	Total a.i. needed (lbs)	Total a.i. needed (g)	Total test material needed for plot (mL)	Total spray solution for plot (L)	Total spray solution for plot (mL)	Total water (mL)	Total spray time (sec)
0.005509642	0.002754821	1.249564738	2.635378547	0.44593528	445.9352804	443.2999018	41.79337211

Total application solution	
Overage	5.691781932
Total mL test material	15.00
Total mL water	2523.1664

Spray time per treatment plot:

Plots 1-9:	25.08	Sec
Plots 10-15:	16.72	Sec

Study No.: PRTF RT25 Ring Test 2021Initial/Date: CO 08 JUN 21Application Event: 1**Application Information**Amount of Water: 2523.17 mLTime of Mixing: 1012Container No.: 21-34Amount used: 15 mL

Application Pass Times (sec):	
Plot	Time (sec)
TRT1	<u>26</u>
TRT2	<u>16</u>
Total time:	<u>42</u>

Used TIM 16 8JUN21 @

Application Start Time: <u>1023</u>
Application End Time: <u>1026</u>
Estimated Volume Remaining: <u>1750</u>
Disposal Method: <u>Fallow Field</u>
Total Pass Time: <u>42 sec</u>
Applied By: [REDACTED] <u>08 June 2021</u>

Application Rate Verification Calculation:Expected Passtime
(Based on
$$\text{Time per pass} \times \text{No. of passes} = \text{Expected Total Passtime}$$

Actual Passtime:

$$\frac{\text{Total Pass time}}{\text{Expected Pass time}} = \text{Percent Application Rate}$$

Actual a.i. output:

$$\text{Target FP application} \times \text{Percent Application Rate} = \text{Total FP Applied}$$

APPLICATION 2021 TRIAL 2

Study No.: PRTF RT25 Ring Test 2021 Trial 2

Date: 16 Sep 2021

Application Event: 2021 trial 2

Sprayer ID: SPR 07

Calibration Information

Nozzle output per 20 seconds

Output measured in (mL, L, gal, etc.): mL

Sprayer Pressure: LVL 3

Sprayer Swath: 55 inches

Output (mL)				Total	Avg
	Run 1	Run 2	Run 3		
Time	20.00	20.25	20.18	60.43	20.14 seconds
Nozzle 1 (mL)	175	180	178	533	177.67 mL
Nozzle 2 (mL)	177	180	179	536	178.67 mL
Output (mL)/sec	17.60	17.78	17.69	Average mL/sec: 17.69	
Variance 95%	16.81				
Variance 105%	18.57				

Date: 16 Sep 2021

Initials: [REDACTED]

Timer: TIM 18

072

Study No.: PRTF RT25 2021 trial 2

Date: 16 sep 2021

Application Event: Trial 2

Sprayer Description

Sprayer ID: SPR07

Description: backpack sprayer with internal circulation, modified with 2 nozzles

PSI: LVL 2 of 5

Number of Nozzles: 2

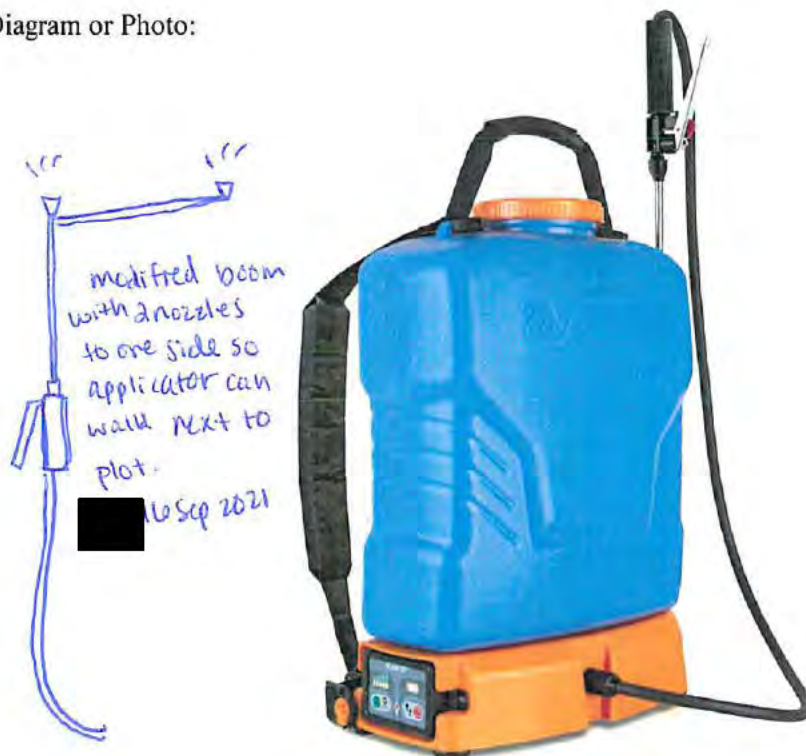
Nozzle screen: red screen, nozzles: yellow teejet
"turbo" 02 fan

Swath width: 55 inches

Nozzle distance to target: ~ 12 inches

Agitation type during mixing: shaken, poured into tank, circulation turned on

Sprayer Diagram or Photo:



Initial/Date: 16 Sep 2021

Application Environmental Conditions

Parameter	Reading	Instrument/Monitor
Temperature (°C)	③ 25.2	③ CRC HOBO station
Humidity	③ ① 80.81.0	③ CRC HOBO station
Wind Speed	③ 0 mph	③ CRC HOBO station
Wind Direction	③ NA	③ N/A
Soil Conditions	Dry / Moist / Wet	N/A
Crop Height	② 18 - 28 cm	N/A
% Crop Coverage in Plot	② 75% - 90%	N/A
Crop Type	Alfalfa	N/A
Species Name	<i>medicago sativa</i>	N/A
Distance between Control and Treatment Plots	② ~34 ft	N/A

① WE 16 sep 2021 ASW

② recorded by AW 16 sep 2021

③ recorded by [REDACTED] 16 sep 2021

Control Application Calculations

Application variables		
Description	Value	Units
Application rate	0.5	lbs a.i./acre
Plot size	128	sq ft.
Sprayer rate	17.69	mL/sec
spray coverage	100	L/acre

Conversions	
Sq ft per acre	43560
mL per L	1000
g per lb	453.592

Plot size (acres)	Total spray solution for plot (L)	Total spray solution for plot (mL)	Total spray time (sec)
0.002938476	0.293847567	293.8475666	16.61094215

Total application solution	
Overage	8.5078125
Total mL water	2500

Study No.: PRTF RT25 2021 Trial 2 (1)
 Initial/Date: 16 Sep 2021 (1)
 Application Event: 2021 Trial 2 (2)
Application Information

Amount of Water: 2500 mL (2) Time of Mixing: 9:40 am (2)
 Container No.: N/A Amount used: N/A

Application Pass Times (sec):	
Plot	Time (sec)
Control 1	15.71 (2)
Control 2	NA (2)

Application Start Time:	9:52 (2)
Application End Time:	9:53 (2)
Estimated Volume Remaining:	NA (1)
Disposal Method:	NA (1)
Total Pass Time:	15.71 (1)
Applied By:	16 Sep 2021 (1)

Application Rate Verification Calculation:

~~Expected Passtime
(Based on~~

~~Time per pass x No. of passes = Expected Total Passtime~~

~~Actual Passtime:~~

~~Total Pass time ÷ Expected Passtime = Percent Application Rate~~

~~Actual a.i. output:~~

~~Target FP application x Percent Application Rate = Total FP Applied~~

(1) recorded by 16 Sep 2021
 (2) recorded by 16 Sep 2021

Treatment Application Calculations

Application variables		
Description	Value	Units
Application rate	0.5	lbs a.i./acre
Plot size	240	sq ft.
Sprayer rate	17.69	mL/sec
spray coverage	100	L/acre
Density	1.1	g/mL
Purity	0.47415	g a.i./mL
Purity	43.5	%

Conversions	
Sq ft per acre	43560
mL per L	1000
g per lb	453.592

Plot size (acres)	Total a.i. needed (lbs)	Total a.i. needed (g)	Total test material needed for plot (mL)	Total spray solution for plot (L)	Total spray solution for plot (mL)	Total water (mL)	Total spray time (sec)
0.005509642	0.002754821	1.249564738	2.635378547	0.550964187	550.9641873	548.3288088	31.14551653

Total application solution	
Overage	3.794521288
Total mL test material	10.00
Total mL water	2080.6453

Spray time per treatment plot:

Plots 1-10:	20.76	Sec
Plots 11-15:	10.38	Sec

Study No.: PRTF RT25 2021 Trial 2 ②

Initial/Date: 16 Sep 2021 ②

Application Event: Trial 2 2021 ②

Application Information

Amount of Water: 2080.6 mL ② Time of Mixing: ③ 0911 Jug shaken for 1 minute
 Container No.: 21-36 ② Amount used: 10.0 mL ②

Application Pass Times (sec):	
Plot	Time (sec)
TRT1	21.31 ③
TRT2	10.8 ③
Total time:	31.39 ③

Application Start Time: ③ 10:08
Application End Time: ③ 10:10 ② 10:10
Estimated Volume Remaining: NA WE 400 16 Sep 21 1250mL ②
Disposal Method: WE 16 Sep 21 NA Fallow Field ②
Total Pass Time: 31.39 ②
Applied By: 16 Sep 2021 ②

Application Rate Verification Calculation:

~~Expected Passtime~~

(Based on) $\frac{\text{Time per pass}}{\text{Total Pass time}} \times \text{No. of passes} = \text{Expected Total Passtime}$

~~Actual Passtime:~~

$\frac{\text{Total Pass time}}{\text{Expected Pass time}} = \text{Percent Application Rate}$

~~Actual a.i. output:~~

$\text{Target FP application} \times \text{Percent Application Rate} = \text{Total FP Applied}$

① WE 16 Sep 2021

② recorded by 16 Sep 2021

③ recorded by 16 Sep 2021

ENVIRONMENTAL CHAMBER CONDITIONS 2021
TRIAL 1

Month June Year 2021

Environmental Chamber ID: ECOL6

Parameter	Temperature	Humidity
Target Range:	34-35 °C	45-90%
Meter Used:	TH309	ECO6H

Date	Temperature °C		% Relative Humidity	Init.
	Min	Max		
02 Jun 2021	34.1	34.2	70	
03 Jun 2021	34.1	34.7	70	
04 Jun 2021	34.5	34.7	70	
05 June 2021	34.4	34.7	70	
06 June 2021	34.6	34.7	70	
07 June 2021	34.4	34.7	70	
08 Jun 2021	33.9	34.7	70	
09 Jun 2021	34.2	34.7	70	
10 Jun 2021	34.4	34.7	70	
11 June 2021	34.3	34.8	70	
12 Jun 2021	34.2	34.8	70	
13 Jun 2021	34.2	34.8	70	
14 Jun 2021	34.2	34.8	70	
15 Jun 2021	34.3	34.6	70	
16 Jun 2021	34.2	34.9	70	
17 Jun 2021	34.0	35.3	70	
18 Jun 2021	34.0	35.3	70	

① WE [REDACTED] "Jun" 16 Jun 2021

[illegible]

080

ENVIRONMENTAL CHAMBER CONDITIONS 2021

TRIAL 2

MORTALITY AND OBSERVATIONS 2021
TRIAL 1

Field Location: Smithers	≤ 4 - Hour Observations		24 - Hour Observations	
Post-application interval: 6 Hours	In/Date: [REDACTED] 08 June 2021 Time: 1714		In/Date: [REDACTED] 09 Jun 2021 Time: 1700	
Time bees exposed: 1700	Test Concentration		Test Concentration	
	Control	T1	Control	T1
Replicate	# Dead	# Dead	# Dead	# Dead
1	0	0	0	25
2	0	0	0	25
3	0	0	0	25
4	0	0	0	25
5	0	0	0	25
6	0	0	0	23
Total	0	0	0	148

Note: 25 bees per replicate. Number dead per replicate = cumulative number dead for that replicate since dead bees are not removed.

Field Location: Smithers

Post-application interval: 6 Hours

Replicate	Behavioral Observations - Control	
	Initial/Date: [REDACTED] 08 Jun 2021	Initial/Date: [REDACTED] 09 Jun 2021
	≤ 4 - Hour Observations	24 - Hour Observations
1	2 lethargic N WE AW 08 Jun 2021	N
2	N	N
3	N	N
4	N	N
5	N	N
6	N	N

Observation abbreviations:

N=normal
MO=moribund

IN=intoxicated
AT=ataxia

TR=trembling

084

Field Location: Smithers

Post-application interval: 6 Hours

Replicate	Behavioral Observations - Treatment	
	Initial/Date: [REDACTED] 08 June 2021	Initial/Date: [REDACTED] 09 June 2021
	≤ 4 - Hour Observations	24 - Hour Observations
1	2 lethargic	all dead
2	N	all dead
3	N	all dead
4	1 lethargic	all dead
5	2 lethargic	AW 09 June 2021 2 remaining bees all dead
6	N	2 remaining bees moving slowly

Observation abbreviations:

N=normal
MO=moribund

IN=intoxicated
AT=ataxia

TR=trembling

Field Location: Smithers	≤ 4 - Hour Observations		24 - Hour Observations	
Post-application interval: 24 Hours	In/Date: [REDACTED] 09 June 2021 Time: 1525		In/Date: [REDACTED] 10 June 2021 Time: 1126	
Time bees exposed: 1126	Test Concentration		Test Concentration	
	Control	T1	Control	T1
Replicate	# Dead	# Dead	# Dead	# Dead
1	0	0	0	0
2	0	0	0	1
3	0	1	1	1
4	0	0	0	14
5	0	0	0	1
6	0	0	0	0
Total	0	1	1	17

Note: 25 bees per replicate. Number dead per replicate = cumulative number dead for that replicate since dead bees are not removed.

Field Location: Smithers

Post-application interval: 24 Hours

Behavioral Observations - Control		
	Initial/Date: [REDACTED] 09 June 2021	Initial/Date: [REDACTED] 10 June 2021
Replicate	≤ 4 - Hour Observations	24 - Hour Observations
1	N	N
2	N	N
3	N	N
4	N	N
5	N	N
6	N	N

Observation abbreviations:

N=normal
MO=moribund

IN=intoxicated
AT=ataxia

TR=trembling

086

Field Location: Smithers

Post-application interval: 24 Hours

Behavioral Observations - Treatment		
Initial/Date: [REDACTED] 09 June 2021		Initial/Date: [REDACTED] 10 June 2021
Replicate	≤ 4 - Hour Observations	24 - Hour Observations
1	N	N
2	N	N
3	N	N
4	N	N
5	N	N
6	N	N

Observation abbreviations:

N=normal
MO=moribund

IN=intoxicated
AT=ataxia

TR=trembling

Biological Observations for
Foliar Bee Exposure Study

PRTF RT25 Ring Test 2021

Field Location: Eurofins	≤ 4 - Hour Observations		24 - Hour Observations	
Post-application interval: 6 Hours	In/Date: <u>09 June 2021</u> Time: <u>1826</u>		In/Date: <u>10 June 2021</u> Time: <u>1820</u>	
Time bees exposed: <u>1820</u>	Test Concentration		Test Concentration	
	Control	T1	Control	T1
Replicate	# Dead	# Dead	# Dead	# Dead
1	0	0	0	24
2	0	0	0	25
3	0	0	0	25
4	0	0	0	① 25
5	0	0	0	① 24
6	0	0	1	① X 24
Total	0	0	1	14 ^{WF} 2021

Note: 25 bees per replicate. Number dead per replicate = cumulative number dead for that replicate since dead bees are not removed.

① WF 10 June 2021

Field Location: Eurofins

Post-application interval: 6 Hours

Replicate	Behavioral Observations - Control	
	Initial/Date: <u>09 June 2021</u>	Initial/Date: <u>10 June 2021</u>
	≤ 4 - Hour Observations	24 - Hour Observations
1	N	N
2	N	N
3	N	N
4	N	N
5	N	N
6	N	N

Observation abbreviations:

N=normal
MO=moribund

IN=intoxicated
AT=ataxia

TR=trembling

088

Field Location: Eurofins

Post-application interval: 6 Hours

Behavioral Observations - Treatment		
	Initial/Date: [REDACTED] 09 June 2021	Initial/Date: [REDACTED] 10 Jun 21
Replicate	≤ 4 - Hour Observations	24 - Hour Observations
1	N	remaining 1 bee lethargic
2	1 lethargic	all dead
3	N	all dead
4	N	all dead
5	N	remaining 1 bee lethargic
6	N	remaining 1 bee lethargic

Observation abbreviations:

N=normal
MO=moribund

IN=intoxicated
AT=ataxia

TR=trembling

Field Location: Eurofins	≤ 4 - Hour Observations		24 - Hour Observations	
Post-application interval: 24 Hours	In/Date: <u>10 June 2021</u>	In/Date: <u>11 Jun 2021</u>		
	Time: <u>1608</u>	Time: <u>1222</u>		
Time bees exposed: <u>1222</u>	Test Concentration		Test Concentration	
	Control	T1	Control	T1
Replicate	# Dead	# Dead	# Dead	# Dead
1	0	0	0	0
2	0	0	0	0
3	0	0	0	0
4	0	0	0	0
5	0	0	0	0
6	0	0	1	0
Total	0	0	1	0

Note: 25 bees per replicate. Number dead per replicate = cumulative number dead for that replicate since dead bees are not removed.

Field Location: Eurofins

Post-application interval: 24 Hours

Replicate	Behavioral Observations - Control	
	Initial/Date: <u>10 June 2021</u>	Initial/Date: <u>11 June 2021</u>
	≤ 4 - Hour Observations	24 - Hour Observations
1	N	N
2	N	N
3	N	N
4	N	N
5	N	N
6	N	N

Observation abbreviations:

N=normal
MO=moribund

IN=intoxicated
AT=ataxia

TR=trembling

Field Location: Eurofins

Post-application interval: 24 Hours

Behavioral Observations - Treatment		
	Initial/Date: [REDACTED] 10 June 2021	Initial/Date: [REDACTED] 11 June 2021
Replicate	≤ 4 - Hour Observations	24 - Hour Observations
1	N	N
2	N	N
3	N	N
4	N	N
5	N	N
6	N	N

Observation abbreviations:

N=normal
MO=moribund

IN=intoxicated
AT=ataxia

TR=trembling

091

MORTALITY AND OBSERVATIONS 2021
TRIAL 2

Field Location: Eurofins	≤ 4 - Hour Observations		24 - Hour Observations	
Post-application interval: 6 Hours	In/Date: [REDACTED] 14 Sep 2021 Time: 1735		In/Date: [REDACTED] 15 Sep 2021 Time: 1705	
Time bees exposed: 1705	Test Concentration		Test Concentration	
	Control	T1	Control	T1
Replicate	# Dead	# Dead	# Dead	# Dead
1	0	0	2	25
2	0	0	5	25
3	0	0	9	25
4	0	0	7	25
5	0	0	5	25
6	0	0	5	25
Total	0	0	33	150 ^{NP} 15 Sep 21

Note: 25 bees per replicate. Number dead per replicate = cumulative number dead for that replicate since dead bees are not removed.

Field Location: Eurofins

Post-application interval: 6 Hours

Behavioral Observations - Control		
	Initial/Date: [REDACTED] 14 Sep 2021	Initial/Date: [REDACTED] 15 Sep 2021
Replicate	≤ 4 - Hour Observations	① 24 - Hour Observations
1	all N	all N
2	all N	all N
3	all N	all N
4	all N	all N
5	all N	all N
6	all N	all N

Observation abbreviations:

N=normal
MO=moribund

IN=intoxicated
AT=ataxia

TR=trembling

093

① Bees appear to be having a somewhat more difficult time climbing wall of cages than normal [REDACTED] 15 Sep 2021

Field Location: Eurofins

Post-application interval: 6 Hours

	Behavioral Observations - Treatment	
	Initial/Date: [REDACTED] 14 sep 2021	Initial/Date: [REDACTED] 15 sep 2021
Replicate	≤ 4 - Hour Observations	24 - Hour Observations
1	all N	all dead
2	all N	all dead
3	all N	all dead
4	all N	all dead
5	all N	all dead
6	all N	all dead

Observation abbreviations:

N=normal
MO=moribund

IN=intoxicated
AT=ataxia

TR=trembling

Field Location: Eurofins	≤ 4 - Hour Observations		24 - Hour Observations	
Post-application interval: 24 Hours	In/Date: [REDACTED] 15 Sep 2021	In/Date: [REDACTED] 16 Sep 2021		
	Time: 1211	Time: 1132		
Time bees exposed: 1132	Test Concentration		Test Concentration	
	Control	T1	Control	T1
Replicate	# Dead	# Dead	# Dead	# Dead
1	0	0	2	0
2	0	0	4	1
3	0	0	2	2
4	0	0	1	1
5	0	0	0	1
6	0	0	4	2
Total	0	0	13	7

Note: 25 bees per replicate. Number dead per replicate = cumulative number dead for that replicate since dead bees are not removed.

Field Location: Eurofins

Post-application interval: 24 Hours

Replicate	Behavioral Observations - Control	
	Initial/Date: [REDACTED] 15 Sep 2021	Initial/Date: [REDACTED] 16 Sep 2021
	≤ 4 - Hour Observations	24 - Hour Observations
1	all N	all N
2	all N	all N
3	all N	all N
4	all N	all N
5	all N	all N
6	all N	all N

Observation abbreviations:

N=normal
MO=moribund

IN=intoxicated
AT=ataxia

TR=trembling

095

Field Location: Eurofins

Post-application interval: 24 Hours

Behavioral Observations - Treatment		
	Initial/Date: [REDACTED] 15 Sep 2021	Initial/Date: 16 Sep 2021 [REDACTED]
Replicate	≤ 4 - Hour Observations	24 - Hour Observations
1	all N	all N
2	all N	all N
3	all N	all N
4	all N	all N
5	all N	all N
6	all N	all N

Observation abbreviations:

N=normal
MO=moribund

IN=intoxicated
AT=ataxia

TR=trembling

Field Location: Smithers	≤ 4 - Hour Observations		24 - Hour Observations	
Post-application interval: 6 Hours	In/Date: [redacted] 16 sep 2021 Time: 1739		In/Date: [redacted] 17 sep 2021 Time: 1720	
Time bees exposed: 1709	Test Concentration		Test Concentration	
	Control	T1	Control	T1
Replicate	# Dead	# Dead	# Dead	# Dead
1	0	0	0	2
2	0	0	1	10
3	0	0	2	2
4	0	0	0	0
5	0	0	1	2
6	0	0	1	0
Total	0	0	5	16

Note: 25 bees per replicate. Number dead per replicate = cumulative number dead for that replicate since dead bees are not removed.

Field Location: Smithers

Post-application interval: 6 Hours

Replicate	Behavioral Observations - Control	
	Initial/Date: [redacted] 16 sep 2021	Initial/Date: [redacted] 17 sep 2021
	≤ 4 - Hour Observations	24 - Hour Observations
1	all N	all N
2	all N	all N
3	all N	all N
4	all N	all N
5	all N	all N
6	all N	all N

Observation abbreviations:

N=normal
MO=moribund

IN=intoxicated
AT=ataxia

TR=trembling

097

Field Location: Smithers

Post-application interval: 6 Hours

	Behavioral Observations - Treatment	
	Initial/Date: [REDACTED] 16 Sep 2021	Initial/Date: [REDACTED] 17 Sep 2021
Replicate	≤ 4 - Hour Observations	24 - Hour Observations
1	all N	all N
2	all N	all N
3	all N	all N
4	all N	all N
5	all N	all N
6	all N	all N

Observation abbreviations:

N=normal
MO=moribund

IN=intoxicated
AT=ataxia

TR=trembling

Field Location: Smithers	≤ 4 - Hour Observations		24 - Hour Observations	
Post-application interval: 24 Hours	In/Date: [redacted] 17 SEP 2021 Time: 1739 1140		In/Date: [redacted] 18 SEP 2021 Time: 1055	
Time bees exposed: 1709/1055	Test Concentration		Test Concentration	
	Control	T1	Control	T1
Replicate	# Dead	# Dead	# Dead	# Dead
1	0	1	1	16
2	0	4	2	23
3	2	2	3	9
4	0	0	5	18
5	0	2	1	21
6	1	0	1	12
Total	3	9	13	99

Note: 25 bees per replicate. Number dead per replicate = cumulative number dead for that replicate since dead bees are not removed.

Field Location: Smithers

Post-application interval: 24 Hours

Replicate	Behavioral Observations - Control	
	Initial/Date: [redacted] 17 SEP 2021	Initial/Date: [redacted] 18 SEP 2021
	≤ 4 - Hour Observations	24 - Hour Observations
1	all N	all N
2	all N	all N
3	all N	all N
4	all N	all N
5	all N	all N
6	all N	all N

Observation abbreviations:

N=normal
MO=moribund

IN=intoxicated
AT=ataxia

TR=trembling

099

Field Location: Smithers

Post-application interval: 24 Hours

	Behavioral Observations - Treatment	
	Initial/Date: [REDACTED] 17 sep 2021	Initial/Date: [REDACTED] 18 sep 2021
Replicate	≤ 4 - Hour Observations	24 - Hour Observations
1	all N	all N
2	all N	all N
3	all N	all N
4	all N	all N
5	all N	all N
6	all N	all N

Observation abbreviations:

N=normal
MO=moribund

IN=intoxicated
AT=ataxia

TR=trembling

Field Location: Smithers	≤ 4 - Hour Observations		24 - Hour Observations	
Post-application interval: 48 Hours	In/Date: [REDACTED] 18 Sep 2021 Time: 1258		In/Date: [REDACTED] 19 Sep 2021 Time: 1203	
Time bees exposed: 1203	Test Concentration		Test Concentration	
	Control	T1	Control	T1
Replicate	# Dead	# Dead	# Dead	# Dead
1	0	0	0	0
2	0	0	0	0
3	0	0	2	1
4	0	0	0	0
5	0	0	1	0
6	0	0	0	0
Total	0	0	3	1

Note: 25 bees per replicate. Number dead per replicate = cumulative number dead for that replicate since dead bees are not removed.

Field Location: Smithers

Post-application interval: 48 Hours

Replicate	Behavioral Observations - Control	
	Initial/Date: [REDACTED] 18 Sep 2021	Initial/Date: [REDACTED] 19 Sep 2021
	≤ 4 - Hour Observations	24 - Hour Observations
1	all N	all N
2	all N	all N
3	all N	all N
4	all N	all N
5	all N	all N
6	all N	all N

Observation abbreviations:

N=normal
MO=moribund

IN=intoxicated
AT=ataxia

TR=trembling

Field Location: Smithers

Post-application interval: 48 Hours

	Behavioral Observations - Treatment	
	Initial/Date: [REDACTED] 18 Sep 2021	Initial/Date: [REDACTED] 19 Sep 2021
Replicate	≤ 4 - Hour Observations	24 - Hour Observations
1	all N	all N
2	all N	all N
3	all N	all N
4	all N	all N
5	all N	all N
6	all N	all N

Observation abbreviations:

N=normal
MO=moribund

IN=intoxicated
AT=ataxia

TR=trembling

FOLIAGE COLLECTION 2021

TRIAL 1

TRIAL 1
MF [REDACTED] 20 DEC 2021

Location: Smithers

Post application Interval: 6hrs; harvest time:

WE 08 Jun 2021
~~0070~~ 1600

Cloud Cover: 4070

Weight of Foliage Aliquots (target 15 g)		
Replicate	Control (g)	T1 (g)
1	15.0	15.0
2	15.0	15.0
3	15.0	15.0
4	15.0	15.0
5	15.0	15.0
6	15.0	15.0

Time of exposure

Control: 1645

T1: ~~1658~~ WE [REDACTED] 08 Jun 2021 1700

Balance used: E030

Date/Initial: 08 Jun 2021 [REDACTED]

Trial 1
 (ME) 20 Dec 21

Location: Smithers

Post application Interval: 24hrs; harvest time: 1030

Cloud Cover: 95% very cloudy, no rain 09 June 2021

Weight of Foliage Aliquots (target 15 g)		
Replicate	Control (g)	T1 (g)
1	15.0	15.0
2	15.0	15.0
3	15.0	15.0
4	15.0	15.0
5	15.0	15.0
6	15.0	15.0

Time of exposure

Control: 1110

T1: 1126

Balance used: E030

Date/Initial: 09 June 2021

Location: Eurofins

Post application Interval: 6hrs

Trial 1
09 June 2021

Weight of Foliage Aliquots (target 15 g)		
Replicate	Control (g)	T1 (g)
1	15.0	15.0
2	15.0	15.0
3	15.0	15.0
4	15.0	15.0
5	15.0	15.0
6	15.0	15.0

Time of exposure

Control: 1810

T1: 1820

Balance used: 2030

Date/Initial: 09 June 2021

Location: Eurofins

Post application Interval: 24hrs

Trial 1
100224

Weight of Foliage Aliquots (target 15 g)		
Replicate	Control (g)	T1 (g)
1	15.0	15.0
2	15.0	15.0
3	15.0	15.0
4	15.0	15.0
5	15.0	15.0
6	15.0	15.0

Time of exposure

Control: 1213

T1: 1222

Balance used: E030

Date/Initial: 10 Jun 2021

FOLIAGE COLLECTION 2021

TRIAL 2

Location: Eurofins

Post application Interval: 6hrs

Weight of Foliage Aliquots (target 15 g)

Replicate	Control (g)	T1 (g)
1	15.0	15.0
2	15.0	15.0
3	15.0	15.0
4	15.0	15.0
5	15.0	15.0
6	15.0	15.0

Time of exposure

Control:

T1:

Balance used:

Date/Initial:

Location: Eurofins

Post application Interval: 24hrs

Trial 2
15 Sep 2021

Weight of Foliage Aliquots (target 15 g)		
Replicate	Control (g)	T1 (g)
1	15.0	15.0
2	15.0	15.0
3	15.0	15.0
4	15.0	15.0
5	15.0	15.0
6	15.0	15.0

Time of exposure

Control: 11:16

T1: 11:32

Balance used: E030

Date/Initial: 15 Sep 2021

Trial 2
MF 16 Sep 2021

Location: Smithers

Post application Interval: 6hrs

cloud cover: 100% overcast

Weight of Foliage Aliquots (target 15 g)		
Replicate	Control (g)	T1 (g)
1	15.0	15.0
2	15.0	15.0
3	15.0	15.0
4	15.0	15.0
5	15.0	15.0
6	15.0	15.0

MF 16 Sep 2021

Time of exposure

Control: 1700

T1: 1709

Balance used: E030

Date/Initial 16 Sep 2021

Location: Smithers

Post application Interval: 24hrs

cloud cover: 100%

Weight of Foliage Aliquots (target 15 g)

Replicate	Control (g)	T1 (g)
1	15.0	15.0
2	15.0	15.0
3	15.0	15.0
4	15.0	15.0
5	15.0	15.0
6	15.0	15.0

Time of exposure

Control: 1041

T1: 1055

Balance used: E030

Date/Initial: 17 Sep 2021

Location: Smithers

Post application Interval: 48hrs

cloud cover: 30% (MF)

18 Sep 2021

Weight of Foliage Aliquots (target 15 g)

Replicate	Control (g)	T1 (g)
1	15.0	15.0
2	15.0	15.0
3	15.0	15.0
4	15.0	15.0
5	15.0	15.0
6	15.0	15.0

Time of exposure

Control: 1200

T1: 1203

Balance used: FSS

Date/Initial:

18 Sep 2021

SAMPLE INFORMATION 2021

TRIAL 1

Treatment	Timing	Matrix	Sample ID	Pre-weight, card or bag (g)	Scale ID: E030
UTC	0hr	Spray Card	SM.UTC.SC1	0.7	In/Date: 04 June 2021
UTC	0hr	Spray Card	SM.UTC.SC2	0.6	
UTC	0hr	Spray Card	SM.UTC.SC3	0.6	
TRT	0hr	Spray Card	SM.TRT.SC1	0.7	
TRT	0hr	Spray Card	SM.TRT.SC2	0.7	
TRT	0hr	Spray Card	SM.TRT.SC3	0.6	
UTC	0hr	Foliage	SM.UTC.0hr.FA	3.9	
UTC	0hr	Foliage	SM.UTC.0hr.FB	4.0	
TRT	0hr	Foliage	SM.TRT.0hr.FA	3.9	
TRT	0hr	Foliage	SM.TRT.0hr.FB	3.9	
UTC	6hr	Foliage	SM.UTC.6hr.FA	3.9	
UTC	6hr	Foliage	SM.UTC.6hr.FB	3.9	
TRT	6hr	Foliage	SM.TRT.6hr.FA	4.0	
TRT	6hr	Foliage	SM.TRT.6hr.FB	3.9	
UTC	24hr	Foliage	SM.UTC.24hr.FA	3.9	
UTC	24hr	Foliage	SM.UTC.24hr.FB	4.0	
TRT	24hr	Foliage	SM.TRT.24hr.FA	3.9	
TRT	24hr	Foliage	SM.TRT.24hr.FB	3.8	
UTC	48hr	Foliage	SM.UTC.48hr.FA	3.9	
UTC	48hr	Foliage	SM.UTC.48hr.FB	4.1	
TRT	48hr	Foliage	SM.TRT.48hr.FA	3.7	
TRT	48hr	Foliage	SM.TRT.48hr.FB	3.9	

Sample List and Handling

PRTF RT25 Ring Test 2021

Treatment	Timing	Matrix	Sample ID	Time Collected	Gross Weight (g)	Sample Weight (g)	Scale ID	Time in Freezer	Date	Initial
UTC	0hr	Tank Mix	SM.UTC.TMA		N/A	N/A	N/A			
UTC	0hr	Tank Mix	SM.UTC.TMB		N/A	N/A	N/A			
TRT	0hr	Tank Mix	SM.TRT.TMA	1016	N/A	N/A	N/A	1111	08 June 2021	
TRT	0hr	Tank Mix	SM.TRT.TMB	1016	N/A	N/A	N/A	1111	08 Jun 2021	
UTC	0hr	Spray Card	SM.UTC.SC1	1002	5.1 ⁰	0.4	E025	1111	08 Jun 2021	
UTC	0hr	Spray Card	SM.UTC.SC2	1002	1.5 ⁰	0.5	E025	1111	08 Jun 2021	
UTC	0hr	Spray Card	SM.UTC.SC3	1002	1.0 ⁰	0.4	E025	1111	08 Jun 2021	
TRT	0hr	Spray Card	SM.TRT.SC1	1027	4.8 ⁰	0.1	E025	1111	08 Jun 2021	
TRT	0hr	Spray Card	SM.TRT.SC2	1027	5.0 ⁰	0.3	E025	1111	08 Jun 2021	
TRT	0hr	Spray Card	SM.TRT.SC3	1027	4.7 ⁰	0.2	E025	1111	08 Jun 2021	
UTC	0hr	Foliage	SM.UTC.0hr.FA	1045	37.2	33.3	E025	1111	08 Jun 2021	
UTC	0hr	Foliage	SM.UTC.0hr.FB	1045	49.6	45.7	E025	1111	08 Jun 2021	
TRT	0hr	Foliage	SM.TRT.0hr.FA	1101	55.4	51.5	E025	1111	08 Jun 2021	
TRT	0hr	Foliage	SM.TRT.0hr.FB	1101	57.4	53.5	E025	1111	08 Jun 2021	

08 JUN 21 @

08 June 2021

Sample (MF) 08 June 2021

Treatment	Timing	Matrix	Sample ID	Time Collected	Gross Weight (g)	Sample Weight (g)	Scale ID	Time in Freezer	Date	Initial
UTC	6hr	Foliage	SM.UTC.6hr.FA	16:29	15.0	14.0	E030	1702	08 June 2021	
UTC	6hr	Foliage	SM.UTC.6hr.FB	16:30	15.1		E030	1702	08 June 2021	
TRT	6hr	Foliage	SM.TRT.6hr.FA	16:53	15.3		E030	1702	08 June 2021	
TRT	6hr	Foliage	SM.TRT.6hr.FB	16:55	15.2		E030	1702	08 June 2021	
UTC	24hr	Foliage	SM.UTC.24hr.FA	10:59	17.3		E030	1132	09 Jun 21	
UTC	24hr	Foliage	SM.UTC.24hr.FB	10:59	16.8		E030	1132	09 Jun 2021	
TRT	24hr	Foliage	SM.TRT.24hr.FA	11:15	16.0		E030	1132	09 Jun 2021	
TRT	24hr	Foliage	SM.TRT.24hr.FB	11:17	16.2		E030	1132	09 Jun 2021	
UTC	48hr	Foliage	SM.UTC.48hr.FA							
UTC	48hr	Foliage	SM.UTC.48hr.FB							
TRT	48hr	Foliage	SM.TRT.48hr.FA							
TRT	48hr	Foliage	SM.TRT.48hr.FB							

NA Samples weighed out in weigh bag + sample weight not a factor 08 June 2021

NA 09 Nov 2021

NA 09 Nov 2021

SAMPLE INFORMATION 2021

TRIAL 2

Treatment	Timing	Matrix	Sample ID	Pre-weight, card or bag (g)	Scale ID: <u>E033</u>
UTC	0hr	Spray Card	SM2.UTC.SC1	4.7	In/Date: <u>13 Sep 2021</u>
UTC	0hr	Spray Card	SM2.UTC.SC2	4.5	
UTC	0hr	Spray Card	SM2.UTC.SC3	4.5	
TRT	0hr	Spray Card	SM2.TRT.SC1	4.6	
TRT	0hr	Spray Card	SM2.TRT.SC2	4.5	
TRT	0hr	Spray Card	SM2.TRT.SC3	4.5	
UTC	0hr	Foliage	SM2.UTC.0hr.FA	3.9	
UTC	0hr	Foliage	SM2.UTC.0hr.FB	3.9	
TRT	0hr	Foliage	SM2.TRT.0hr.FA	3.8	
TRT	0hr	Foliage	SM2.TRT.0hr.FB	3.9	
UTC	6hr	Foliage	SM2.UTC.6hr.FA	3.9	
UTC	6hr	Foliage	SM2.UTC.6hr.FB	3.8	
TRT	6hr	Foliage	SM2.TRT.6hr.FA	3.9	
TRT	6hr	Foliage	SM2.TRT.6hr.FB	3.8	
UTC	24hr	Foliage	SM2.UTC.24hr.FA	3.9	
UTC	24hr	Foliage	SM2.UTC.24hr.FB	3.9	
TRT	24hr	Foliage	SM2.TRT.24hr.FA	3.9	
TRT	24hr	Foliage	SM2.TRT.24hr.FB	3.8	

Sample List and Handling

PRTF RT25 Ring Test 2021-Trial 2

Treatment	Timing	Matrix	Sample ID	Time Collected	Gross Weight (g)	Sample Weight (g)	Scale ID	Time in Freezer	Date	Initial
UTC	0hr	Tank Mix	SM2.UTC.TMA	0954	N/A	N/A	N/A	1056	16 Sep 21	
UTC	0hr	Tank Mix	SM2.UTC.TMB	0954	N/A	N/A	N/A	1056	16 Sep 21	
TRT	0hr	Tank Mix	SM2.TRT.TMA	1006	N/A	N/A	N/A	1056	16 Sep 21	
TRT	0hr	Tank Mix	SM2.TRT.TMB	1006	N/A	N/A	N/A	1056	16 Sep 21	
UTC	0hr	Spray Card	SM2.UTC.SC1	1006	5.0	0.3	E025	1056	16 Sep 21	
UTC	0hr	Spray Card	SM2.UTC.SC2	1000	4.8	0.3	E025	1056	16 Sep 21	
UTC	0hr	Spray Card	SM2.UTC.SC3	1000	5.0	0.5	E025	1056	16 Sep 21	
TRT	0hr	Spray Card	SM2.TRT.SC1	1015	5.2	0.6	E025	1056	16 Sep 21	
TRT	0hr	Spray Card	SM2.TRT.SC2	1015	4.8	0.3	E025	1056	16 Sep 21	
TRT	0hr	Spray Card	SM2.TRT.SC3	1015	4.8	0.3	E025	1056	16 Sep 21	
UTC	0hr	Foliage	SM2.UTC.0hr.FA	1024	38.5	34.6	E025	1056	16 Sep 21	
UTC	0hr	Foliage	SM2.UTC.0hr.FB	1024	24.7	20.8	E025	1056	16 Sep 21	
TRT	0hr	Foliage	SM2.TRT.0hr.FA	1028	30.8	27.0	E025	1056	16 Sep 21	
TRT	0hr	Foliage	SM2.TRT.0hr.FB	1028	33.7	29.8	E025	1056	16 Sep 21	

Treatment	Timing	Matrix	Sample ID	Time Collected	Gross Weight (g)	Sample Weight (g)	Scale ID	Time in Freezer	Date	Initial
UTC	6hr	Foliage	SM2.UTC.6hr.FA	1600	19.4	15.5	E025	1725	16sep21	
UTC	6hr	Foliage	SM2.UTC.6hr.FB	1600	18.9	15.1	E025	1725	16sep21	
TRT	6hr	Foliage	SM2.TRT.6hr.FA	1608	22.2	18.3	E025	1725	16sep21	
TRT	6hr	Foliage	SM2.TRT.6hr.FB	1608	22.8	19.0	E025	1725	16sep21	
UTC	24hr	Foliage	SM2.UTC.24hr.FA	1057 0957 17sep21	22.0	18.1	E01	1120	17sep21	
UTC	24hr	Foliage	SM2.UTC.24hr.FB	0957	24.0	20.1	E01	1120	17sep21	
TRT	24hr	Foliage	SM2.TRT.24hr.FA	1010	20.4	16.5	E01	1120	17sep21	
TRT	24hr	Foliage	SM2.TRT.24hr.FB	1010	23.0	19.2	E01	1120	17sep21	

Sample List and Handling

PRTF RT25 Ring Test 2021-Trial 2

Treatment	Timing	Matrix	Sample ID	Time Collected	Gross Weight (g)	Sample Weight (g)	Scale ID	Time in Freezer	Date	Initial
UTC	48hr	Foliage	SM2.UTC.48hr.FA	11:00		24.0	FSS	12:50	18 Sep 21	
UTC	48hr	Foliage	SM2.UTC.48hr.FB	11:06		25.2	FSS	12:50	18 Sep 21	
TRT	48hr	Foliage	SM2.TRT.48hr.FA	11:05		19.7	FSS	12:50	18 Sep 21	
TRT	48hr	Foliage	SM2.TRT.48hr.FB	11:05		16.9	FSS	12:50	18 Sep 21	

NOTES TO FILE

Study Number PRTF RTAS Ring Test 2021

Daily Log (Date & Initial each entry)

██████ +6hr samples transferred to ██████
via Max Feken. Samples packed into a cooler with
insulated dry ice and a temperature probe. ██████
confirmed that the temp was 8.9°C upon arrival
██████ 09 June 2021

██████ +24 samples transferred to ██████ via
Eric Peterson packed in a cooler with insulated dry
ice and a temperature monitor. Samples left around
10:40 am 09 June 2021 ██████ 09 June 2021

██████ +6hr samples arrived at 1755 via Max Feken,
min temp: 2.5 max: 5.5 °C ██████ 09 June 2021

██████ +24 samples arrived at 1157 via max Feken
samples placed on blue ice for transport, no temperature
monitor ██████ 10 June 2021

NA ██████ 09 Nov 2021

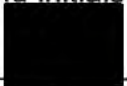
NOTE TO FILE

Study Number or Logbook Name: PRTF RT25 2021 Trial 2

Describe situation or observation:

On day 0, the day of application, the weather was overcast and 100% cloud cover though thin was present from the time of application until the 4hr foliage collection. At 1600. some light rain occurred, but only a very light sprinkle with a few raindrops here and there.

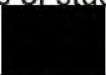
Date and initials of recorder:

 16 sep 2021

Study director or management assessment (if needed):

 16 sep 2021

Date and initials of study director/management:

 16 sep 2021

Appendix C

Anonymized Data Submittal – Lab B





FINAL SUMMARY REPORT

**Phase 1 Ring Test:
Standardization of Honey Bee Toxicity of Residues on Foliage (RT₂₅) Study Design –
Phase II**

TEST REGULATORY GUIDELINE(S)

EPA OCSPP 850.3030: Honey Bee Toxicity of Residues on Foliage

STUDY TRIAL COMPLETION DATE

September 18, 2021

STUDY DIRECTOR AND AUTHOR




TESTING FACILITY



SPONSOR

Pollinator Research Task Force (PRTF)

STUDY IDENTIFICATION

 Study Number: S21-04089

PAGE COUNT

1 of 14

TABLE OF CONTENTS

TABLE OF CONTENTS.....	2
GENERAL INFORMATION.....	3
STUDY PERSONNEL AND PERFORMING FACILITIES	3
I. SUMMARY.....	4
TABLE 1: Result Summary.....	9
FIGURE 1: Treatment Plot Layout.....	10
APPENDIX A: Detailed Field Conditions from Application to Last Foliage Sampling	11
APPENDIX B: Detailed Bee Testing Conditions (Bioassays 1 to 3).....	12
APPENDIX C: Application Calculations	13

GENERAL INFORMATION
STUDY PERSONNEL AND PERFORMING FACILITIES

[REDACTED] *Study Trial Number:* S21-04089

Study Title: Standardization of Honey Bee Toxicity of Residues on Foliage
(RT₂₅) Study Design – Phase II

Test Substances: Dimethoate 400 EC Formulation

Study Representative: Joseph D. Wisk

Sponsor: BASF Corporation
Agricultural Solutions
26 Davis Drive
Research Triangle Park, NC 27709-3528
USA

Testing Facility:

[REDACTED]

Testing Facility Personnel:

[REDACTED]

Study Trial Execution Dates:

Study Initiation Date (final protocol provided):

Biological Experimental Start Date: June 8, 2021

Biological Experimental Termination Date: September 18, 2021

I. SUMMARY

Study Objectives:

To determine the length of time over which field-weathered foliar residues of Dimethoate 400 EC formulation on alfalfa leaves remain toxic to the honey bee *Apis mellifera* L. The 24-hour RT₂₅ was determined. The RT₂₅ was the residual time at which the bee mortality observed in the treated groups was less than or equal to 25%.

Material and Methods:

Test item (T):	Dimethoate 400 EC
Description:	Liquid Formulation, Emulsifiable Concentrate
Lot/Batch No.:	01707-006
Active ingredients (AI):	Dimethoate
Content purity:	43.5% (nominal, see product label)
Expiration Date:	NA
Received Date:	10 Jun 2020
Last Used for Study:	14 Sep 2020
Storage conditions:	Kept Ambient

Test Species:	<i>Apis mellifera</i> L.
Age:	Young newly emerged adult workers of similar age and feeding status (3 to 5 days old)

Source:	Queen-right, healthy colonies were used from the stock hives maintained at the Eurofins Agrosience Services, LLC laboratory apiary located near Prospect Hill, NC, USA; they were monitored by the facility beekeeper. The hives used were 21165 and 21167 (June trial) and 21A146 (September trial).
---------	---

Preparation of Test System:	Young adult bees that were approximately the same age were emerged from capped brood frames that were placed into an emergence box. The emerged bees were brushed into separate large holding containers at different time points for proper and traceable aging of the bees and for acclimation until used for bioassays. The newly emerged bees were collected from the holding boxes and then introduced directly into the test units on the day of exposure. It was not necessary to anaesthetize the bees with CO ₂ prior to their introduction into the test units. Moribund or dead bees were rejected and replaced by healthy bees before starting the test. Reserve bees from the respective holding box were used as the replacement bees. Bees were fed <i>ad libitum</i> with untreated 50 % (w/v) aqueous sucrose solution.
-----------------------------	---

Test Unit:	Transparent 32 oz plastic containers (upper diameter = approx. 11 cm, base diameter = approx. 9 cm; height = approx. 14 cm) were used as test units. The top of the test units were covered with a screened lid to allow ventilation but prevent the escape of the honey bees. The lid of the test unit also had a hole where a feeding syringe was deposited. The feeder itself helped plug up the hole so that no bees can escape. The base and side walls of each test unit were covered with approximately 15 g of alfalfa foliage sampled in the field prior to exposure. As soon as the test units were prepared, the bees were transferred directly into the test units pre-loaded with untreated/treated alfalfa.
Test design:	<p>Extended laboratory study; one untreated water control (C) and one dose of Dimethoate 400 EC (T) were applied to alfalfa foliage under field conditions. A canopy was not needed over the treated plots because there was no rain at least 3 hrs after application. Treated foliage was collected at 6 hrs and 24 hrs after application (HAA) for bioassay 1 and 2, respectively. Each bioassay consisted of 6 replicates of 25 bees per replicate per group (150 bees per group). Bees were exposed to treated foliage (leaves and stems) in the laboratory and mortality and behavioural abnormalities were recorded 24 hrs after start of exposure. Additional bioassays were conducted using foliage provided by another laboratory after similar treatment with the control and test items.</p> <p>An initial trial was conducted in June 2021, and a second trial with similar study design was conducted in September 2021.</p>
Analytical Samples Taken:	<p>June and September Trials:</p> <ul style="list-style-type: none">• Untreated & Treated Tank Mix, A & R (50 mL each)• Fiber Glass Discs from Control & Treated Plots (3 reps each)• 1-hr Residual Foliage Samples, C & T plots, A & R (≥ 15 g each)• 6-hr Residual Foliage Samples, C & T plots, A & R (≥ 15 g each)• 24-hr Residual Foliage Samples, C & T plots, A & R (≥ 15 g each).

Field Conditions During
Application:
(See Appendix A
for details)

June Trial ([REDACTED]):

Avg. Temperature: 31°C (88°F)

Avg. Humidity: 61%

Avg. Wind Speed: 0.3 m/s (0.6 mph)

Wind Direction: East to West

Precipitation: No rain from start of application (09 Jun 2021 at 10:47) to last foliage sampling (10 Jun 2021 at 10:34)

September Trial ([REDACTED]):

Avg. Temperature: 25°C (77°F)

Avg. Humidity: 72%

Avg. Wind Speed: 0.4 m/s (1 mph)

Wind Direction: East to West

Precipitation: No rain from start of application (14 Sep 2021 at 09:21) to last foliage sampling (15 Sep 2021 at 09:13)

Bee Testing Conditions:
(See Appendix B
for details)

June Trial (all bioassays):

Target Temperature: 25 to 35°C (77 to 95°F)

Actual Temperature: 30 to 31°C (86 to 87°F)

Target Humidity: 50 to 80%

Actual Humidity: 36 to 53%

Lighting: Complete darkness, except during Assessments

September Trial (all bioassays):

Target Temperature: 25 to 35°C (77 to 95°F)

Actual Temperature: 26 to 28°C (79 to 83°F)

Target Humidity: 50 to 80%

Actual Humidity: 64 to 77%

Lighting: Complete darkness, except during Assessments

Target application volume: 200 L spray mix/ha

Calibrated output rate:

June Trial ([REDACTED]):

75.34 mL spray mix/sec

September Trial ([REDACTED]):

50.12 mL spray mix/sec

Test rates: (See Appendix C for detailed application calculations)	<i>Control (C):</i> untreated water spray <i>Dimethoate 400 EC (T):</i> <u>T Target Rate:</u> 560.4 g a.i./ha (1288.3 g product/ha) <u>June Trial T Actual Rate:</u> based on total actual application duration (9.46 sec) and calibrated output rate was 555 g a.i./ha (1274 g product/ha) <u>September Trial T Actual Rate:</u> based on total actual application duration (14.36 sec) and calibrated output rate was 560 g a.i./ha (1287 g product/ha)
Application equipment & procedure:	Calibrated boom sprayer (NC-SPR-12) with 6 nozzles (flat fan, TeeJet), giving 10 ft (3 m) coverage; Application performed at approximately 18 in above canopy.
Application Date & Times:	Untreated water control (C) plot: 09 Jun 2021 @ 10:30 EST Treated (T) plot: 09 Jun 2021 @ 10:47 EST Untreated water control (C) plot: 14 Sep 2021 @ 09:21 EST Treated (T) plot: 14 Sep 2021 @ 09:31 EST
Crop:	Alfalfa (<i>Medicago sativa</i>)
Crop Information:	June: Height = 12-14 in; BBCH approx. 60; Groundcover = 100% September: Height = 12 in; BBCH 58; Groundcover = 90%
Main Plot Size:	C and T main plots both 12 m x 3 m (See Figure 1)
Subplot size:	1 m x 1 m (see Figure 1)

Findings:*June Trial:***[REDACTED]** application

In the control group of 6 HAA and 24 HAA bioassays, the 24-hr cumulative mortality was 2% and 11%, respectively, which was below the 20% control mortality threshold set by the validity criteria (see Table 1). For the test item, total cumulative mortality in the 6 HAA and 24 HAA bioassays was 100% and 14% (equivalent to control-corrected mortality of 100% and 3%), respectively.

[REDACTED] application

In the control group of 6 HAA and 24 HAA bioassays, the 24-hr cumulative mortality was 0% and 9%, respectively, which was below the 20% control mortality threshold set by the validity criteria (see Table 1). For the test item, total cumulative mortality in the 6 HAA and 24 HAA bioassays was 100% and 5% (equivalent to control-corrected mortality of 100% and 4%), respectively.

*September Trial:***[REDACTED]** application

In the control group of 6 HAA and 24 HAA bioassays, the 24-hr cumulative mortality was 0% and 4%, respectively, which was below the 20% control mortality threshold set by the validity criteria (see Table 1). For the test item, total cumulative mortality in the 6 HAA and 24 HAA bioassays was 99% and 19% (equivalent to control-corrected mortality of 99% and 16%), respectively.

[REDACTED] application

In the control group of 6 HAA and 24 HAA bioassays, the 24-hr cumulative mortality was 3% and 5%, respectively, which was below the 20% control mortality threshold set by the validity criteria (see Table 1). For the test item, total cumulative mortality in the 6 HAA and 24 HAA bioassays was 37% and 21% (equivalent to control-corrected mortality of 36% and 16%), respectively.

Conclusions:

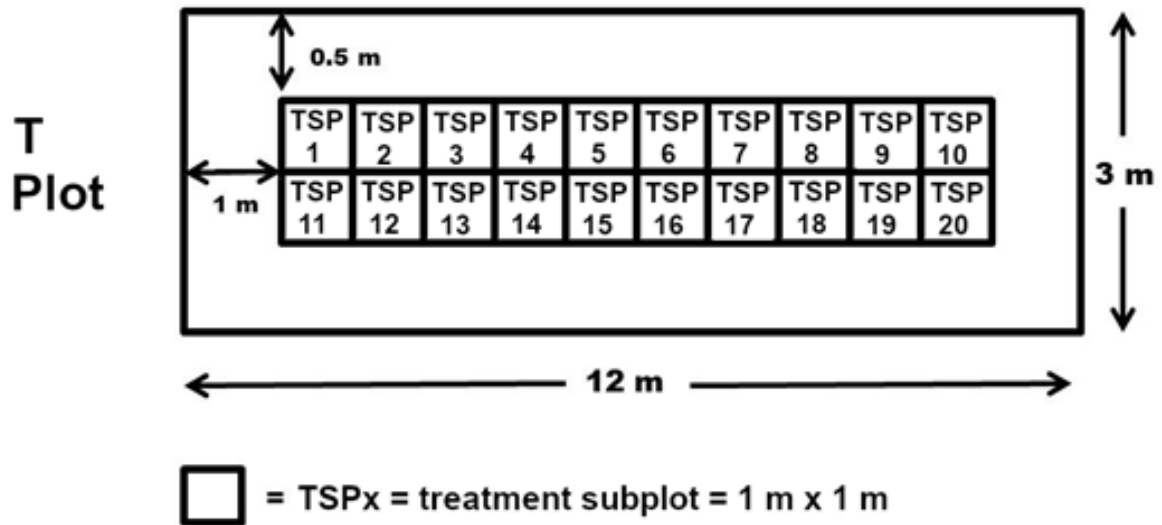
This study was deemed valid because the control mortality at the end of test (24 hrs) was below the 20% acceptance threshold for all bioassays.

According to the results of the study, the 24-hr RT₂₅ for the test formulation Dimethoate 400 EC (T) was determined to be > 6 hrs and < 24 hrs, at the target application rate of 560.4 g a.i./ha (0.5 lb a.i./Ac).

TABLE 1: Result Summary

Experimental Group ^A	Residual Time	Exposure Duration	# Bees Used	# Bees Dead	% Cumulative Mortality	% Corrected Mortality ^B
June Trial						
[REDACTED] Application						
C = Untreated Control	6 HR	24 HR	150	3	2	-
T= Dimethoate 400 EC	6 HR	24 HR	150	150	100	100
C = Untreated Control	24 HR	24 HR	150	17	11	-
T= Dimethoate 400 EC	24 HR	24 HR	150	21	14	3
[REDACTED] Application						
C = Untreated Control	6 HR	24 HR	150	0	0	-
T= Dimethoate 400 EC	6 HR	24 HR	150	150	100	100
C = Untreated Control	24 HR	24 HR	150	13	9	-
T= Dimethoate 400 EC	24 HR	24 HR	150	8	5	4
September Trial						
[REDACTED] Application						
C = Untreated Control	6 HR	24 HR	150	0	0	-
T= Dimethoate 400 EC	6 HR	24 HR	150	149	99	99
C = Untreated Control	24 HR	24 HR	150	6	4	-
T= Dimethoate 400 EC	24 HR	24 HR	150	29	19	16
[REDACTED] Application						
C = Untreated Control	6 HR	24 HR	150	4	3	-
T= Dimethoate 400 EC	6 HR	24 HR	150	56	37	36
C = Untreated Control	24 HR	24 HR	150	7	5	-
T= Dimethoate 400 EC	24 HR	24 HR	150	31	21	16

^A T= Dimethoate 400 EC = 560.4 g a.i./ha = 0.5 lb a.i./Ac (target rate)^B Corrected Mortality= (% T - % C)/(100 - % C) x 100

FIGURE 1: Typical Treatment Plot Layout**NOTES:**

- Treatment plot was 34 ft (10.4 m) from the untreated control plot.
- For 1 hr residual, 3 randomly selected subplots were sampled and foliage (leaves and stems) combined into one bulk sample. This foliage sample was used for analytical testing.
- For each residual timing after 1 hr residual (i.e., 3 hr, 6 hr, 24 hr), 3 randomly selected subplots were sampled and combined into one bulk sample. This foliage sample was used to set up the bioassay and supply material for analytical testing.

APPENDIX A: Daily Field Conditions from Application to Last Foliage Sampling*Weather Station ID: NC-WST-1 ([REDACTED])**Coordinates: 36.23022 N, 79.23882 W; Elevation: 226 m*

Date	Air Temp (@ 6 ft) [°F]			Humidity (@ 6 ft) [%]			Wind Speed [m/s]			Precip- itation [mm]
	Avg	Min	Max	Avg	Min	Max	Avg	Min	Max	Total
June 9, 2021	76	70	89	89	64	100	1.8	0	5.5	0
June 10, 2021	75	69	84	94	72	100	2.0	0	5.7	0.26
September 16, 2021	74	67	86	87	62	100	1.1	0	4.7	0
September 17, 2021	75	69	87	86	58	100	1.1	0	5.4	0

Note that data shown covers period from application on 09 Jun 2021 to last sampling on 10 Jun 2021, and from application on 16 Sep 2021 to last sampling on 17 Sep 2021.

APPENDIX B: Detailed Bee Testing Conditions

Bioassay	Date & Time Interval ^A	Test Chamber (Data Logger)	Temp (°F)			RH (%)		
			MIN	MAX	AVG	MIN	MAX	AVG
6HAA [REDACTED]	09 Jun (17:17) to 10 Jun (20:21) 2021	NC-CCR-2 (NC-WST-36)	86.5	87.4	87.1	42	53	51
24HAA [REDACTED]	10 Jun (11:22) to 11 Jun (11:22) 2021		86.4	87.4	86.8	36	53	45
6HAA [REDACTED]	08 Jun (17:49) to 09 Jun (17:47) 2021	NC-CCR-2 (NC-WST-36)	86.2	87.5	87.1	42	53	51
24HAA [REDACTED]	09 Jun (12:50) to 10 Jun (12:37) 2021		86.5	87.5	87.1	42	53	51
6HAA [REDACTED]	14 Sep (16:09) to 15 Sep (16:01) 2021	NC-CCR-6 (NC-WST-5)	79.9	82.7	80.9	70	77	75
24HAA [REDACTED]	15 Sep (10:24) to 16 Sep (10:23) 2021		79.5	81.6	80.4	70	76	74
6HAA [REDACTED]	16 Sep (17:32) to 17 Sep (17:35) 2021	NC-CCR-6 (NC-WST-5)	79.1	80.3	79.5	64	77	73
24HAA [REDACTED]	17 Sep (11:32) to 18 Sep (11:23) 2021		78.7	80.4	79.2	64	72	69

^A This date and time interval is from start of exposure to last assessment.

APPENDIX C: Application Calculations

APPENDIX C.1: Calculations for Spray Mix Preparation

STEP 1: What is the spray coverage?

$$\text{Spray Coverage} = \boxed{200 \text{ L mix / ha}}$$

STEP 2: What is your target rate?

$$T = [0.5 \text{ lb a.i./Ac}] \times [453.592 \text{ g a.i. / 1 lb a.i.}] = 226.796 \text{ g a.i./Ac}$$

$$T = [226.796 \text{ g a.i./Ac}] \times [1 \text{ Ac / 0.404686 ha}] = 560.425 \text{ g a.i./ha}$$

Product nominal purity = 43.5 %

$$T = [560.425 \text{ g a.i./ha}] / [0.435] = \boxed{1288.3 \text{ g Prod/ha}}$$

STEP 3: Mixing test item product into water

For 1 ha, mix 1288.3 g Prod into 200 L water OR $[1288.3 \text{ g Prod / 200 L water}] / [1 \text{ ha}]$

STEP 4: Application calculations for spray area

$$\text{Plot} = 3 \text{ m} \times 12 \text{ m} = 36 \text{ m}^2$$

$$[36 \text{ m}^2] \times [1 \text{ ha / 10,000 m}^2] = 0.0036 \text{ ha}$$

How much product needed for 0.0036 ha?

$$[1288.3 \text{ g Prod / 1 ha}] = [X \text{ g Prod / 0.0036 ha}]$$

$$X = [(1288.3 \text{ g Prod}) \times (0.0036 \text{ ha})] / [1] = \boxed{4.638 \text{ g Prod}}$$

How much water needed for mix?

$$[1288.3 \text{ g Prod / 200 L water}] = [4.638 \text{ g Prod / Y Liter water}]$$

$$Y = [(200 \text{ L water}) \times (4.638 \text{ g Prod})] / [1288.3 \text{ g Prod}] = 0.72 \text{ Liter water}$$

$$Y = \boxed{720 \text{ mL water}} \text{ added to } 4.638 \text{ g Prod}$$

STEP 5: Calculating for 2000 mL for extra spray material

$$[4.638 \text{ g Prod}] / [720 \text{ mL water}] = [X \text{ g Prod}] / [2000 \text{ mL water}]$$

$$X = [(4.638 \text{ g Prod}) \times (2000 \text{ mL water})] / [720 \text{ mL water}]$$

$$X = \boxed{12.88 \text{ g Prod added to 2000 mL of water}}$$

** Note that only 720 mL of this 2000 mL mix will be sprayed into the treatment plot to hit the target rate. The purpose of the overage is for tank mix sampling, priming the boom, and maintaining tank pressure.

APPENDIX C.2: Calculation for Target Pass Time**June Trial:****Given:**Total output volume desired = 720 mL = **OV**Boom Output Rate = 75.34 mL/sec = **OR**
(from calibrations)Total Target Spray Duration (SD) = [OV] / [OR] = 9.56 sec = **SD**

Note that this calculation is for a single pass.

Since the boom has coverage of 3 m (10 ft) and the plot was the same width, one pass was performed east to west within the treatment (T) plot. The actual pass time for the T Plot was 9.46 seconds.

Application on the control (C) plot was performed in the same manner except only water was applied. A separate spray tank was used to hold the water for the control application and water control application was performed before performing the treated application. The actual pass time for the control plot was 9.81 seconds.

September Trial:**Given:**Total output volume desired = 720 mL = **OV**Boom Output Rate = 50.12 mL/sec = **OR**
(from calibrations)Total Target Spray Duration (SD) = [OV] / [OR] = 14.37 sec = **SD**

Note that this calculation is for a single pass.

Since the boom has coverage of 3 m (10 ft) and the plot was the same width, one pass was performed east to west within the treatment (T) plot. The actual pass time for the T Plot was 14.36 seconds.

Application on the control (C) plot was performed in the same manner except only water was applied. A separate spray tank was used to hold the water for the control application and water control application was performed before performing the treated application. The actual pass time for the control plot was 14.28 seconds.

Appendix D

Dimethoate Analysis Report for Spray Tank Solutions, Spray Cards, and Treated Alfalfa



ANALYTICAL REPORT

STUDY TITLE

Determination of Dimethoate Dislodgeable Residues in Alfalfa Foliage,
Application Spray Cards, and Tank Mix Samples

DATA REQUIREMENT

U.S. EPA Ecological Effects Test Guidelines
OCSPP 850.3030 Honey Bee Toxicity of Residues on Foliage

AUTHOR

Timothy D. Ballard, M.S.

STUDY INITIATION DATE

August 11, 2021

ANALYTICAL COMPLETION DATE

March 3, 2022

STUDY SPONSOR

Pollinator Research Task Force, LLC
Corteva Agriscience
9330 Zionsville Road
Indianapolis, IN 46268
Phone: (833) 267-8382

SPONSOR REPRESENTATIVE

Joseph Wisk
BASF Corporation
26 Davis Drive
Research Triangle Park, NC 27709-3528
Phone: (919) 547-2619

**STUDY DIRECTOR
AND TESTING FACILITY**

Timothy D. Ballard, M.S.
EN-CAS Analytical Laboratories
2359 Farrington Point Drive
Winston-Salem, NC 27107
Phone: (336) 785-3252

STUDY ID

EN-CAS Project #: 21-0001


Total Number of Pages = 41

CERTIFICATION

We, the undersigned, declare that the work described in this report was performed under our supervision, and that this report provides an accurate record of the procedures and results.

EN-CAS Laboratories **Certification:**

Study Director:

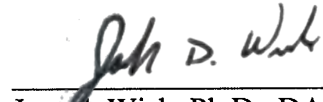


Timothy D. Ballard, M.S.
Laboratory Manager

3-Mar-2022
Date

Pollinator Research Task Force, LLC **Certification:**

Approved by:



Joseph Wisk, Ph.D., DABT
BASF
Study Representative

March 7, 2022
Date

TABLE OF CONTENTS

<u>SUBJECT</u>	<u>PAGE</u>
TITLE PAGE	1
CERTIFICATION	2
TABLE OF CONTENTS	3
1.0 INTRODUCTION	5
2.0 SUMMARY OF METHOD	5
3.0 CALCULATIONS	7
4.0 RESULTS AND DISCUSSION	8
5.0 REFERENCES	8

TABLES

TABLE 1	Determination of Dimethoate Procedural Controls and Recoveries in Alfalfa Dislodgeable Foliar Samples.....	9
TABLE 2	Determination of Dimethoate in Treated Alfalfa Dislodgeable Foliar Samples	10
TABLE 3	Determination of Dimethoate Procedural Controls and Recoveries in Spray Card Samples.....	11
TABLE 4	Determination of Dimethoate in Treated Spray Card Samples	12
TABLE 5	Determination of Dimethoate in Treated Tank Mix Samples	13
TABLE 6	Determination of Dimethoate in Dimethoate Formulation Assay	14

EXAMPLE CHROMATOGRAPHY

FIGURE 1	Typical Chromatogram: 0.0125 µL/mL Standard.....	15
FIGURE 2	Typical Chromatogram: Alfalfa Foliage Blank.....	16
FIGURE 3	Typical Chromatogram: Alfalfa Foliage Control.....	17
FIGURE 4	Typical Chromatogram: Alfalfa Foliage Spike	18
FIGURE 5	Typical Chromatogram: Alfalfa Foliage Sample	19
FIGURE 6	Typical Calibration Curve: Alfalfa Foliage.....	20
FIGURE 7	Typical Chromatogram: 1.0 µL/mL Standard.....	21
FIGURE 8	Typical Chromatogram: Spray Card Blank.....	22
FIGURE 9	Typical Chromatogram: Spray Card Control	23
FIGURE 10	Typical Chromatogram: Spray Card Spike	24
FIGURE 11	Typical Chromatogram: Spray Card Sample	25
FIGURE 12	Typical Calibration Curve: Spray Card.....	26

FIGURE 13	Typical Chromatogram: 150 µL/mL Standard.....	27
------------------	---	----

TABLE OF CONTENTS (continued)

<u>SUBJECT</u>	<u>PAGE</u>
FIGURE 14	Typical Chromatogram: Tank Mix Blank 28
FIGURE 15	Typical Chromatogram: Tank Mix Control 29
FIGURE 16	Typical Chromatogram: Tank Mix Sample..... 30
APPENDIX	31
1-01-AN	32
1-02-AN	33
1-03-AN	34
1-04-AN	35
1-05-AN	36
2-01-AN	37
2-02-AN	38
3-01-AN	40
3-02-AN	41

1.0 INTRODUCTION

Dislodgeable residues of dimethoate were determined in alfalfa foliage and sample spray cards. The dimethoate concentration was determined in samples of tank mix solution.

A dimethoate alfalfa control foliage sample was received at EN-CAS on May 14, 2021 and was logged in on May 26, 2021. The remaining dimethoate alfalfa foliage samples were received on June 23, 2021, October 21, 2021 and November 3, 2021, and were logged in on June 30, 2021, November 12, 2021 and November 15, 2021, respectively. Dimethoate spray card samples were received at EN-CAS on June 22, 2021, October 21, 2021 and November 3, 2021 and were logged in on June 28, 2021, November 12, 2021 and November 15, 2021, respectively. Dimethoate tank mix samples were received at EN-CAS on June 23, 2021, October 21, 2021 and November 3, 2021, and were logged in on June 30, 2021, November 12, 2021 and November 15, 2021, respectively. The dimethoate formulation sample was received on October 22, 2021 and was logged in on November 9, 2021. The samples were assigned unique EN-CAS identification numbers.

2.0 SUMMARY OF METHOD

Foliar dislodgeable residues of dimethoate were extracted by shaking ca. 15 g with a 200-mL 0.004% dioctyl sodium sulfosuccinate (DSS) solution and cleaned up by partitioning a 25-mL aliquot with 50:50 methylene chloride (DCM):chloroform in the presence of saturated sodium chloride solution. After drying with sodium sulfate, the combined DCM:chloroform fractions were concentrated to incipient dryness by rotary evaporation. The samples were reconstituted with 10 mL of acetonitrile containing 0.1% polyethylene glycol (PEG) and then sonicated. Gas chromatographic (GC) standards were also prepared in the acetonitrile (with PEG) solution in order to normalize GC column and detector non-linearity effects that were often present in organo-phosphate quantitation.¹

Residues of dimethoate on spray card samples were extracted with 100 mL of acetone.² GC calibration standards were prepared in acetone. The same GC conditions were used to determine the amount of dimethoate on each spray card.

GC Conditions

Instrument:	HP/Agilent 6890
Phase:	DB-5MS (# 474)
Diameter:	30 m
Length:	0.53 mm
Film Thickness:	1.5 µm
Gases:	Carrier: Air = 100 mL/min.

Detector: Helium = 70 mL/min.
Makeup = 20 mL/min.

Volume Injection: 1.0 µL

Detector: Flame Photometric

Temperatures: Detector: 200°C
Injector Oven: 250°C

Temperature
Program: Initial Temperature: 160°C
Initial Time: 1.0 min.
Ramp: 15°C/min.
Temperature: 235°C
Hold Time: 5.0 min.
Ramp: 35°C/min.
Temperature: 250°C
Final Hold Time: 5.0 min.

Retention
Time: 7.25 min.

Run
Time: 15 min.

The concentration of dimethoate in tank mix samples was determined by HPLC/UV.³
Samples were extracted with mobile phase prior to analysis.

HPLC Conditions

Phase: Zorbax RX-C18 (# 109)
Diameter: 2.1 mm
Length: 150 mm
Particle Size: 5.0 µm

Mobile Phase: Acetonitrile:Water:Acetic Acid.

Pump: Waters Alliance 2695 at a flowrate of 0.300 mL/min., Isocratic
(600:400:1)

Autoinjector: Waters Alliance 2695

Detector: Waters 2996 PDA Detection at 210 nm

Controller: Waters 2695

Data Acquisition

System: Empower2

Volume Injection: 10 µL

Column Oven: Shimadzu CTO-10AS
Temperature = 30°C

Retention

Time: 1.6 min.

Run

Time: 8.0 min.

3.0 CALCULATIONS

3.1 Calculations for Dimethoate Alfalfa Plant Samples

$$\mu\text{g/mL found} = \frac{\text{Sample Response} - \text{Intercept}}{\text{Slope}}$$

$$\mu\text{g/mL corrected} = \mu\text{g/mL found} \times \text{Dilution Factor}$$

$$\mu\text{g sample} = \frac{\mu\text{g/mL corrected} \times 100 \text{ mL}}{\frac{1}{8} \text{ total sample extracted}}$$

$$\frac{w}{w} \text{ ppm} = \frac{\mu\text{g sample}}{\text{grams sample}}$$

$$\text{Recovery \%} = \frac{\text{ppm found}}{\text{ppm Spike added}} \times 100\%$$

3.2 Calculations for Dimethoate Treatment Samples

$$\mu\text{g/mL found} = \frac{\text{Sample Response}}{\text{Average standard response}} \times \text{standard } \mu\text{g/mL}$$

$$\mu\text{g/mL Solution} = \mu\text{g/mL found} * \text{Dilution Factor}$$

$$\text{Original Conc, } \mu\text{g} = \mu\text{g/mL Solution} \times 100 \text{ mL extraction volume}$$

$$\text{Found \% ppm} = \frac{\text{Original Conc, } \mu\text{g}}{\text{Sample weight, g}} \times 100\%$$

3.3 Calculations for Dimethoate Card Solutions

$$\mu\text{g/mL found} = \frac{\text{Sample Response} - \text{Intercept}}{\text{Slope}}$$

$$\mu\text{g card} = \mu\text{g/mL found} \times \text{Extraction Volume}$$

$$\text{Recovery \%} = \frac{\mu\text{g card}}{\mu\text{g Spike added}} \times 100\%$$

4.0 RESULTS AND DISCUSSION

The concentration of dimethoate dislodgeable foliar residues found on alfalfa samples ranged between 0 and 32.8 ppm, see DFR Table 2. QC recoveries from the method ranged from 77-94%, see Table 1. Dimethoate found on spray cards ranged from 257 to 863 μg , see Table 4. QC recoveries from the method ranged from 100-108%, see Table 3. Dimethoate tank mix sample average concentrations ranged from 1958 to 2575 $\mu\text{g/mL}$, see Table 5. Dimethoate average formulation assay was 37.5%, see Table 6.

5.0 REFERENCES

1. EN-CAS Analytical Method No. ENC-5/97, entitled Analytical Method for the Determination of Dimethoate and Omethoate in Dioctyl Sodium Sulfosuccinate (DSS) Solution, issued February 27, 1998.
2. CIPAC Handbook E, p. 154-157, Dimethoate Technical Gas Chromatographic Method.
3. CIPAC Handbook E, p. 69-70, Dimethoate Technical High Performance Liquid Chromatography Method.

Table 1

Determination of Dimethoate Procedural Controls and Recoveries
in Alfalfa Dislodgeable Foliar Samples

EN-CAS ID	Study ID	Set #	Fortification Level (ppm)	Fortification / Extraction Date	% Recovery
EU13648-C1	SM.UTC.0hr.FA	1-01-AN	--	9/15/21	--
EU13648-S1	SM.UTC.0hr.FA	1-01-AN	2.6	9/15/21	94
EU13671-C1	S21-04089-L1-C-LEAF-1HAA-A	1-02-AN	--	9/23/21	--
EU13671-S1	S21-04089-L1-C-LEAF-1HAA-A	1-02-AN	5.2	9/23/21	89
EU13626-C2	Control	1-03-AN	--	9/28/21	--
EU13626-S4	Control + Spike	1-03-AN	5.2	9/28/21	89
EU13626-C3	Control	1-04-AN	--	11/23/21	--
EU13626-S5	Control + Spike	1-04-AN	2.6	11/23/21	85
EU13626-S6	Control + Spike	1-04-AN	5.2	11/23/21	94
EU13737-C1	SM2.UTC.0HR.FA	1-05-AN	--	11/29/21	--
EU13737-S1	SM2.UTC.0HR.FA	1-05-AN	5.2	11/29/21	77

Table 2

Determination of Dimethoate in Treated Alfalfa Dislodgeable Foliar Samples

EN-CAS Sample ID	Study ID	Set #	w/w ppm Found
EU13648-C1	SM.UTC.0hr.FA	1-01-AN	0.0
EU13650	SM.TRT.0hr.FA	1-01-AN	22.1
EU13652	SM.UTC.6hr.FA	1-01-AN	0.0
EU13654	SM.TRT.6hr.FA	1-01-AN	32.8
EU13656	SM.UTC.24hr.FA	1-01-AN	0.0
EU13658	SM.TRT.24hr.FA	1-01-AN	21.6
EU13671-C1	S21-04089-L1-C-LEAF-1HAA-A	1-02-AN	0.0
EU13670	S21-04089-L1-C-LEAF-6HAA-A	1-02-AN	0.0
EU13672	S21-04089-L1-C-LEAF-24HAA-A	1-02-AN	0.0
EU13664	S21-04089-L1-T-LEAF-1HAA-A	1-02-AN	10.6
EU13665	S21-04089-L1-T-LEAF-6HAA-A	1-02-AN	8.0
EU13666	S21-04089-L1-T-LEAF-24HAA-A	1-02-AN	2.3
EU13655	SM.TRT.6hr.FB	1-02-AN	7.8
EU13651	SM.TRT.0hr.FB	1-03-AN	14.5
EU13659	SM.TRT.24hr.FB	1-03-AN	7.3
EU13780	S21-04089-L2-T-LEAF-1HAA-A	1-04-AN	19.5
EU13781	S21-04089-L2-T-LEAF-6HAA-A	1-04-AN	19.4
EU13782	S21-04089-L2-T-LEAF-24HAA-A	1-04-AN	12.3
EU13737-C1	SM2.UTC.0hr.FA	1-05-AN	0.0
EU13741	SM2.UTC.6hr.FA	1-05-AN	0.0
EU13745	SM2. UTC.24hr.FA	1-05-AN	0.0
EU13749	SM2. UTC.48hr.FA	1-05-AN	0.0
EU13739	SM2.TRT.0hr.FA	1-05-AN	15.2
EU13743	SM2.TRT.6hr.FA	1-05-AN	6.4
EU13747	SM2.TRT.24hr.FA	1-05-AN	18.7
EU13751	SM2.TRT.48hr.FA	1-05-AN	5.2

Table 3

Determination of Dimethoate Procedural Controls and Recoveries
in Spray Card Samples

EN-CAS ID	Study ID	Set #	Fort. Level (µg)	Fort. / Extract. Date	% Recovery
EU13676	S21-04089-L1-C-DISC-0HBA-Rep1	3-01-AN	--	10/8/21	--
EU13677	S21-04089-L1-C-DISC-0HBA-Rep2	3-01-AN	249	10/8/21	101
EU13678	S21-04089-L1-C-DISC-0HBA-Rep3	3-01-AN	499	10/8/21	102
EU13770	S21-04089-L2-C-DISC-0HBA-Rep1	3-02-AN	--	10/8/21	--
EU13771	S21-04089-L2-C-DISC-0HBA-Rep2	3-02-AN	249	10/8/21	108
EU13772	S21-04089-L2-C-DISC-0HBA-Rep3	3-02-AN	499	10/8/21	100

Fort. = Fortification

Extract. = Extraction

Table 4

Determination of Dimethoate in Treated Spray Card Samples

EN-CAS Sample ID	Study ID	Set #	µg/Card Found
EU13676	S21-04089-L1-C-DISC-0HBA-Rep1	3-01-AN	0.0
EU13645	SM.TRT.SC1	3-01-AN	422
EU13646	SM.TRT.SC2	3-01-AN	863
EU13647	SM.TRT.SC3	3-01-AN	440
EU13661	S21-04089-LT-T-DISC-0HAA-Rep1	3-01-AN	275
EU13662	S21-04089-LT-T-DISC-0HAA-Rep2	3-01-AN	257
EU13663	S21-04089-LT-T-DISC-0HAA-Rep3	3-01-AN	293
EU13770	S21-04089-L2-C-DISC-0HBA-Rep1	3-02-AN	0.0
EU13777	S21-04089-L2-T-DISC-0HAA-Rep1	3-02-AN	383
EU13778	S21-04089-L2-T-DISC-0HAA-Rep2	3-02-AN	379
EU13779	S21-04089-L2-T-DISC-0HAA-Rep3	3-02-AN	584
EU13734	SM2.TRT.SC1	3-02-AN	347
EU13735	SM2.TRT.SC2	3-02-AN	510
EU13736	SM2.TRT.SC3	3-02-AN	537

Table 5

Determination of Dimethoate in Tank Mix Samples

EN-CAS Sample ID	Study ID	Set #	µg/mL Found	Average µg/mL Found
EU13679-A	S21-04089-L1-C-PRE-0HBA-A	2-01-AN	0.0	
EU13679-B	S21-04089-L1-C-PRE-0HBA-A	2-01-AN	0.0	0.0
EU13681-A	S21-04089-L1-C-PRE-0HBA-A	2-01-AN	0.0	
EU13681-B	S21-04089-L1-C-PRE-0HBA-A	2-01-AN	0.0	0.0
EU13683-A	S21-04089-L1-C-PRE-0HBA-A	2-01-AN	2465	
EU13683-B	S21-04089-L1-C-PRE-0HBA-A	2-01-AN	2512	2488
EU13685-A	S21-04089-L1-C-PRE-0HBA-A	2-01-AN	2550	
EU13685-B	S21-04089-L1-C-PRE-0HBA-A	2-01-AN	2574	2562
EU13640-A	SM.TRT.TMA	2-01-AN	2251	
EU13640-B	SM.TRT.TMA	2-01-AN	2277	2264
EU13727-A	SM2.UTC.TMA	2-02-AN	0.0	
EU13727-B	SM2.UTC.TMA	2-02-AN	0.0	0.0
EU13769-A	S21-04089-L2-CT-Tank-0HBA-A	2-02-AN	0.0	
EU13769-B	S21-04089-L2-CT-Tank-0HBA-A	2-02-AN	0.0	0.0
EU13729-A	SM2.TRT.TMA	2-02-AN	1962	
EU13729-B	SM2.TRT.TMA	2-02-AN	1954	1958
EU13776-A	S21-04089-L2-T-Tank-0HBA-A	2-02-AN	2578	
EU13776-B	S21-04089-L2-T-Tank-0HBA-A	2-02-AN	2572	2575

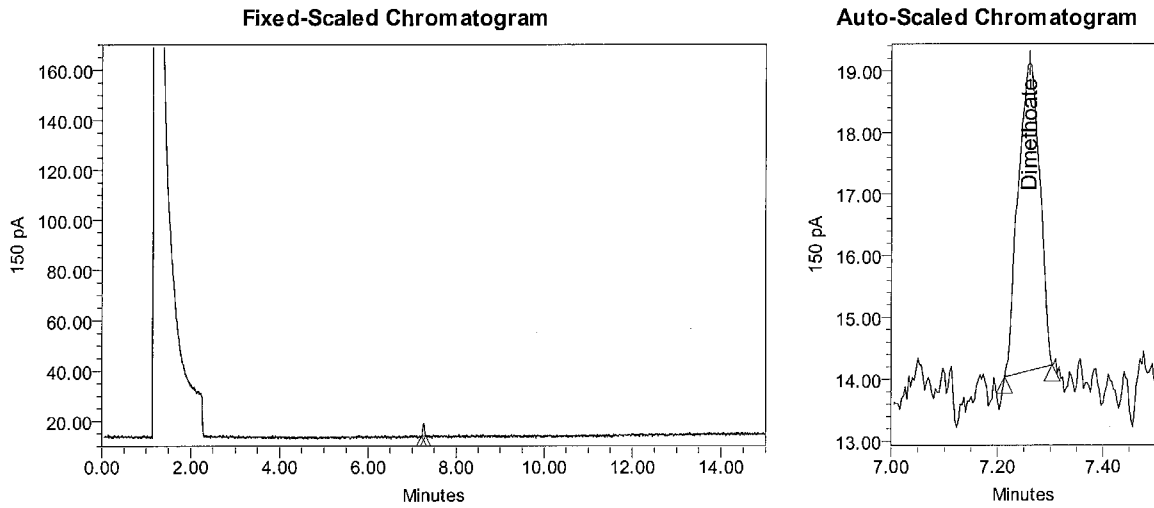
Table 6

Determination of Dimethoate in Dimethoate Formulation Assay

EN-CAS Sample ID	Study ID	Set #	% Purity	Average % Purity
EU13765-A	Dimethoate aliquot for	2-02-AN	37.2	
EU13765-B	analysis of purity	2-02-AN	37.8	37.5

FIGURE 1

Typical Chromatogram
0.0125 $\mu\text{L/mL}$ Alfalfa Foliage Standard



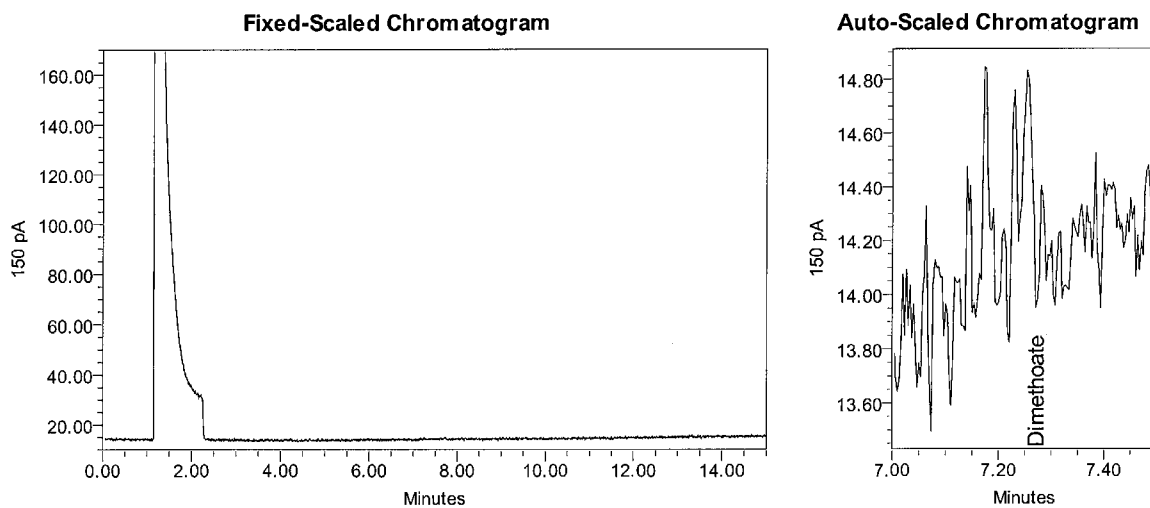
Peak Results

	Name	RT	Area	Baseline Start	Baseline End	Int Type
1	Dimethoate	7.260	14	7.213	7.303	BB

GC/FPD Run # 94651, Set # 1-02-AN, Dated 24 Sep 2021

FIGURE 2

Typical Chromatogram
Alfalfa Foliage Blank



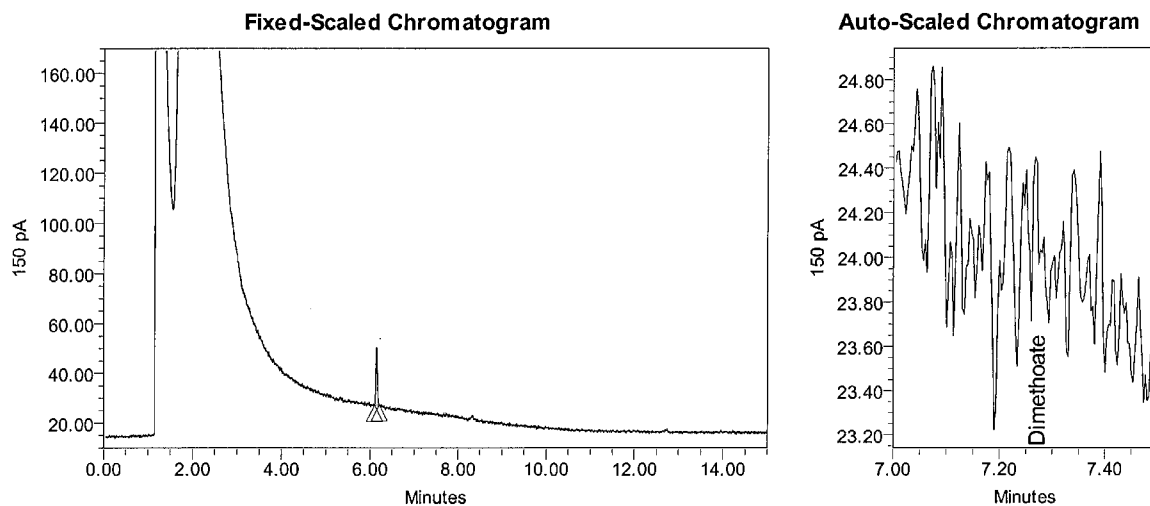
Peak Results

	Name	RT	Area	Baseline Start	Baseline End	Int Type
1	Dimethoate	7.273				Missing

EN-CAS Sample ID #: Blank
Dimethoate w/w ppm: 0.0 w/w ppm
GC/FPD Run # 94651, Set # 1-02-AN, Dated 24 Sep 2021

FIGURE 3

Typical Chromatogram
Alfalfa Foliage Control
Study ID: S21-04089-L1-C-LEAF-1HAA-A



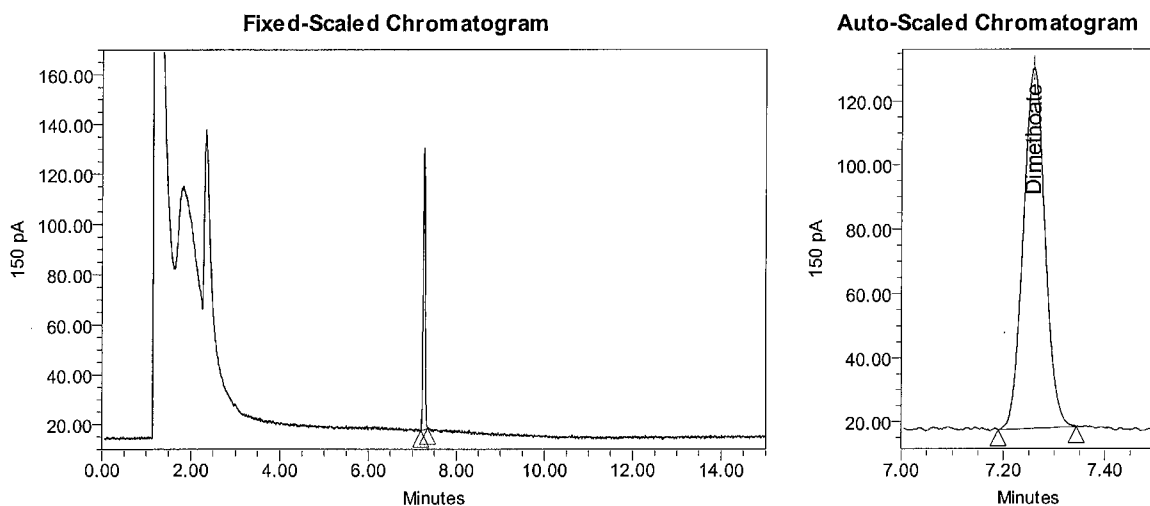
Peak Results

	Name	RT	Area	Baseline Start	Baseline End	Int Type
1	Dimethoate	7.273				Missing

EN-CAS Sample ID #: EU13671-C1
Dimethoate w/w ppm: 0.0 w/w ppm
GC/FPD Run # 94651, Set # 1-02-AN, Dated 24 Sep 2021

FIGURE 4

Typical Chromatogram
Alfalfa Foliage Control + 5.2 ppm Dimethoate
Study ID: S21-04089-L1-C-LEAF-1HAA-A



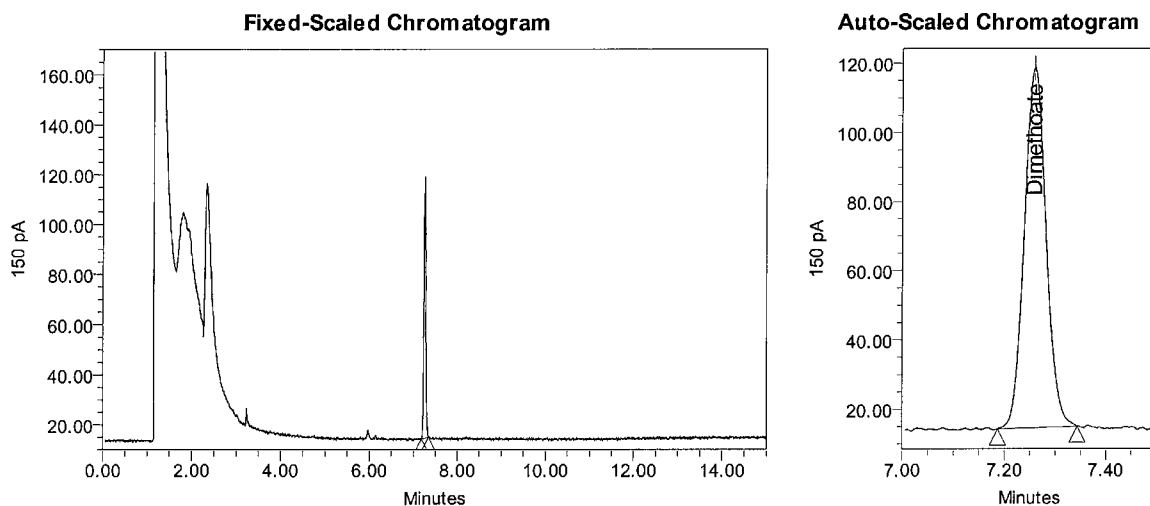
Peak Results

	Name	RT	Area	Baseline Start	Baseline End	Int Type
1	Dimethoate	7.259	333	7.190	7.343	BB

EN-CAS Sample ID #: EU13671-S1
Dimethoate % Recovery: 89%
GC/FPD Run # 94651, Set # 1-02-AN, Dated 24 Sep 2021
1 to 20 Dilution

FIGURE 5

Typical Chromatogram
Alfalfa Foliage Sample
Study ID: S21-04089-L1-T-LEAF-6HAA-A



Peak Results

	Name	RT	Area	Baseline Start	Baseline End	Int Type
1	Dimethoate	7.259	309	7.187	7.343	BB

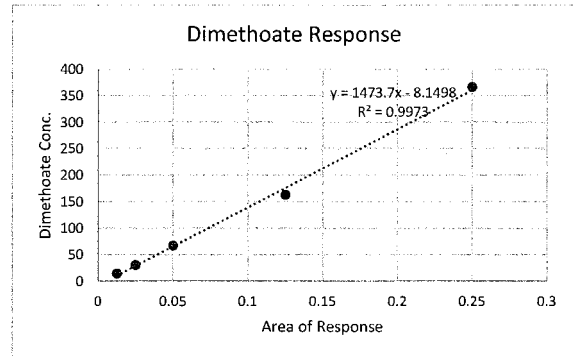
EN-CAS Sample ID #: EU13665
Dimethoate w/w ppm: 8.0 w/w ppm
GC/FPD Run # 94651, Set # 1-02-AN, Dated 24 Sep 2021
1 to 20 Dilution

FIGURE 6

Typical GC Calibration Curve
For Alfalfa Foliage

Standard Curve

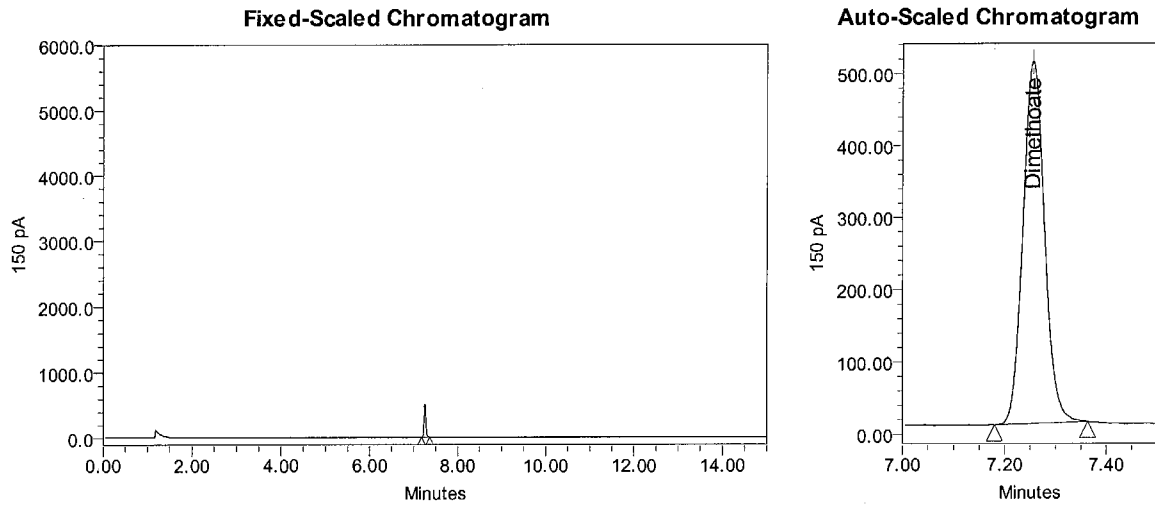
Standard	Conc.	Response
0.0125µg/mL Dimethoate	0.0125	14.1232
0.025µg/mL Dimethoate	0.025	30.4462
0.05µg/mL Dimethoate	0.05	67.1843
0.125µg/mL Dimethoate	0.125	162.9546
0.25µg/mL Dimethoate	0.25	366.1375
Slope		1474
Intercept		-8.15
r^2		0.99730



GC/FPD Run # 94651, Set # 1-02-AN, Dated 24 Sep 2021

FIGURE 7

Typical Chromatogram
1.0 µL/mL Spray Card Standard



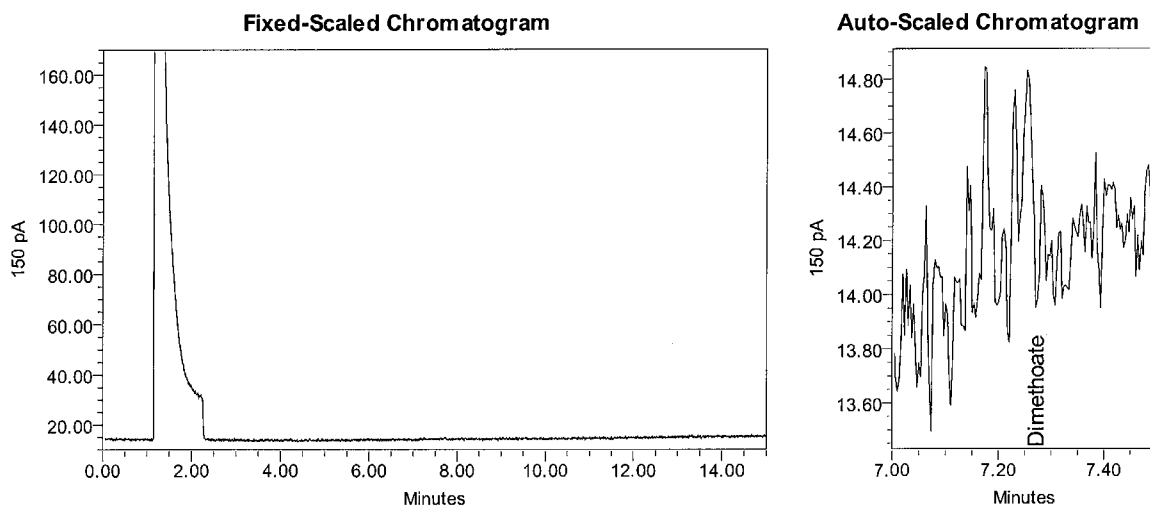
Peak Results

	Name	RT	Area	Baseline Start	Baseline End	Int Type
1	Dimethoate	7.255	1476	7.180	7.363	BB

GC/FPD Run # 94657, Set # 3-01-AN, Dated 08 Oct 2021

FIGURE 8

Typical Chromatogram
Spray Card Blank



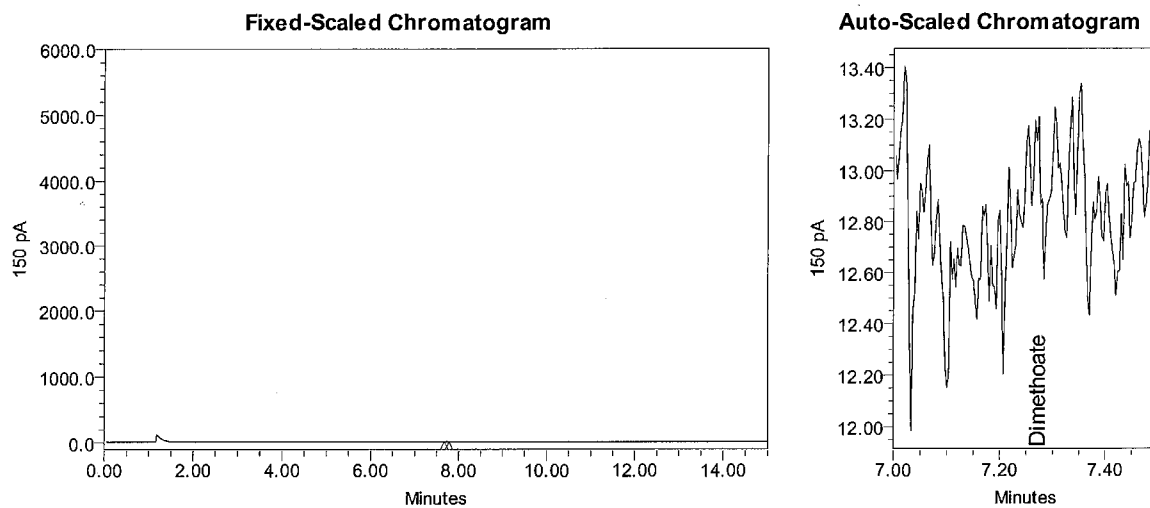
Peak Results

	Name	RT	Area	Baseline Start	Baseline End	Int Type
1	Dimethoate	7.273				Missing

EN-CAS Sample ID #: Blank
Dimethoate $\mu\text{g}/\text{card}$ Found: 0.0 $\mu\text{g}/\text{card}$ Found
GC/FPD Run # 94657, Set # 3-01-AN, Dated 08 Oct 2021

FIGURE 9

Typical Chromatogram
Spray Card Control
Study ID: S21-04089-L1-C-DISC-0HBA-Rep1



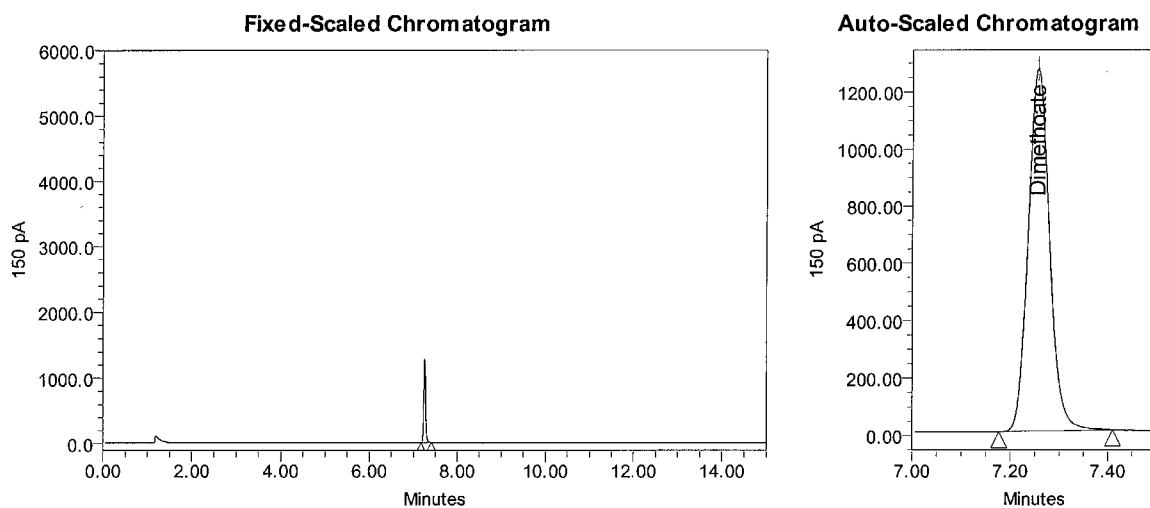
Peak Results

	Name	RT	Area	Baseline Start	Baseline End	Int Type
1	Dimethoate	7.273				Missing

EN-CAS Sample ID #: EU13676
Dimethoate $\mu\text{g}/\text{card}$ Found: 0.0 $\mu\text{g}/\text{card}$ Found
GC/FPD Run # 94657, Set # 3-01-AN, Dated 08 Oct 2021
1 to 100 Dilution

FIGURE 10

Typical Chromatogram
Spray Card Control + 249 µg Dimethoate
Study ID: S21-04089-L1-C-DISC-0HBA-Rep2



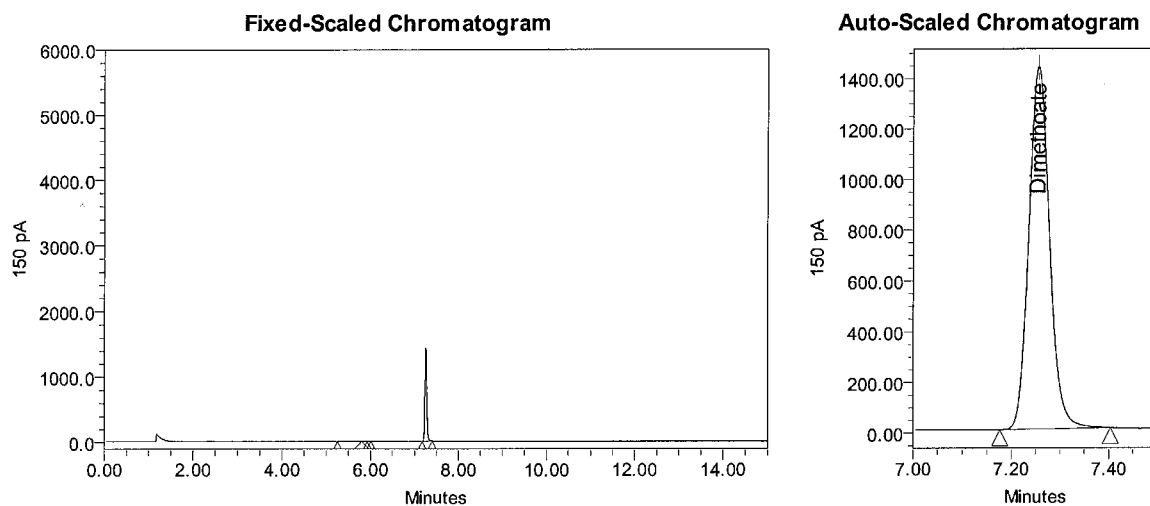
Peak Results

	Name	RT	Area	Baseline Start	Baseline End	Int Type
1	Dimethoate	7.256	3832	7.177	7.410	BB

EN-CAS Sample ID #: EU13677
Dimethoate % Recovery: 101%
GC/FPD Run # 94657, Set # 3-01-AN, Dated 08 Oct 2021
1 to 100 Dilution

FIGURE 11

Typical Chromatogram
Spray Card Sample
Study ID: S21-04089-LT-T-DISC-0HAA-Rep1



Peak Results

	Name	RT	Area	Baseline Start	Baseline End	Int Type
1	Dimethoate	7.255	4212	7.177	7.403	BB

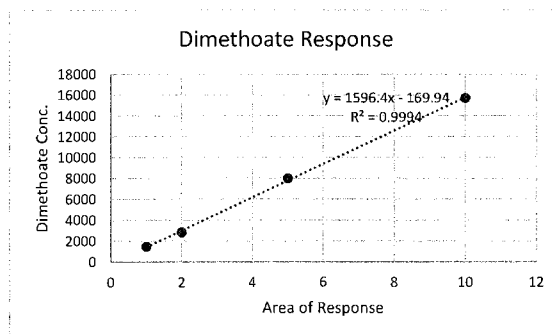
EN-CAS Sample ID #: EU13661
Dimethoate $\mu\text{g}/\text{card}$ Found: 275 $\mu\text{g}/\text{card}$ Found
GC/FPD Run # 94657, Set # 3-01-AN, Dated 08 Oct 2021
1 to 100 Dilution

FIGURE 12

Typical GC Calibration Curve
For Spray Cards

Standard Curve

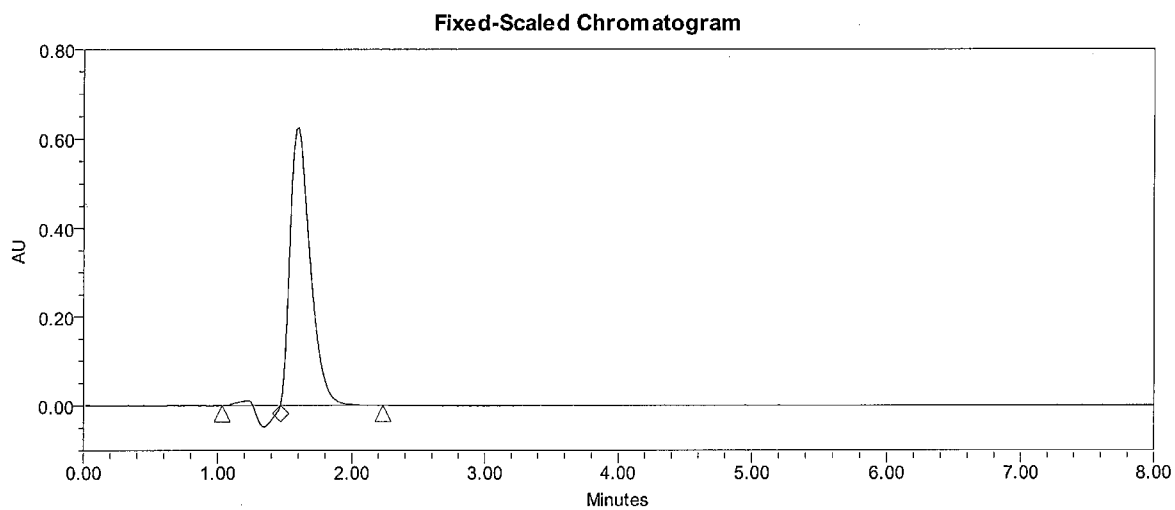
Standard	Conc.	Response
1µg/mL Dimethoate	1	1476
2µg/mL Dimethoate	2	2850
5µg/mL Dimethoate	5	7997
10µg/mL Dimethoate	10	15730
Slope		1596
Intercept		-169.94
r^2		0.99943



GC/FPD Run # 94657, Set # 3-01-AN, Dated 08 Oct 2021

FIGURE 13

Typical Chromatogram
150 µL/mL Tank Mix Standard



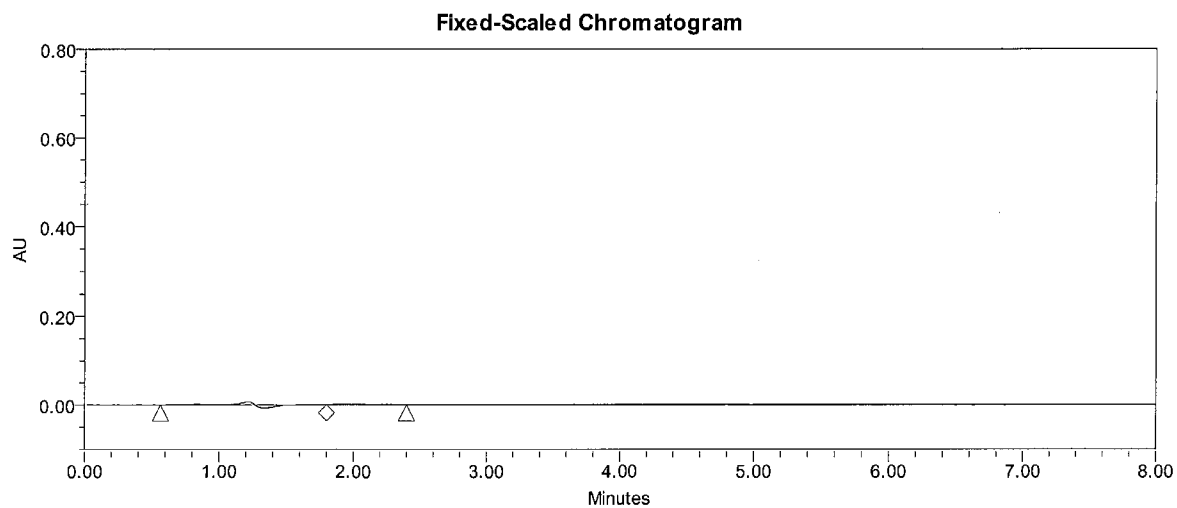
Peak Results

	Name	RT	Area	Baseline Start	Baseline End	Int Type
1	Dimethoate	1.596	6535695	1.033	2.233	vb

HPLC/UV Run # 94655, Set # 2-01-AN, Dated 05 Oct 2021

FIGURE 14

Typical Chromatogram
Tank Mix Blank



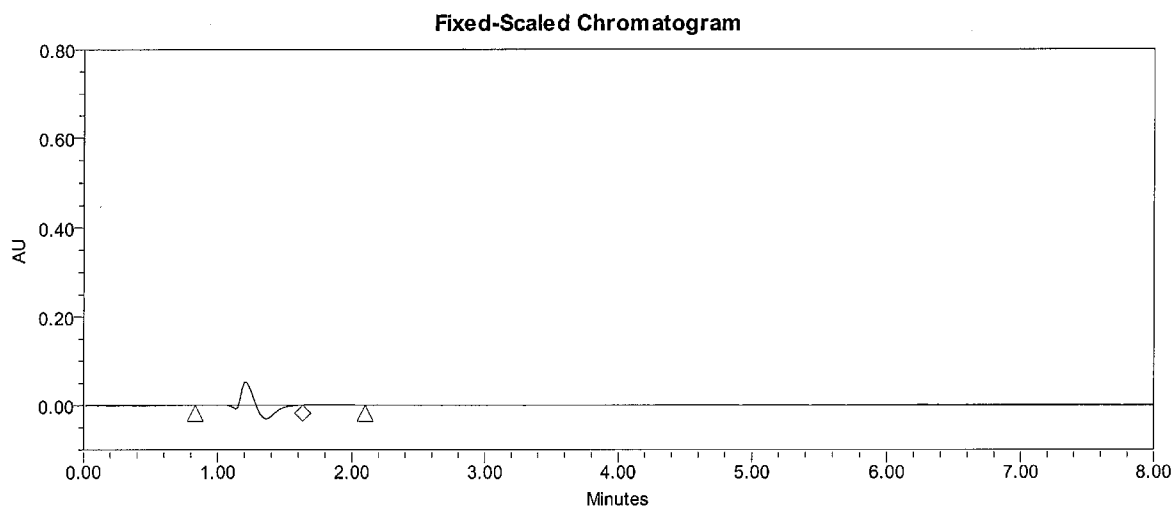
Peak Results

	Name	RT	Area	Baseline Start	Baseline End	Int Type
1	Dimethoate	1.592				Missing

EN-CAS Sample ID #: Blank
Dimethoate $\mu\text{g/mL}$ Found: 0.0 $\mu\text{g/mL}$ Found
HPLC/UV Run # 94655, Set # 2-01-AN, Dated 05 Oct 2021

FIGURE 15

Typical Chromatogram
Tank Mix Control
Study ID: S21-04089-L1-C-PRE-0HAA-A



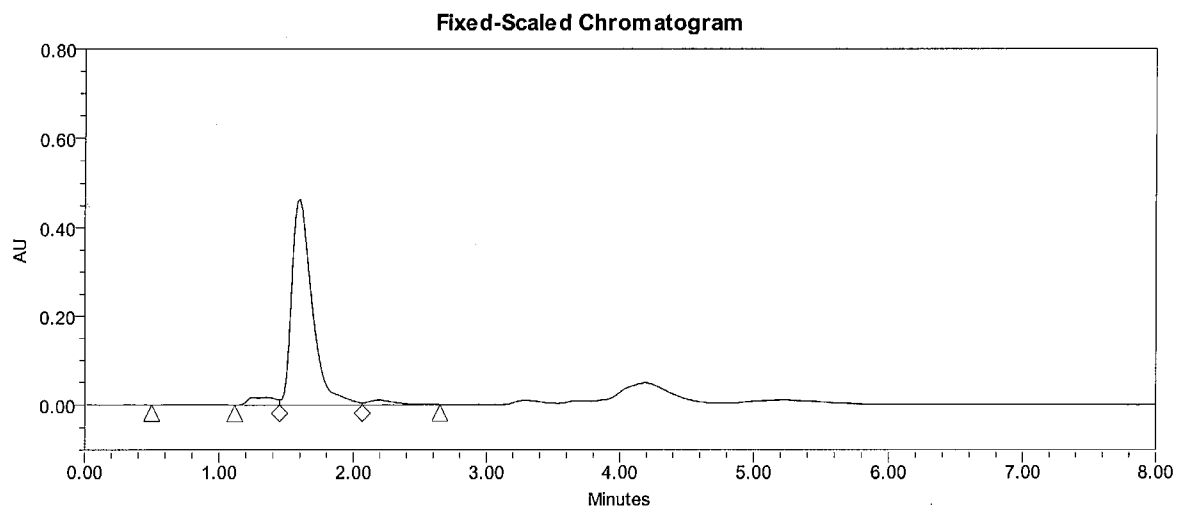
Peak Results

	Name	RT	Area	Baseline Start	Baseline End	Int Type
1	Dimethoate	1.592				Missing

EN-CAS Sample ID #: EU13679
Dimethoate $\mu\text{g/mL}$ Found: 0.0 $\mu\text{g/mL}$ Found
HPLC/UV Run # 94655, Set # 2-01-AN, Dated 05 Oct 2021
1 to 2 Dilution

FIGURE 16

Typical Chromatogram
Tank Mix Sample
Study ID: SM.TRT.TMA



Peak Results

	Name	RT	Area	Baseline Start	Baseline End	Int Type
1	Dimethoate	1.596	4918410	1.117	2.650	Vv

EN-CAS Sample ID #: EU13640
Dimethoate $\mu\text{g/mL}$ Found: 2282 $\mu\text{g/mL}$ Found
HPLC/UV Run # 94655, Set # 2-01-AN, Dated 05 Oct 2021
1 to 20 Dilution

APPENDIX

Calculation Spreadsheets

Dimethoate 1-01-AN

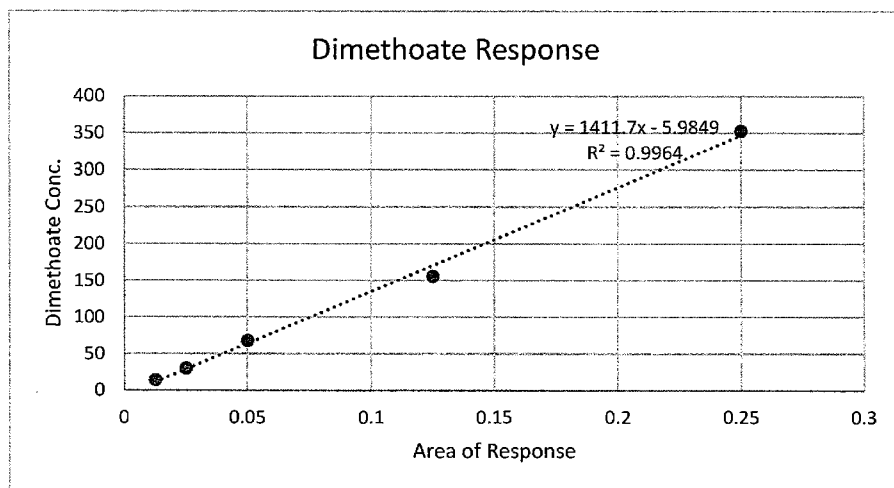
Project:21-0001

MDB 9/16/21

Run# 95500

Standard Curve

Standard	Conc.	Response
0.0125µg/mL Dimethoate	0.0125	14.813
0.025µg/mL Dimethoate	0.025	30.5326
0.05µg/mL Dimethoate	0.05	68.5328
0.125µg/mL Dimethoate	0.125	155.9906
0.25µg/mL Dimethoate	0.25	353.115
Slope		1412
Intercept		-5.98
r ²		0.99644



En-Cas ID	Study ID	g Sample	Response	ug/mL Found	Dil Factor	ug/mL Corrected	ug sample	w/w ppm	Recovery %
EU13648-C1	SM.UTC.0hr.FA	16.16	0.0000	0.0000	1	0.0000	0.0000	0.0	n/a
EU13648-S1	SM.UTC.0hr.FA	15.75	166.1200	0.1219	20	2.4383	195.0620	12.4	94%
EU13650	SM.TRT.0hr.FA	15.62	298.0940	0.2154	20	4.3080	344.6400	22.1	n/a
EU13652	SM.UTC.6hr.FA	14.63	0.0000	0.0000	1	0.0000	0.0000	0.0	n/a
EU13654	SM.TRT.6hr.FA	14.82	208.2649	0.1518	40	6.0707	485.6572	32.8	n/a
EU13656	SM.UTC.24hr.FA	16.74	0.0000	0.0000	1	0.0000	0.0000	0.0	n/a
EU13658	SM.TRT.24hr.FA	15.50	289.7504	0.2095	20	4.1898	335.1835	21.6	n/a

Dimethoate 1-02-AN

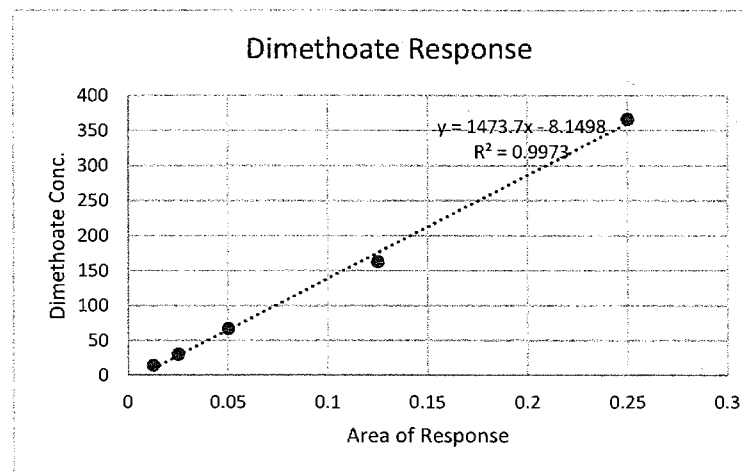
Project:21-0001

MDB 9/24/21

Run# 94651

Standard Curve

Standard	Conc.	Response
0.0125µg/mL Dimethoate	0.0125	14.1232
0.025µg/mL Dimethoate	0.025	30.4462
0.05µg/mL Dimethoate	0.05	67.1843
0.125µg/mL Dimethoate	0.125	162.9546
0.25µg/mL Dimethoate	0.25	366.1375
Slope		1474
Intercept		-8.15
r ²		0.99730



En-Cas ID	Study ID	g Sample	Response	ug/mL Found	Dil Factor	ug/mL Corr.	ug sample	w/w ppm	Recovery %
EU13671-C1	S21-04089-L1-C-LEAF-1HAA-A	16.20	0.0000	0.0000	1	0.0000	0.0000	0.0	n/a
EU13671-S1	S21-04089-L1-C-LEAF-1HAA-A	15.81	333.0287	0.2315	20	4.6302	370.4138	23.4	89%
EU13670	S21-04089-L1-C-LEAF-6HAA-A	15.11	0.0000	0.0000	1	0.0000	0.0000	0.0	n/a
EU13672	S21-04089-L1-C-LEAF-24HAA-A	15.61	0.0000	0.0000	1	0.0000	0.0000	0.0	n/a
EU13664	S21-04089-L1-T-LEAF-1HAA-A	14.97	138.6370	0.0996	20	1.9921	159.3648	10.6	n/a
EU13665	S21-04089-L1-T-LEAF-6HAA-A	15.27	104.2673	0.0763	20	1.5256	122.0500	8.0	n/a
EU13666	S21-04089-L1-T-LEAF-24HAA-A	15.18	23.8669	0.0217	20	0.4345	34.7602	2.3	n/a
EU13655	SM.TRT.6hr.FB	14.76	97.4912	0.0717	20	1.4337	114.6933	7.8	n/a

Dimethoate 1-03-AN

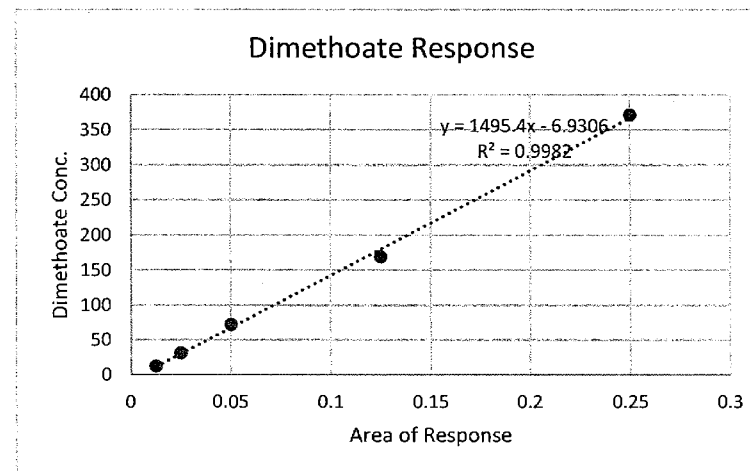
Project:21-0001

MDB 9/29/21

Run# 94652

Standard Curve

Standard	Conc.	Response
0.0125µg/mL Dimethoate	0.0125	12.6596
0.025µg/mL Dimethoate	0.025	31.7386
0.05µg/mL Dimethoate	0.05	72.1238
0.125µg/mL Dimethoate	0.125	169.1192
0.25µg/mL Dimethoate	0.25	371.3232
Slope		1495
Intercept		-6.93
r ²		0.99817



En-Cas ID	Study ID	g Sample	Response	ug/mL Found	Dil Factor	ug/mL Corr.	ug sample	w/w ppm	Recovery %
EU13626-C2	Control	14.48	0.0000	0.0000	1	0.0000	0.0000	0.0	n/a
EU13626-S4	Control+ Spike	14.67	338.7166	0.2311	20	4.6228	369.8272	25.2	89%
EU13651	SM.TRT.0hr.FB	15.97	101.5622	0.0726	40	2.9021	232.1650	14.5	n/a
EU13659	SM.TRT.24hr.FB	15.33	97.4912	0.0698	20	1.3966	111.7267	7.3	n/a

Dimethoate 1-04-AN

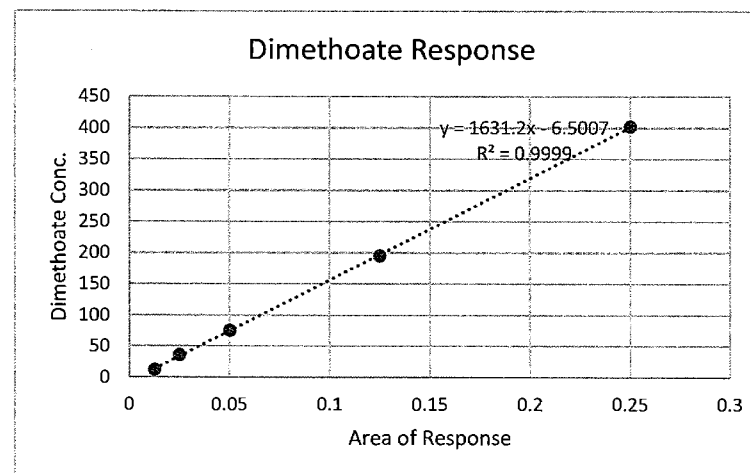
Project:21-0001

MDB 12/1/21

Run# 94664

Standard Curve

Standard	Conc.	Response
0.0125µg/mL Dimethoate	0.0125	12.6004
0.025µg/mL Dimethoate	0.025	36.3227
0.05µg/mL Dimethoate	0.05	75.531
0.125µg/mL Dimethoate	0.125	195.407
0.25µg/mL Dimethoate	0.25	402.0513
Slope		1631
Intercept		-6.50
r ²		0.99990

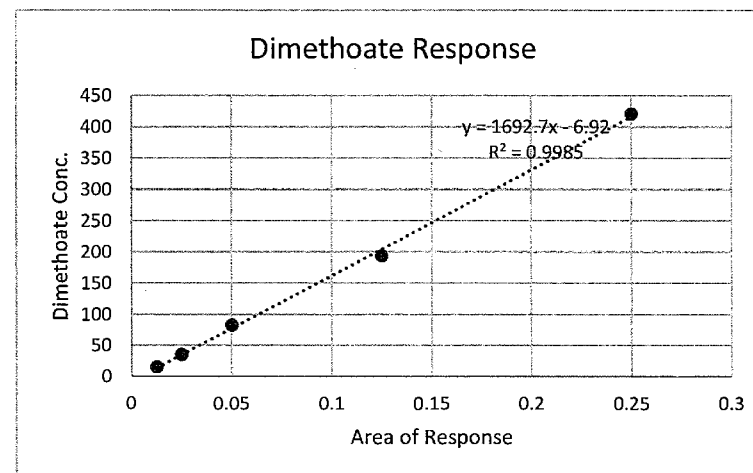


En-Cas ID	Study ID	g Sample	Response	ug/mL Found	Dil Factor	ug/mL Corr.	ug sample	w/w ppm	Recovery %
EU13626-C3	Control	15.20	0.0000	0.0000	1	0.0000	0.0000	0.0	n/a
EU13626-S5	Control +Spike	14.98	173.2860	0.1102	20	2.2044	176.3512	11.8	85%
EU13626-S6	Control +Spike	15.44	389.1166	0.2425	20	4.8507	388.0576	25.1	94%
EU13780	S21-04089-L2-T-LEAF-1HAA-A	15.20	295.4596	0.1851	20	3.7024	296.1902	19.5	n/a
EU13781	S21-04089-L2-T-LEAF-6HAA-A	15.03	290.4533	0.1820	20	3.6410	291.2796	19.4	n/a
EU13782	S21-04089-L2-T-LEAF-24HAA-A	15.07	183.1970	0.1163	20	2.3259	186.0728	12.3	n/a

Dimethoate 1-05-AN
 Project:21-0001
 MDB 12/2/21
 Run# 94665

Standard Curve

Standard	Conc.	Response
0.0125µg/mL Dimethoate	0.0125	15.3278
0.025µg/mL Dimethoate	0.025	35.6292
0.05µg/mL Dimethoate	0.05	82.9895
0.125µg/mL Dimethoate	0.125	193.7542
0.25µg/mL Dimethoate	0.25	420.5936
Slope		1693
Intercept		-6.92
r ²		0.99849



En-Cas ID	Study ID	g Sample	Response	ug/mL Found	Dil Factor	ug/mL Corr.	ug sample	w/w ppm	Recovery %
EU13737-C1	SM2.UTC.0hr.FA	15.39	0.0000	0.0000	1	0.0000	0.0000	0.0	n/a
EU13737-S1	SM2.UTC.0hr.FA	17.34	329.3639	0.1987	20	3.9732	317.8591	18.3	77%
EU13741	SM2.UTC.6hr.FA	15.00	0.0000	0.0000	1	0.0000	0.0000	0.0	n/a
EU13745	SM2.UTC.24hr.FA	17.53	0.0000	0.0000	1	0.0000	0.0000	0.0	n/a
EU13749	SM2.UTC.48hr.FA	16.57	0.0000	0.0000	1	0.0000	0.0000	0.0	n/a
EU13739	SM2.TRT.0hr.FA	15.03	234.2835	0.1425	20	2.8499	227.9881	15.2	n/a
EU13743	SM2.TRT.6hr.FA	17.74	112.6094	0.0706	20	1.4123	112.9805	6.4	n/a
EU13747	SM2.TRT.24hr.FA	15.00	289.4142	0.1751	20	3.5012	280.0982	18.7	n/a
EU13751	SM2.TRT.48hr.FA	16.78	84.8421	0.0542	20	1.0842	86.7345	5.2	n/a

Dimethoate 2-01-AN
 Project:21-0001
 MDB 10/5/21
 Run# 94655

Standard Suitability

Standard	Response
150µg/mL Dimethoate	6535695
150µg/mL Dimethoate	6534303
150µg/mL Dimethoate	6556662
150µg/mL Dimethoate	6497603
150µg/mL Dimethoate	6489675
Average	6522788
Standard Deviation	28184
%CV	0.43209

En-Cas ID	Study ID	Response	ug/mL Found	Dil Factor	Sample ug/mL	Average
EU13679-A	S21-04089-L1-C-PRE-OHBA-A	0	0	2	0	0
EU13679-B	S21-04089-L1-C-PRE-OHBA-A	0	0	2	0	
EU13681-A	S21-04089-L1-C-PRE-OHBA-A	0	0	2	0	0
EU13681-B	S21-04089-L1-C-PRE-OHBA-A	0	0	2	0	
150µg/mL Dimethoate		6512749	149	n/a	n/a	
EU13683-A	S21-04089-L1-T-PRE-OHBA-A	5385833	123	20	2465	2488
EU13683-B	S21-04089-L1-T-PRE-OHBA-A	5488322	126	20	2512	
EU13685-A	S21-04089-L1-T-PRE-OHBA-A	5571772	128	20	2550	2562
EU13685-B	S21-04089-L1-T-PRE-OHBA-A	5623781	129	20	2574	
EU13640-A	SM.TRT.TMA	4918410	113	20	2251	2264
EU13640-B	SM.TRT.TMA	4974916	114	20	2277	
150µg/mL Dimethoate		6491962	149	n/a	n/a	

Dimethoate 2-02-AN

Project:21-0001

MDB 11/17/21

Run# 94662

Standard Suitability

Standard	Response
150µg/mL Dimethoate	6537454
150µg/mL Dimethoate	6545791
150µg/mL Dimethoate	6517652
150µg/mL Dimethoate	6482356
150µg/mL Dimethoate	6525192
Average	6521689
Standard Deviation	24521
%CV	0.37599

En-Cas ID	Study ID	Response	ug/mL Foun	Dil Factor	ug/mL sample	Average
150µg/mL Dimethoate		6533614	149	n/a	n/a	
EU13727-A	SM2.UTC.TMA	0	0	2	0	
EU13727-B	SM2.UTC.TMA	0	0	2	0	0
EU13769-A	S21-04089-L2-CT-Tank-0HBA-A	0	0	2	0	
EU13769-B	S21-04089-L2-CT-Tank-0HBA-A	0	0	2	0	0
150µg/mL Dimethoate		6570515	149	n/a	n/a	
EU13729-A	SM2.TRT.TMA	4328572	98	20	1962	
EU13729-B	SM2.TRT.TMA	4312254	98	20	1954	1958
EU13776-A	S21-04089-L2-T-Tank-0HBA-A	5688240	129	20	2578	
EU13776-B	S21-04089-L2-T-Tank-0HBA-A	5676673	129	20	2572	2575
150µg/mL Dimethoate		6563805	149	n/a	n/a	

Dimethoate 2-02-AN
 Project:21-0001
 MDB 11/17/21
 Run# 94662

Standard Suitability

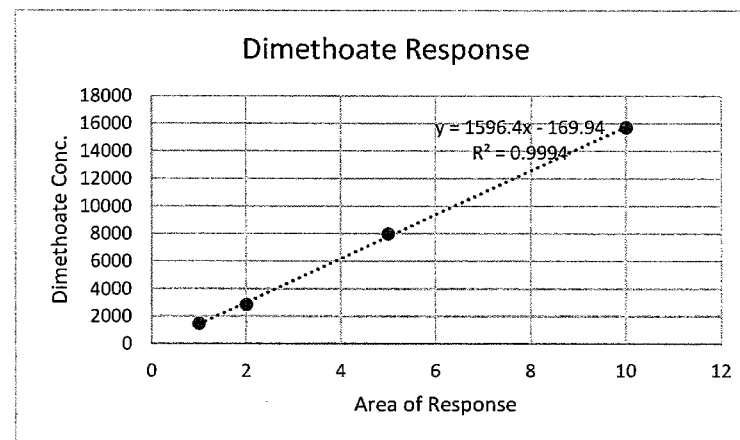
Standard	Response
150µg/mL Dimethoate	6537454
150µg/mL Dimethoate	6545791
150µg/mL Dimethoate	6517652
150µg/mL Dimethoate	6482356
150µg/mL Dimethoate	6525192
Average	6521689
Standard Deviation	24521
%CV	0.37599

En-Cas ID	Study ID	Response	Weighed	ug/mL Found	Dil Factor	ug/mL Solution	Original Conc.			
							ug/mL	Average	Found	Average
150µg/mL Dimethoate		6563805		149	n/a	n/a				
EU13765-A	Dimethoate aliquot	5594943	0.17067	127	5	634	63421		37.2%	
EU13765-B	Dimethoate aliquot	5777584	0.17317	131	5	655	65491	64456	37.8%	37.5%
150µg/mL Dimethoate		6563243		149	n/a	n/a				

Dimethoate 3-01-AN
 Project:21-0001
 MDB 10/8/21
 Run# 94657

Standard Curve

Standard	Conc.	Response
1µg/mL Dimethoate	1	1476
2µg/mL Dimethoate	2	2850
5µg/mL Dimethoate	5	7997
10µg/mL Dimethoate	10	15730
Slope		1596
Intercept		-169.94
r ²		0.99943

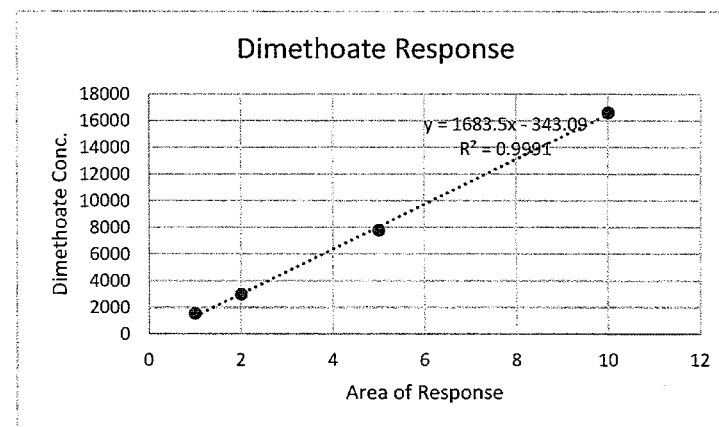


En-Cas ID	Study ID	Sample	Response	ug/mL Found	Dil Factor	ug card	Recovery %
EU13676	Control	1 disc	0	0.000	100	0.0000	n/a
EU13677	Control+ Spike	1 disc	3832	2.507	100	250.6974	101%
EU13678	Control+ Spike	1 disc	7936	5.078	100	507.7582	102%
EU13645	SM.TRT.SC1	1 disc	6559	4.215	100	421.5406	n/a
EU13646	SM.TRT.SC2	1 disc	13604	8.629	100	862.8685	n/a
EU13647	SM.TRT.SC3	1 disc	6849	4.397	100	439.6819	n/a
EU13661	S21-04089-LT-T-DISC-0HAA-Rep1	1 disc	4212	2.745	100	274.5078	n/a
EU13662	S21-04089-LT-T-DISC-0HAA-Rep2	1 disc	3931	2.569	100	256.8803	n/a
EU13663	S21-04089-LT-T-DISC-0HAA-Rep3	1 disc	4508	2.930	100	293.0299	n/a

Dimethoate 3-02-AN
 Project:21-0001
 MDB 11/22/21
 Run# 94663

Standard Curve

Standard	Conc.	Response
1µg/mL Dimethoate	1	1528
2µg/mL Dimethoate	2	2984
5µg/mL Dimethoate	5	7801
10µg/mL Dimethoate	10	16618
Slope		1684
Intercept		-343.09
r ²		0.99908



En-Cas ID	Study ID	Sample	Response	ug/mL Found	Dil Factor	ug card	Recovery %
EU13770	S21-04089-L2-C-DISC-OHAA-Rep1	1 disc	0	0.000	100	0.0000	n/a
EU13771	S21-04089-L2-C-DISC-OHAA-Rep2	1 disc	4165	2.678	100	267.7688	108%
EU13772	S21-04089-L2-C-DISC-OHAA-Rep3	1 disc	8047	4.983	100	498.3391	100%
EU13777	S21-04089-L2-T-DISC-OHAA-Rep1	1 disc	6104	3.829	100	382.9465	n/a
EU13778	S21-04089-L2-T-DISC-OHAA-Rep2	1 disc	6039	3.791	100	379.0704	n/a
EU13779	S21-04089-L2-T-DISC-OHAA-Rep3	1 disc	9496	5.845	100	584.4543	n/a
EU13734	SM2.TRT.SC1	1 disc	5493	3.467	100	346.6567	n/a
EU13735	SM2.TRT.SC2	1 disc	8238	5.097	100	509.7198	n/a
EU13736	SM2.TRT.SC3	1 disc	8702	5.373	100	537.2586	n/a

Appendix E

Summary of Statistics for the Toxicity of Facility A June Alfalfa Application Tested By Lab A



CETIS Analytical Report

 Report Date: 01 Apr-22 13:28 (p 1 of 2)
 Test Code/ID: LabA_S_T1_6h / 18-7014-0043

Foliar Acute Bee Test										A	
Analysis ID: 07-8946-6093		Endpoint: 24-hr Mortality		CETIS Version: CETISv1.9.7							
Analyzed: 01 Apr-22 13:02		Analysis: Nonparametric-Two Sample		Status Level: 1							
Edit Date: 01 Apr-22 12:59		MD5 Hash: DB2007035F9B72D25D1D89DAAFBA801C		Editor ID: 001-771-848-3							
Batch ID: 18-5358-3757		Test Type: Acute Bee Survival		Analyst: Alison Briden							
Start Date: 08 Jun-21		Protocol: OCSP 850.3030		Diluent: Not Applicable							
Ending Date: 09 Jun-21		Species: Apis Mellifera		Brine: Not Applicable							
Test Length: 24h		Taxon:		Source:						Age:	
Sample ID: 08-1259-0939		Code: LabA_S_T1_6h		Project: 36326							
Sample Date: 09 Jun-21		Material: Dimethoate		Source: Pacific EcoRisk							
Receipt Date: 09 Jun-21		CAS (PC):		Station: Lab A							
Sample Age: ---		Client:									
Comments: Post-application interval: +6h Smithers Alfalfa, Trial 1											
Data Transform		Alt Hyp		Comparison Result				PMSD			
Angular (Corrected)		C < T		0.5lbs ai/a failed 24-hr mortality endpoint				2.43%			
Wilcoxon Rank Sum Two-Sample Test											
Control	vs	Conc-lbs ai/a	Test Stat	Critical	Ties	DF	P-Type	P-Value	Decision(α:5%)		
Control		0.5*	21	---	0	10	Exact	0.0011	Significant Effect		
ANOVA Table											
Source	Sum Squares		Mean Square		DF	F Stat	P-Value	Decision(α:5%)			
Between	5.38168		5.38168		1	1850	<1.0E-05	Significant Effect			
Error	0.0290129		0.0029013		10						
Total	5.41069				11						
ANOVA Assumptions Tests											
Attribute	Test		Test Stat	Critical	P-Value	Decision(α:1%)					
Variance	Variance Ratio F Test		2.09E+15	14.9	<1.0E-05	Unequal Variances					
Distribution	Shapiro-Wilk W Normality Test		0.561	0.802	5.2E-05	Non-Normal Distribution					
24-hr Mortality Summary											
Conc-lbs ai/a	Code	Count	Mean	95% LCL	95% UCL	Median	Min	Max	Std Err	CV%	%Effect
0	00	6	0.000	0.000	0.000	0.000	0.000	0.000	0.000	---	0.00%
0.5		6	0.987	0.952	1.000	1.000	0.920	1.000	0.013	3.31%	98.67%
Angular (Corrected) Transformed Summary											
Conc-lbs ai/a	Code	Count	Mean	95% LCL	95% UCL	Median	Min	Max	Std Err	CV%	%Effect
0	00	6	0.100	0.100	0.100	0.100	0.100	0.100	0.000	0.00%	100.00%
0.5		6	1.440	1.360	1.520	1.470	1.280	1.470	0.031	5.29%	6.96%
24-hr Mortality Detail											
Conc-lbs ai/a	Code	Rep 1	Rep 2	Rep 3	Rep 4	Rep 5	Rep 6				
0	00	0.000	0.000	0.000	0.000	0.000	0.000				
0.5		1.000	1.000	1.000	1.000	1.000	0.920				
Angular (Corrected) Transformed Detail											
Conc-lbs ai/a	Code	Rep 1	Rep 2	Rep 3	Rep 4	Rep 5	Rep 6				
0	00	0.100	0.100	0.100	0.100	0.100	0.100				
0.5		1.470	1.470	1.470	1.470	1.470	1.280				
24-hr Mortality Binomials											
Conc-lbs ai/a	Code	Rep 1	Rep 2	Rep 3	Rep 4	Rep 5	Rep 6				
0	00	0/25	0/25	0/25	0/25	0/25	0/25				
0.5		25/25	25/25	25/25	25/25	25/25	23/25				

CETIS Analytical Report

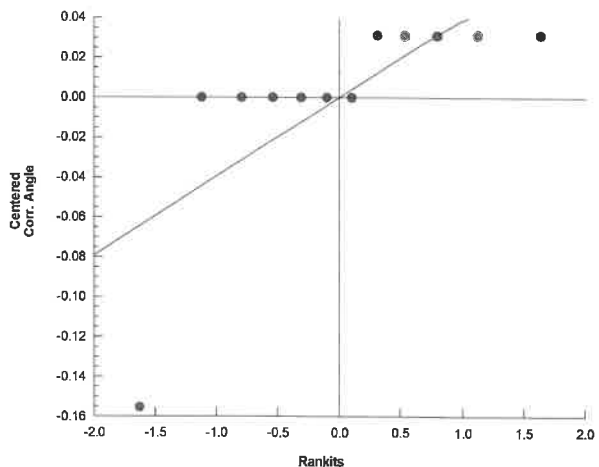
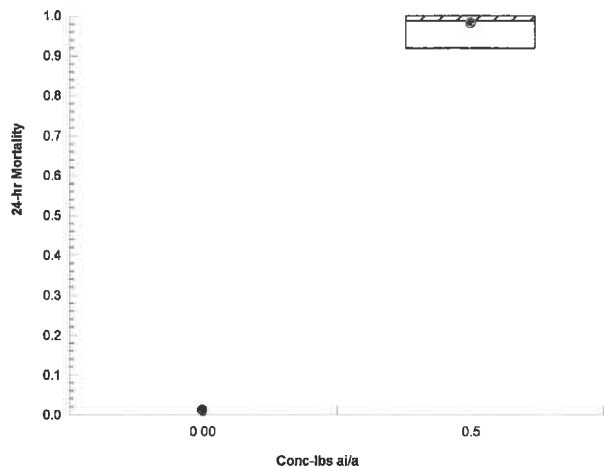
Report Date: 01 Apr-22 13:28 (p 2 of 2)
 Test Code/ID: LabA_S_T1_6h / 18-7014-0043

Foliar Acute Bee Test

A

Analysis ID: 07-8946-6093	Endpoint: 24-hr Mortality	CETIS Version: CETISv1.9.7
Analyzed: 01 Apr-22 13:02	Analysis: Nonparametric-Two Sample	Status Level: 1
Edit Date: 01 Apr-22 12:59	MD5 Hash: DB2007035F9B72D25D1D89DAAFBA801C	Editor ID: 001-771-848-3

Graphics



CETIS Analytical Report

Report Date: 01 Apr-22 13:32 (p 1 of 2)
Test Code/ID: LabA_S_T1_24h / 09-3734-5003

Foliar Acute Bee Test												A
Analysis ID: 18-2723-9049		Endpoint: 24-hr Mortality		CETIS Version: CETISv1.9.7								
Analyzed: 01 Apr-22 13:30		Analysis: Nonparametric-Two Sample		Status Level: 1								
Edit Date: 01 Apr-22 13:28		MD5 Hash: BEF602E4FF7FC4D968A9E404BEE3A092		Editor ID: 001-771-848-3								
Batch ID: 00-7565-9634		Test Type: Acute Bee Survival		Analyst: Alison Briden								
Start Date: 09 Jun-21		Protocol: OCSP 850.3030		Diluent: Not Applicable								
Ending Date: 10 Jun-21		Species: Apis Mellifera		Brine: Not Applicable								
Test Length: 24h		Taxon:		Source: Age:								
Sample ID: 03-4146-1007		Code: LabA_S_T1_24h		Project: 36326								
Sample Date: 09 Jun-21		Material: Dimethoate		Source: Pacific EcoRisk								
Receipt Date: 09 Jun-21		CAS (PC):		Station: Lab A								
Sample Age: ---		Client:										
Comments: Post-application interval: +24h Smithers Alfalfa, Trial 1												
Data Transform		Alt Hyp		Comparison Result						PMSD		
Angular (Corrected)		C < T		0.5lbs ai/a passed 24-hr mortality endpoint						9.86%		
Wilcoxon Rank Sum Two-Sample Test												
Control	vs	Conc-lbs ai/a	Test Stat	Critical	Ties	DF	P-Type	P-Value	Decision(α:5%)			
Control		0.5	29.5	---	2	10	Exact	0.0985	Non-Significant Effect			
ANOVA Table												
Source	Sum Squares		Mean Square		DF	F Stat	P-Value	Decision(α:5%)				
Between	0.0748536		0.0748536		1	1.82	0.2072	Non-Significant Effect				
Error	0.411455		0.0411455		10							
Total	0.486308				11							
ANOVA Assumptions Tests												
Attribute	Test		Test Stat	Critical	P-Value	Decision(α:1%)						
Variance	Variance Ratio F Test		47.2	14.9	0.0007	Unequal Variances						
Distribution	Shapiro-Wilk W Normality Test		0.662	0.802	0.0004	Non-Normal Distribution						
24-hr Mortality Summary												
Conc-lbs ai/a	Code	Count	Mean	95% LCL	95% UCL	Median	Min	Max	Std Err	CV%	%Effect	
0	00	6	0.007	0.000	0.024	0.000	0.000	0.040	0.007	244.95%	0.00%	
0.5		6	0.113	0.000	0.344	0.040	0.000	0.560	0.090	193.85%	10.74%	
Angular (Corrected) Transformed Summary												
Conc-lbs ai/a	Code	Count	Mean	95% LCL	95% UCL	Median	Min	Max	Std Err	CV%	%Effect	
0	00	6	0.117	0.074	0.160	0.100	0.100	0.201	0.017	35.30%	100.00%	
0.5		6	0.275	-0.023	0.573	0.201	0.100	0.846	0.116	103.23%	42.56%	
24-hr Mortality Detail												
Conc-lbs ai/a	Code	Rep 1	Rep 2	Rep 3	Rep 4	Rep 5	Rep 6					
0	00	0.000	0.000	0.040	0.000	0.000	0.000					
0.5		0.000	0.040	0.040	0.560	0.040	0.000					
Angular (Corrected) Transformed Detail												
Conc-lbs ai/a	Code	Rep 1	Rep 2	Rep 3	Rep 4	Rep 5	Rep 6					
0	00	0.100	0.100	0.201	0.100	0.100	0.100					
0.5		0.100	0.201	0.201	0.846	0.201	0.100					
24-hr Mortality Binomials												
Conc-lbs ai/a	Code	Rep 1	Rep 2	Rep 3	Rep 4	Rep 5	Rep 6					
0	00	0/25	0/25	1/25	0/25	0/25	0/25					
0.5		0/25	1/25	1/25	14/25	1/25	0/25					

CETIS Analytical Report

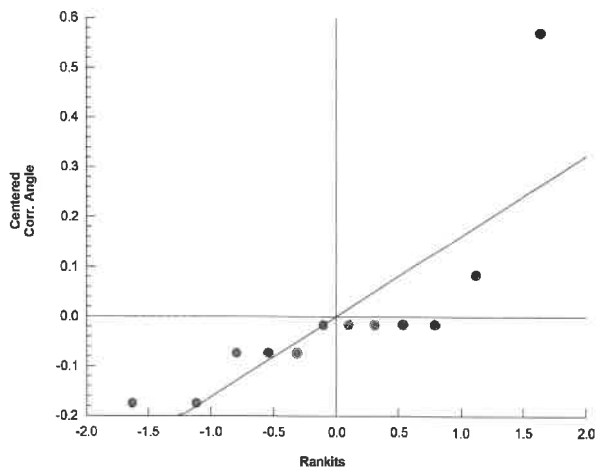
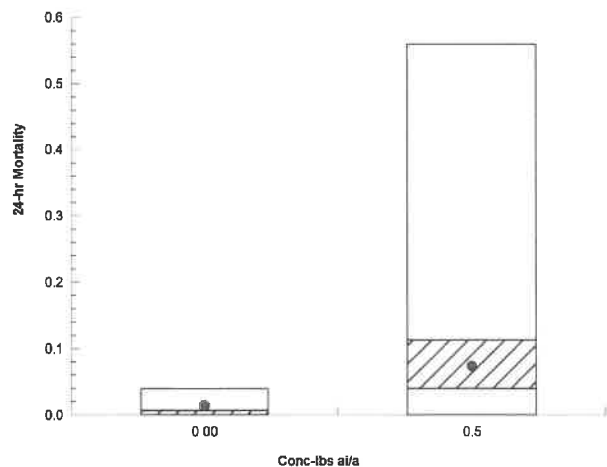
Report Date: 01 Apr-22 13:32 (p 2 of 2)
Test Code/ID: LabA_S_T1_24h / 09-3734-5003

Foliar Acute Bee Test

A

Analysis ID: 18-2723-9049 Endpoint: 24-hr Mortality CETIS Version: CETISv1.9.7
Analyzed: 01 Apr-22 13:30 Analysis: Nonparametric-Two Sample Status Level: 1
Edit Date: 01 Apr-22 13:28 MD5 Hash: BEF602E4FF7FC4D968A9E404BEE3A092 Editor ID: 001-771-848-3

Graphics



CETIS Analytical Report

Report Date: 07 Apr-22 10:24 (p 1 of 1)
Test Code/ID: LabA_S_T1_RT25 / 03-3173-0248

Foliar Acute Bee Test										A	
Analysis ID: 00-0331-4203		Endpoint: 24-hr Mortality RT25			CETIS Version: CETISv1.9.7						
Analyzed: 07 Apr-22 10:24		Analysis: Linear Interpolation (ICPIN)			Status Level: 1						
Edit Date: 07 Apr-22 10:24		MD5 Hash: 2B1D6E6B26F1B66B7506A35E7E4A7A07			Editor ID: 001-771-848-3						
Batch ID: 13-3570-0412		Test Type: Acute Bee Survival			Analyst: Alison Briden						
Start Date: 08 Jun-21		Protocol: OCSPP 850.3030			Diluent: Not Applicable						
Ending Date: 10 Jun-21		Species: Apis Mellifera			Brine: Not Applicable						
Test Length: 48h		Taxon:			Source: Age:						
Sample ID: 05-7284-0636		Code: LabA_S_T1_RT25			Project: 36326						
Sample Date: 08 Jun-21		Material: Dimethoate			Source: Pacific EcoRisk						
Receipt Date: 08 Jun-21		CAS (PC):			Station: Lab A						
Sample Age: ---		Client:									
Comments: RT25, Smithers alfalfa, Trial 1											
Linear Interpolation Options											
X Transform	Y Transform	Seed	Resamples	Exp 95% CL	Method						
Linear	Linear	355542	1	Yes	Two-Point Interpolation						
Point Estimates											
Level	T-hrs	95% LCL	95% UCL								
IC10	7.78	---	---								
IC15	8.8	---	---								
IC20	9.82	---	---								
IC25	10.8	---	---								
IC40	13.9	---	---								
IC50	16	---	---								
24-hr Mortality RT25 Summary											
		Calculated Variate							Isotonic Variate		
T-hrs	Code	Count	Mean	Median	Min	Max	CV%	%Effect	Mean	%Effect	
0		1	100	100	100	100	---	---	100		
6		1	98.7	98.7	98.7	98.7	---	---	98.7		
24		1	10.7	10.7	10.7	10.7	---	---	10.7		
24-hr Mortality RT25 Detail											
T-hrs	Code	Rep 1									
0		100									
6		98.7									
24		10.7									
Graphics											

Test Item: T = Dimethoate 400 EC Formulation Application Date: 8 Jun 2021 @ 10:23
 Treatment Rate: T = 0.5 lb a.i./Ac = 560.4 g a.i./ha
 Bee Colony Used: 20-A-10 Crop: Alfalfa
 * Corrected Mortality = $(\% T - \% C) / (100 - \% C) * 100$

Residual Timepoint: 6 Hours After Application
 Harvest Time: 06/08/2021 @ 16:00
 Exposure Time: 06/08/2021 @ 17:00

	Date:		8-Jun-21	9-Jun-21	24 Hr.		
Treatment	Cage No.	No. Bees	Number of Dead Bees		Cumulative Total	% Cumulative Mortality	% Corrected Mortality
			≤4hr	24 hr			
C (Untreated Water Spray Alfalfa)	1	25	0	0	0	0.0	NA
	2	25	0	0			
	3	25	0	0			
	4	25	0	0			
	5	25	0	0			
	6	25	0	0			
Total		150	0	0			
% Cumulative Mortality			0.0	0.0			
T (Dimethoate 400 EC Treated Alfalfa)	1	25	0	25	148	98.7	98.7
	2	25	0	25			
	3	25	0	25			
	4	25	0	25			
	5	25	0	25			
	6	25	0	23			
Total		150	0	148			
% Cumulative Mortality			0.0	98.7			

Residual Timepoint: 24 Hours After Application
 Harvest Time: 06/09/2021 @ 10:30
 Exposure Time: 06/09/2020 @ 11:26

	Date:		9-Jun-21	10-Jun-21	24 Hr.		
Treatment	Cage No.	No. Bees	Number of Dead Bees		Cumulative Total	% Cumulative Mortality	% Corrected Mortality
			≤4hr	24 hr			
C (Untreated Water Spray Alfalfa)	1	25	0	0	1	0.7	NA
	2	25	0	0			
	3	25	0	1			
	4	25	0	0			
	5	25	0	0			
	6	25	0	0			
Total		150	0	1			
% Cumulative Mortality			0.0	0.7			
T (Dimethoate 400 EC Treated Alfalfa)	1	25	0	0	17	11.3	10.7
	2	25	0	1			
	3	25	1	1			
	4	25	0	14			
	5	25	0	1			
	6	25	0	0			
Total		150	1	17			
% Cumulative Mortality			0.7	11.3			

Post-application interval: +6hr	≤ 4 - Hour Observations		24 - Hour Observations	
Location: Smithers	Test Concentration		Test Concentration	
Replicate	Control	T1	Control	T1
	Observation	Observation	Observation	Observation
1	All N	All N	2 lethargic	All dead
2	All N	All N	All N	All dead
3	All N	All N	All N	All dead
4	All N	All N	1 lethargic	All dead
5	All N	All N	2 lethargic	All dead
6	All N	All N	All N	2 remaining bees moving slowly, 23 dead
Total	All N	All N	5 lethargic	148 dead, 2 bees moving slowly

Post-application interval: +24hr	≤ 4 - Hour Observations		24 - Hour Observations	
Location: Smithers	Test Concentration		Test Concentration	
Replicate	Control	T1	Control	T1
	Observation	Observation	Observation	Observation
1	All N	All N	All N	All N
2	All N	All N	All N	All N
3	All N	All N	All N	All N
4	All N	All N	All N	All N
5	All N	All N	All N	All N
6	All N	All N	All N	All N
Total	All N	All N	All N	All N

Appendix F

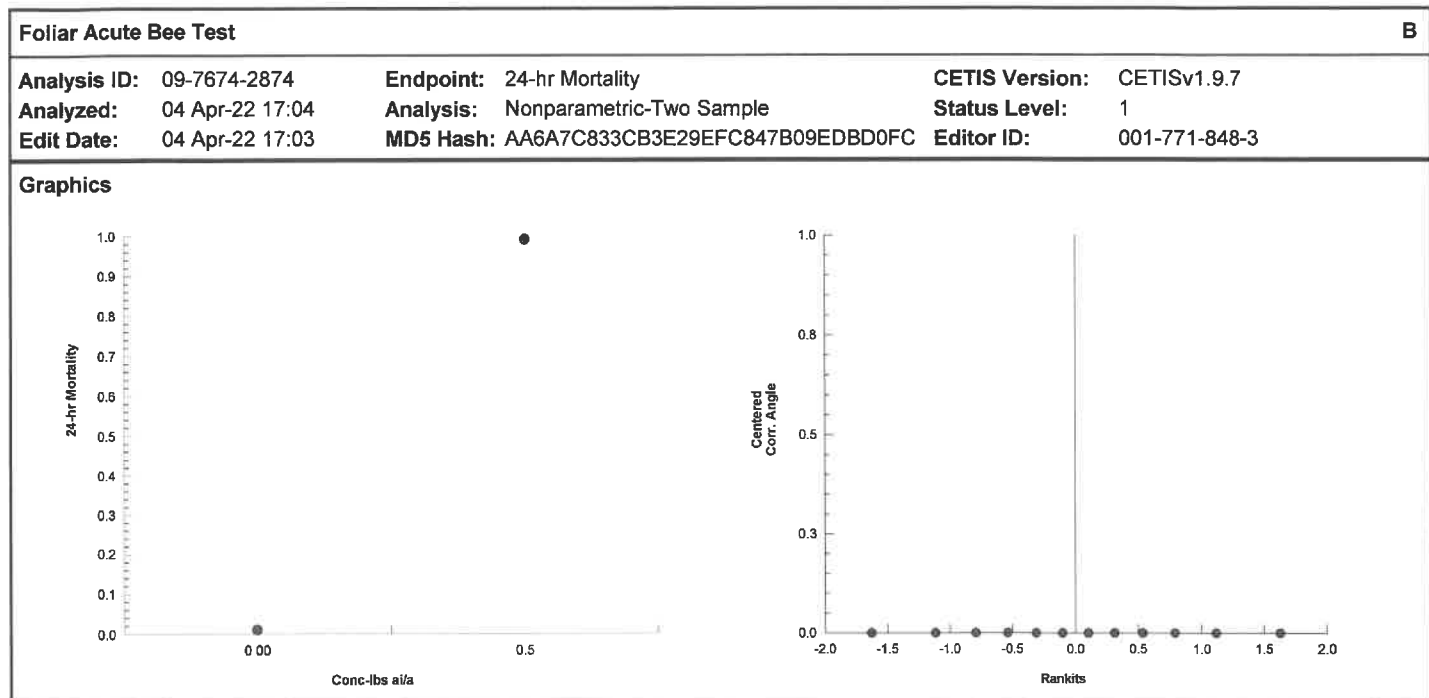
Summary of Statistics for the Toxicity of Facility A June Alfalfa Application Tested By Lab B



CETIS Analytical Report

 Report Date: 04 Apr-22 17:07 (p 1 of 2)
 Test Code/ID: LabB_S_T1_6h / 02-1299-8970

Foliar Acute Bee Test										B	
Analysis ID: 09-7674-2874		Endpoint: 24-hr Mortality		CETIS Version: CETISv1.9.7							
Analyzed: 04 Apr-22 17:04		Analysis: Nonparametric-Two Sample		Status Level: 1							
Edit Date: 04 Apr-22 17:03		MD5 Hash: AA6A7C833CB3E29EFC847B09EDBD0FC		Editor ID: 001-771-848-3							
Batch ID: 01-8123-7054		Test Type: Acute Bee Survival		Analyst: Alison Briden							
Start Date: 08 Jun-21		Protocol: OCSPP 850.3030		Diluent: Not Applicable							
Ending Date: 09 Jun-21		Species: Apis Mellifera		Brine: Not Applicable							
Test Length: 24h		Taxon:		Source:		Age:					
Sample ID: 00-5468-2977		Code: LabB_S_T1_6h		Project: 36326							
Sample Date: 08 Jun-21		Material: Dimethoate		Source: Pacific EcoRisk							
Receipt Date: 08 Jun-21		CAS (PC):		Station: Lab B							
Sample Age: ---		Client:									
Comments: Post-application interval: +6h Smithers alfalfa, Trial 1											
Data Transform		Alt Hyp		Comparison Result				PMSD			
Angular (Corrected)		C < T		0.5lbs ai/a failed 24-hr mortality endpoint				1.00%			
Wilcoxon Rank Sum Two-Sample Test											
Control	vs	Conc-lbs ai/a	Test Stat	Critical	Ties	DF	P-Type	P-Value	Decision(α:5%)		
Control		0.5*	21	---	0	10	Exact	0.0011	Significant Effect		
ANOVA Table											
Source	Sum Squares		Mean Square		DF	F Stat	P-Value	Decision(α:5%)			
Between	5.63449		5.63449		1	3.17E+16	<1.0E-05	Significant Effect			
Error	1.776E-15		1.776E-16		10						
Total	5.63449				11						
ANOVA Assumptions Tests											
Attribute	Test		Test Stat	Critical	P-Value	Decision(α:1%)					
Variance	Variance Ratio F Test		128	14.9	5.7E-05	Unequal Variances					
Distribution	Shapiro-Wilk W Normality Test		0.65	0.802	0.0003	Non-Normal Distribution					
24-hr Mortality Summary											
Conc-lbs ai/a	Code	Count	Mean	95% LCL	95% UCL	Median	Min	Max	Std Err	CV%	%Effect
0	00	6	0.000	0.000	0.000	0.000	0.000	0.000	0.000	---	0.00%
0.5		6	1.000	1.000	1.000	1.000	1.000	1.000	0.000	0.00%	100.00%
Angular (Corrected) Transformed Summary											
Conc-lbs ai/a	Code	Count	Mean	95% LCL	95% UCL	Median	Min	Max	Std Err	CV%	%Effect
0	00	6	0.100	0.100	0.100	0.100	0.100	0.100	0.000	0.00%	100.00%
0.5		6	1.470	1.470	1.470	1.470	1.470	1.470	0.000	0.00%	6.81%
24-hr Mortality Detail											
Conc-lbs ai/a	Code	Rep 1	Rep 2	Rep 3	Rep 4	Rep 5	Rep 6				
0	00	0.000	0.000	0.000	0.000	0.000	0.000				
0.5		1.000	1.000	1.000	1.000	1.000	1.000				
Angular (Corrected) Transformed Detail											
Conc-lbs ai/a	Code	Rep 1	Rep 2	Rep 3	Rep 4	Rep 5	Rep 6				
0	00	0.100	0.100	0.100	0.100	0.100	0.100				
0.5		1.470	1.470	1.470	1.470	1.470	1.470				
24-hr Mortality Binomials											
Conc-lbs ai/a	Code	Rep 1	Rep 2	Rep 3	Rep 4	Rep 5	Rep 6				
0	00	0/25	0/25	0/25	0/25	0/25	0/25				
0.5		25/25	25/25	25/25	25/25	25/25	25/25				



CETIS Analytical Report

 Report Date: 06 Apr-22 10:47 (p 1 of 2)
 Test Code/ID: LabB_S_T1_24h / 14-1306-8091

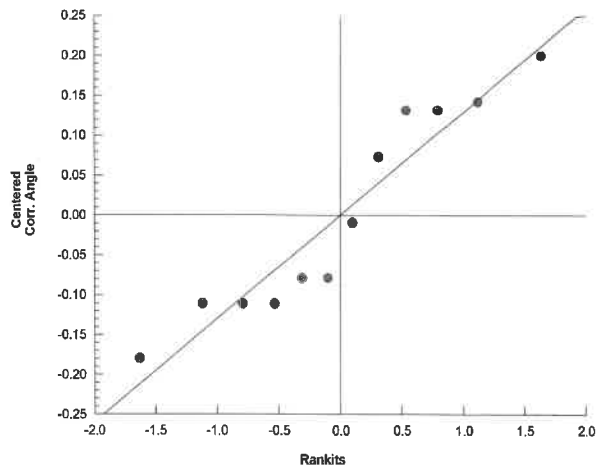
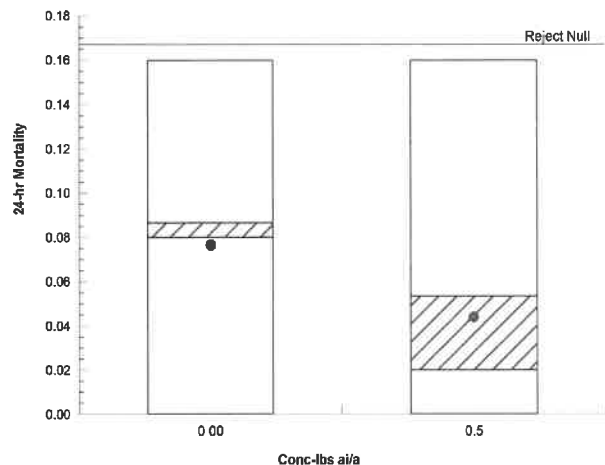
Foliar Acute Bee Test										B	
Analysis ID: 20-6057-9938		Endpoint: 24-hr Mortality		CETIS Version: CETISv1.9.7							
Analyzed: 04 Apr-22 17:08		Analysis: Parametric-Two Sample		Status Level: 1							
Edit Date: 04 Apr-22 17:08		MD5 Hash: 2B6482C864C2768AD5452DCAFC98FF98		Editor ID: 001-771-848-3							
Batch ID: 03-8577-3764		Test Type: Acute Bee Survival		Analyst: Alison Briden							
Start Date: 09 Jun-21		Protocol: OCSPP 850.3030		Diluent: Not Applicable							
Ending Date: 10 Jun-21		Species: Apis Mellifera		Brine: Not Applicable							
Test Length: 24h		Taxon:		Source: Age:							
Sample ID: 15-3883-5158		Code: LabB_S_T1_24h		Project: 36326							
Sample Date: 08 Jun-21		Material: Dimethoate		Source: Pacific EcoRisk							
Receipt Date: 08 Jun-21		CAS (PC):		Station: Lab B							
Sample Age: 24h		Client:									
Comments: Post-application interval: + 24h Smithers Alfalfa, Trial 1											
Data Transform		Alt Hyp		Comparison Result						PMSD	
Angular (Corrected)		C < T		0.5lbs ai/a passed 24-hr mortality endpoint						8.81%	
Equal Variance t Two-Sample Test											
Control	vs	Conc-lbs ai/a	Test Stat	Critical	MSD	DF	P-Type	P-Value	Decision(α:5%)		
Control		0.5	-0.882	1.81	0.141	10	CDF	0.8008	Non-Significant Effect		
ANOVA Table											
Source	Sum Squares		Mean Square		DF	F Stat	P-Value	Decision(α:5%)			
Between	0.0141824		0.0141824		1	0.778	0.3984	Non-Significant Effect			
Error	0.182267		0.0182267		10						
Total	0.196449				11						
ANOVA Assumptions Tests											
Attribute	Test		Test Stat	Critical	P-Value	Decision(α:1%)					
Variance	Variance Ratio F Test		1.15	14.9	0.8812	Equal Variances					
Distribution	Shapiro-Wilk W Normality Test		0.896	0.802	0.1391	Normal Distribution					
24-hr Mortality Summary											
Conc-lbs ai/a	Code	Count	Mean	95% LCL	95% UCL	Median	Min	Max	Std Err	CV%	%Effect
0	00	6	0.087	0.014	0.159	0.080	0.000	0.160	0.028	79.50%	0.00%
0.5		6	0.053	0.000	0.127	0.020	0.000	0.160	0.029	131.34%	-3.65%
Angular (Corrected) Transformed Summary											
Conc-lbs ai/a	Code	Count	Mean	95% LCL	95% UCL	Median	Min	Max	Std Err	CV%	%Effect
0	00	6	0.280	0.143	0.417	0.278	0.100	0.412	0.053	46.50%	100.00%
0.5		6	0.211	0.065	0.358	0.151	0.100	0.412	0.057	66.13%	132.56%
24-hr Mortality Detail											
Conc-lbs ai/a	Code	Rep 1	Rep 2	Rep 3	Rep 4	Rep 5	Rep 6				
0	00	0.160	0.160	0.040	0.040	0.000	0.120				
0.5		0.000	0.040	0.000	0.160	0.000	0.120				
Angular (Corrected) Transformed Detail											
Conc-lbs ai/a	Code	Rep 1	Rep 2	Rep 3	Rep 4	Rep 5	Rep 6				
0	00	0.412	0.412	0.201	0.201	0.100	0.354				
0.5		0.100	0.201	0.100	0.412	0.100	0.354				
24-hr Mortality Binomials											
Conc-lbs ai/a	Code	Rep 1	Rep 2	Rep 3	Rep 4	Rep 5	Rep 6				
0	00	4/25	4/25	1/25	1/25	0/25	3/25				
0.5		0/25	1/25	0/25	4/25	0/25	3/25				

Foliar Acute Bee Test

B

Analysis ID: 20-6057-9938
Analyzed: 04 Apr-22 17:08
Edit Date: 04 Apr-22 17:08Endpoint: 24-hr Mortality
Analysis: Parametric-Two Sample
MD5 Hash: 2B6482C864C2768AD5452DCAFC98FF98CETIS Version: CETISv1.9.7
Status Level: 1
Editor ID: 001-771-848-3

Graphics



CETIS Analytical Report

Report Date: 07 Apr-22 10:25 (p 1 of 1)
 Test Code/ID: LabB_S_T1_RT25 / 14-4052-7390

Foliar Acute Bee Test						B												
Analysis ID: 16-5389-4718		Endpoint: 24-hr Mortality RT25		CETIS Version: CETISv1.9.7														
Analyzed: 07 Apr-22 10:25		Analysis: Linear Interpolation (ICPIN)		Status Level: 1														
Edit Date: 07 Apr-22 10:25		MD5 Hash: 5108EB43A43F62FFA6D4EA8E54E409D8		Editor ID: 001-771-848-3														
Batch ID: 16-4951-7886		Test Type: Acute Bee Survival		Analyst: Alison Briden														
Start Date: 08 Jun-21		Protocol: OCSPP 850.3030		Diluent: Not Applicable														
Ending Date: 10 Jun-21		Species: Apis Mellifera		Brine: Not Applicable														
Test Length: 48h		Taxon:		Source: Age:														
Sample ID: 09-6976-3454		Code: LabB_S_T1_RT25		Project: 36326														
Sample Date: 08 Jun-21		Material: Dimethoate		Source: Pacific EcoRisk														
Receipt Date: 08 Jun-21		CAS (PC):		Station: Lab B														
Sample Age: ---		Client:																
Comments: RT25, Smithers alfalfa, Trial 1																		
Linear Interpolation Options																		
X Transform	Y Transform	Seed	Resamples	Exp 95% CL	Method													
Linear	Linear	1067945	1	Yes	Two-Point Interpolation													
Point Estimates																		
Level	T-hrs	95% LCL	95% UCL															
IC10	7.9	---	---															
IC15	8.85	---	---															
IC20	9.8	---	---															
IC25	10.8	---	---															
IC40	13.6	---	---															
IC50	15.5	---	---															
24-hr Mortality RT25 Summary																		
		Calculated Variate							Isotonic Variate									
T-hrs	Code	Count	Mean	Median	Min	Max	CV%	%Effect	Mean	%Effect								
0		1	100	100	100	100	---	---	100									
6		1	100	100	100	100	---	---	100									
24		1	5.3	5.3	5.3	5.3	---	---	5.3									
24-hr Mortality RT25 Detail																		
T-hrs	Code	Rep 1																
0		100																
6		100																
24		5.3																
Graphics																		
<table border="1"> <caption>Graph Data Points</caption> <thead> <tr> <th>T-hrs</th> <th>24-hr Mortality RT25</th> </tr> </thead> <tbody> <tr> <td>0</td> <td>100</td> </tr> <tr> <td>6</td> <td>100</td> </tr> <tr> <td>24</td> <td>5.3</td> </tr> </tbody> </table>											T-hrs	24-hr Mortality RT25	0	100	6	100	24	5.3
T-hrs	24-hr Mortality RT25																	
0	100																	
6	100																	
24	5.3																	

Test Item: T= Dimethoate 400 EC Formulation
 Application Date: 8 Jun 2021 @ 16:23
 Treatment Rate: T = 0.5 lb a.i./Ac = 566.4 g a.i./ha
 Bee Colony Used: 20-A-10
 Crop: Alfalfa
 * Corrected Mortality = $(\% T - \% C) / (100 - \% C) * 100$

Residual Timepoint: 6 Hours After Application
 Harvest Time: 06/08/2021 @ 16:00
 Exposure Time: 06/08/2021 @ C = 17:49, T = 17:52

	Date:		8-Jun-21	9-Jun-21	24 Hr.		
Treatment	Cage No.	No. Bees	Number of Dead Bees		Cumulative Total	% Cumulative Mortality	% Corrected Mortality
			≤4hr	24 hr			
C (Untreated Water Spray Alfalfa)	1	25	NR	0	0	0.0	NA
	2	25	NR	0			
	3	25	NR	0			
	4	25	NR	0			
	5	25	NR	0			
	6	25	NR	0			
Total		150	0	0			
% Cumulative Mortality			0.0	0.0			
T (Dimethoate 400 EC Treated Alfalfa)	1	25	NR	25	150	100.0	100
	2	25	NR	25			
	3	25	NR	25			
	4	25	NR	25			
	5	25	NR	25			
	6	25	NR	25			
Total		150	0	150			
% Cumulative Mortality			0.0	100.0			

Residual Timepoint: 24 Hours After Application
 Harvest Time: 06/09/2021 @ 10:30
 Exposure Time: 06/09/2021 @ C = 12:50, T = 12:53

	Date:		9-Jun-21	10-Jun-21	24 Hr.		
Treatment	Cage No.	No. Bees	Number of Dead Bees		Cumulative Total	% Cumulative Mortality	% Corrected Mortality
			≤4hr	24 hr			
C (Untreated Water Spray Alfalfa)	1	25	4	4	13	8.7	NA
	2	25	2	4			
	3	25	1	1			
	4	25	1	1			
	5	25	0	0			
	6	25	3	3			
Total		150	11	13			
% Cumulative Mortality			7.3	8.7			
T (Dimethoate 400 EC Treated Alfalfa)	1	25	0	0	8	5.3	-3.6
	2	25	1	1			
	3	25	0	0			
	4	25	1	4			
	5	25	0	0			
	6	25	1	3			
Total		150	3	8			
% Cumulative Mortality			2.0	5.3			

1 affected bee @ 4-hour assessment

Appendix G

Summary of Statistics for the Toxicity of Facility B June Alfalfa Application Tested By Lab A



CETIS Analytical Report

 Report Date: 04 Apr-22 16:55 (p 1 of 2)
 Test Code/ID: LabA_E_T1_6h / 01-1113-8303

Foliar Acute Bee Test										A	
Analysis ID: 16-1533-2294		Endpoint: 24-hr Mortality		CETIS Version: CETISv1.9.7							
Analyzed: 04 Apr-22 16:55		Analysis: Parametric-Two Sample		Status Level: 1							
Edit Date: 04 Apr-22 16:54		MD5 Hash: 11576B6B02B3B936E074495F84043EB0		Editor ID: 001-771-848-3							
Batch ID: 01-7318-7276		Test Type: Acute Bee Survival		Analyst: Alison Briden							
Start Date: 09 Jun-21		Protocol: OCSP 850.3030		Diluent: Not Applicable							
Ending Date: 10 Jun-21		Species: Apis Mellifera		Brine: Not Applicable							
Test Length: 24h		Taxon:		Source:		Age:					
Sample ID: 06-4409-6982		Code: LabA_E_T1_6h		Project: 36326							
Sample Date: 09 Jun-21		Material: Dimethoate		Source: Pacific EcoRisk							
Receipt Date: 09 Jun-21		CAS (PC):		Station: Lab A							
Sample Age: ---		Client:									
Comments: Post-application interval: +6h Eurofins Alfalfa, Trial 1											
Data Transform		Alt Hyp		Comparison Result				PMSD			
Angular (Corrected)		C < T		0.5lbs ai/a failed 24-hr mortality endpoint				2.15%			
Equal Variance t Two-Sample Test											
Control	vs	Conc-lbs ai/a	Test Stat	Critical	MSD	DF	P-Type	P-Value	Decision(α:5%)		
Control		0.5*	46.2	1.81	0.051	10	CDF	<1.0E-05	Significant Effect		
ANOVA Table											
Source	Sum Squares		Mean Square		DF	F Stat	P-Value	Decision(α:5%)			
Between	5.09344		5.09344		1	2130	<1.0E-05	Significant Effect			
Error	0.0238922		0.0023892		10						
Total	5.11733				11						
ANOVA Assumptions Tests											
Attribute	Test		Test Stat	Critical	P-Value	Decision(α:1%)					
Variance	Variance Ratio F Test		1.8	14.9	0.5345	Equal Variances					
Distribution	Shapiro-Wilk W Normality Test		0.844	0.802	0.0306	Normal Distribution					
24-hr Mortality Summary											
Conc-lbs ai/a	Code	Count	Mean	95% LCL	95% UCL	Median	Min	Max	Std Err	CV%	%Effect
0	00	6	0.007	0.000	0.024	0.000	0.000	0.040	0.007	244.95%	0.00%
0.5		6	0.980	0.957	1.000	0.980	0.960	1.000	0.009	2.24%	97.99%
Angular (Corrected) Transformed Summary											
Conc-lbs ai/a	Code	Count	Mean	95% LCL	95% UCL	Median	Min	Max	Std Err	CV%	%Effect
0	00	6	0.117	0.074	0.160	0.100	0.100	0.201	0.017	35.30%	100.00%
0.5		6	1.420	1.360	1.480	1.420	1.370	1.470	0.023	3.90%	8.24%
24-hr Mortality Detail											
Conc-lbs ai/a	Code	Rep 1	Rep 2	Rep 3	Rep 4	Rep 5	Rep 6				
0	00	0.000	0.000	0.000	0.000	0.000	0.040				
0.5		0.960	1.000	1.000	1.000	0.960	0.960				
Angular (Corrected) Transformed Detail											
Conc-lbs ai/a	Code	Rep 1	Rep 2	Rep 3	Rep 4	Rep 5	Rep 6				
0	00	0.100	0.100	0.100	0.100	0.100	0.201				
0.5		1.370	1.470	1.470	1.470	1.370	1.370				
24-hr Mortality Binomials											
Conc-lbs ai/a	Code	Rep 1	Rep 2	Rep 3	Rep 4	Rep 5	Rep 6				
0	00	0/25	0/25	0/25	0/25	0/25	1/25				
0.5		24/25	25/25	25/25	25/25	24/25	24/25				

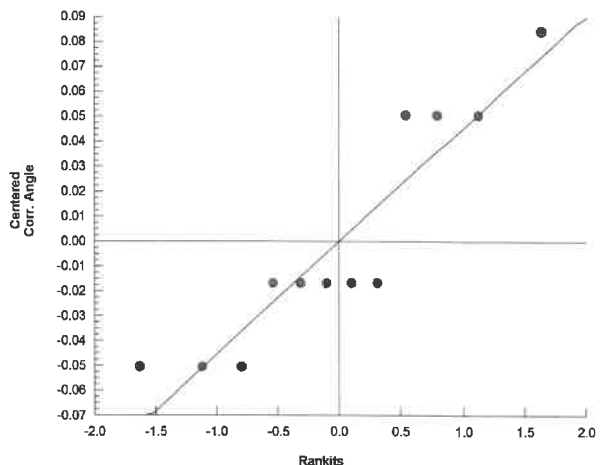
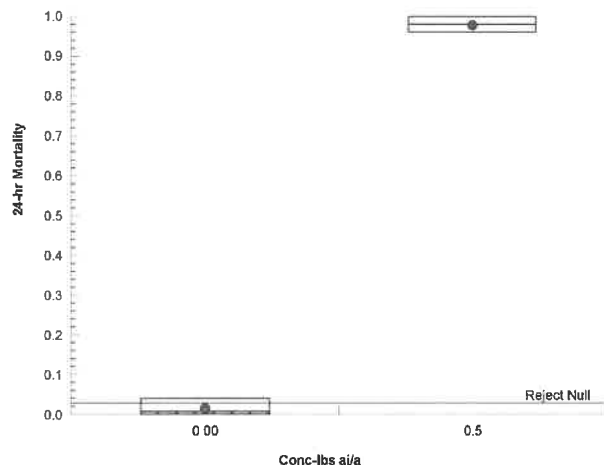
Foliar Acute Bee Test

A

Analysis ID: 16-1533-2294 Endpoint: 24-hr Mortality
Analyzed: 04 Apr-22 16:55 Analysis: Parametric-Two Sample
Edit Date: 04 Apr-22 16:54 MD5 Hash: 11576B6B02B3B936E074495F84043EB0

CETIS Version: CETISv1.9.7
Status Level: 1
Editor ID: 001-771-848-3

Graphics



CETIS Analytical Report

Report Date: 04 Apr-22 16:57 (p 1 of 2)
Test Code/ID: LabA_E_T1_24h / 16-9911-2362

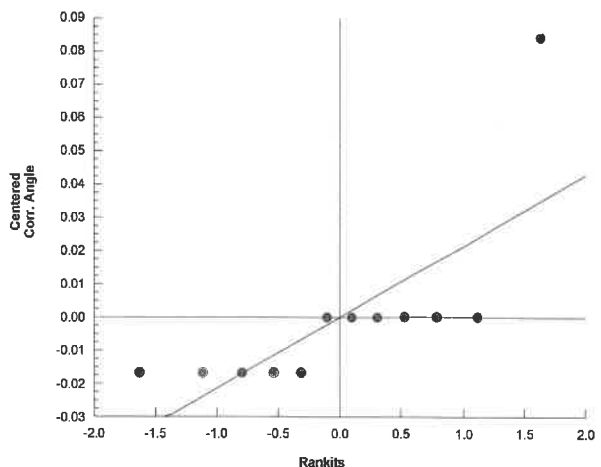
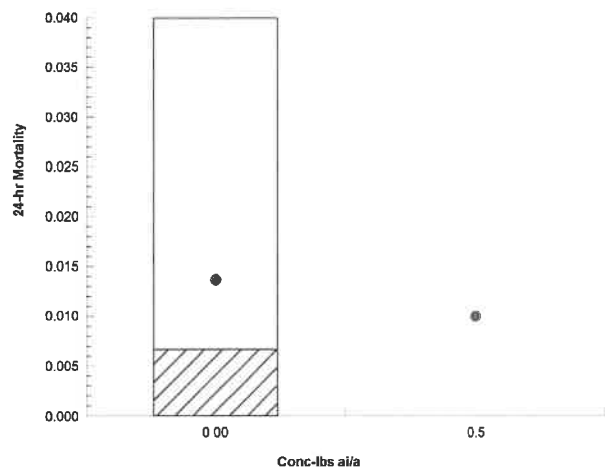
Foliar Acute Bee Test											A
Analysis ID: 05-0596-1482		Endpoint: 24-hr Mortality				CETIS Version: CETISv1.9.7					
Analyzed: 04 Apr-22 16:57		Analysis: Nonparametric-Two Sample				Status Level: 1					
Edit Date: 04 Apr-22 16:57		MD5 Hash: 689DFF7FDC851B54B61088CC95E1ED4C				Editor ID: 001-771-848-3					
Batch ID: 02-8648-3399		Test Type: Acute Bee Survival				Analyst: Alison Briden					
Start Date: 10 Jun-21		Protocol: OCSPP 850.3030				Diluent: Not Applicable					
Ending Date: 11 Jun-21		Species: Apis Mellifera				Brine: Not Applicable					
Test Length: 24h		Taxon:				Source:		Age:			
Sample ID: 20-4643-0655		Code: LabA_E_T1_24h				Project: 36326					
Sample Date: 10 Jun-21		Material: Dimethoate				Source: Pacific EcoRisk					
Receipt Date: 10 Jun-21		CAS (PC):				Station: Lab A					
Sample Age: ---		Client:									
Comments: Post-application interval: +24h Eurofins Alfalfa, Trial 1											
Data Transform		Alt Hyp			Comparison Result					PMSD	
Angular (Corrected)		C < T			0.5lbs ai/a passed 24-hr mortality endpoint					1.51%	
Wilcoxon Rank Sum Two-Sample Test											
Control	vs	Conc-lbs ai/a	Test Stat	Critical	Ties	DF	P-Type	P-Value	Decision(α:5%)		
Control		0.5	42	---	1	10	Exact	1.0000	Non-Significant Effect		
ANOVA Table											
Source	Sum Squares		Mean Square		DF	F Stat	P-Value	Decision(α:5%)			
Between	0.0008533		0.0008533		1	1	0.3409	Non-Significant Effect			
Error	0.0085329		0.0008533		10						
Total	0.0093862				11						
ANOVA Assumptions Tests											
Attribute	Test				Test Stat	Critical	P-Value	Decision(α:1%)			
Variance	Variance Ratio F Test				6.15E+14	14.9	<1.0E-05	Unequal Variances			
Distribution	Shapiro-Wilk W Normality Test				0.561	0.802	5.2E-05	Non-Normal Distribution			
24-hr Mortality Summary											
Conc-lbs ai/a	Code	Count	Mean	95% LCL	95% UCL	Median	Min	Max	Std Err	CV%	%Effect
0	00	6	0.007	0.000	0.024	0.000	0.000	0.040	0.007	244.95%	0.00%
0.5		6	0.000	0.000	0.000	0.000	0.000	0.000	0.000	---	-0.67%
Angular (Corrected) Transformed Summary											
Conc-lbs ai/a	Code	Count	Mean	95% LCL	95% UCL	Median	Min	Max	Std Err	CV%	%Effect
0	00	6	0.117	0.074	0.160	0.100	0.100	0.201	0.017	35.30%	100.00%
0.5		6	0.100	0.100	0.100	0.100	0.100	0.100	0.000	0.00%	116.84%
24-hr Mortality Detail											
Conc-lbs ai/a	Code	Rep 1	Rep 2	Rep 3	Rep 4	Rep 5	Rep 6				
0	00	0.000	0.000	0.000	0.000	0.000	0.040				
0.5		0.000	0.000	0.000	0.000	0.000	0.000				
Angular (Corrected) Transformed Detail											
Conc-lbs ai/a	Code	Rep 1	Rep 2	Rep 3	Rep 4	Rep 5	Rep 6				
0	00	0.100	0.100	0.100	0.100	0.100	0.201				
0.5		0.100	0.100	0.100	0.100	0.100	0.100				
24-hr Mortality Binomials											
Conc-lbs ai/a	Code	Rep 1	Rep 2	Rep 3	Rep 4	Rep 5	Rep 6				
0	00	0/25	0/25	0/25	0/25	0/25	1/25				
0.5		0/25	0/25	0/25	0/25	0/25	0/25				

Foliar Acute Bee Test

A

Analysis ID: 05-0596-1482
Analyzed: 04 Apr-22 16:57
Edit Date: 04 Apr-22 16:57Endpoint: 24-hr Mortality
Analysis: Nonparametric-Two Sample
MD5 Hash: 689DFF7FDC851B54B61088CC95E1ED4CCETIS Version: CETISv1.9.7
Status Level: 1
Editor ID: 001-771-848-3

Graphics



CETIS Analytical Report

Report Date: 07 Apr-22 10:21 (p 1 of 1)
 Test Code/ID: LabA_E_T1_RT25 / 08-5528-8742

Foliar Acute Bee Test						A				
Analysis ID: 17-5130-2753	Endpoint: 24-hr Mortality RT25	CETIS Version: CETISv1.9.7								
Analyzed: 07 Apr-22 10:21	Analysis: Linear Interpolation (ICPIN)	Status Level: 1								
Edit Date: 07 Apr-22 10:21	MD5 Hash: 048272EAF7E5B6B5571B9D70A00EDF00	Editor ID: 001-771-848-3								
Batch ID: 01-0630-2381	Test Type: Acute Bee Survival	Analyst: Alison Briden								
Start Date: 09 Jun-21	Protocol: OCSPP 850.3030	Diluent: Not Applicable								
Ending Date: 11 Jun-21	Species: Apis Mellifera	Brine: Not Applicable								
Test Length: 48h	Taxon:	Source:				Age:				
Sample ID: 16-6323-3594	Code: LabA_E_T1_RT25	Project: 36326								
Sample Date: 09 Jun-21	Material: Dimethoate	Source: Pacific EcoRisk								
Receipt Date: 09 Jun-21	CAS (PC):	Station: Lab A								
Sample Age: ---	Client:									
Comments: RT25, Eurofins alfalfa, Trial 1										
Linear Interpolation Options										
X Transform	Y Transform	Seed	Resamples	Exp 95% CL	Method					
Linear	Linear	1411733	1	Yes	Two-Point Interpolation					
Point Estimates										
Level	T-hrs	95% LCL	95% UCL							
IC10	7.47	---	---							
IC15	8.39	---	---							
IC20	9.31	---	---							
IC25	10.2	---	---							
IC40	13	---	---							
IC50	14.8	---	---							
24-hr Mortality RT25 Summary										
		Calculated Variate							Isotonic Variate	
T-hrs	Code	Count	Mean	Median	Min	Max	CV%	%Effect	Mean	%Effect
0		1	100	100	100	100	---	---	100	
6		1	98	98	98	98	---	---	98	
24		1	0	0	0	0	---	---	0	
24-hr Mortality RT25 Detail										
T-hrs	Code	Rep 1								
0		100								
6		98								
24		0								
Graphics										

Test Item: T = Dimethoate 400 EC Formulation
Application Date: 9 Jun 2021 @ 10:47
Treatment Rate: T = 0.5 lb al/Ac = 560.4 g a.i./ha
Be Colony Used: 20-A-10
Crop: Alfalfa
*** Corrected Mortality =** $(\% T - \% C) / (100 - \% C) * 100$

Residual Timepoint: 6 Hours After Application
Harvest Time: 06/09/2021 @ 16:34
Exposure Time: 06/09/2021 @ C = 17:17, T = 17:22

	Date:		9-Jun-21	10-Jun-21	24 Hr.		
Treatment	Cage No.	No. Bees	Number of Dead Bees		Cumulative Total	% Cumulative Mortality	% Corrected Mortality
			≤4hr	24 hr			
C (Untreated Water Spray Alfalfa)	1	25	0	0	1	0.7	NA
	2	25	0	0			
	3	25	0	0			
	4	25	0	0			
	5	25	0	0			
	6	25	0	1			
Total		150	0	1			
% Cumulative Mortality			0.0	0.7			
T (Dimethoate 400 EC Treated Alfalfa)	1	25	0	24	147	98.0	98
	2	25	0	25			
	3	25	0	25			
	4	25	0	25			
	5	25	0	24			
	6	25	0	24			
Total		150	0	147			
% Cumulative Mortality			0.0	98.0			

Residual Timepoint: 24 Hours After Application
Harvest Time: 06/10/2021 @ 10:34
Exposure Time: 06/10/2021 @ C = 11:22, T = 11:22

	Date:		10-Jun-21	11-Jun-21	24 Hr.		
Treatment	Cage No.	No. Bees	Number of Dead Bees		Cumulative Total	% Cumulative Mortality	% Corrected Mortality
			≤4hr	24 hr			
C (Untreated Water Spray Alfalfa)	1	25	0	0	1	0.7	NA
	2	25	0	0			
	3	25	0	0			
	4	25	0	0			
	5	25	0	0			
	6	25	0	1			
Total		150	0	1			
% Cumulative Mortality			0.0	0.7			
T (Dimethoate 400 EC Treated Alfalfa)	1	25	0	0	0	0.0	-0.7
	2	25	0	0			
	3	25	0	0			
	4	25	0	0			
	5	25	0	0			
	6	25	0	0			
Total		150	0	0			
% Cumulative Mortality			0.0	0.0			

Post-application interval: +6hr	≤ 4 - Hour Observations		24 - Hour Observations	
Location: Eurofins	Test Concentration		Test Concentration	
Replicate	Control	T1	Control	T1
	Observation	Observation	Observation	Observation
1	All N	All N	All N	1 remaining bee lethargic
2	All N	All N	1 lethargic	All dead
3	All N	All N	All N	All dead
4	All N	All N	All N	All dead
5	All N	All N	All N	1 remaining bee lethargic
6	All N	All N	All N	1 remaining bee lethargic
Total	All N	All N	1 lethargic	147 dead, 3 bees lethargic

Post-application interval: +24hr	≤ 4 - Hour Observations		24 - Hour Observations	
Location: Eurofins	Test Concentration		Test Concentration	
Replicate	Control	T1	Control	T1
	Observation	Observation	Observation	Observation
1	All N	All N	All N	All N
2	All N	All N	All N	All N
3	All N	All N	All N	All N
4	All N	All N	All N	All N
5	All N	All N	All N	All N
6	All N	All N	All N	All N
Total	All N	All N	All N	All N

N=normal
MO=moribund

IN=intoxicated
AT=ataxia

TR=trembling

Appendix H

Summary of Statistics for the Toxicity of Facility B June Alfalfa Application Tested By Lab B



CETIS Analytical Report

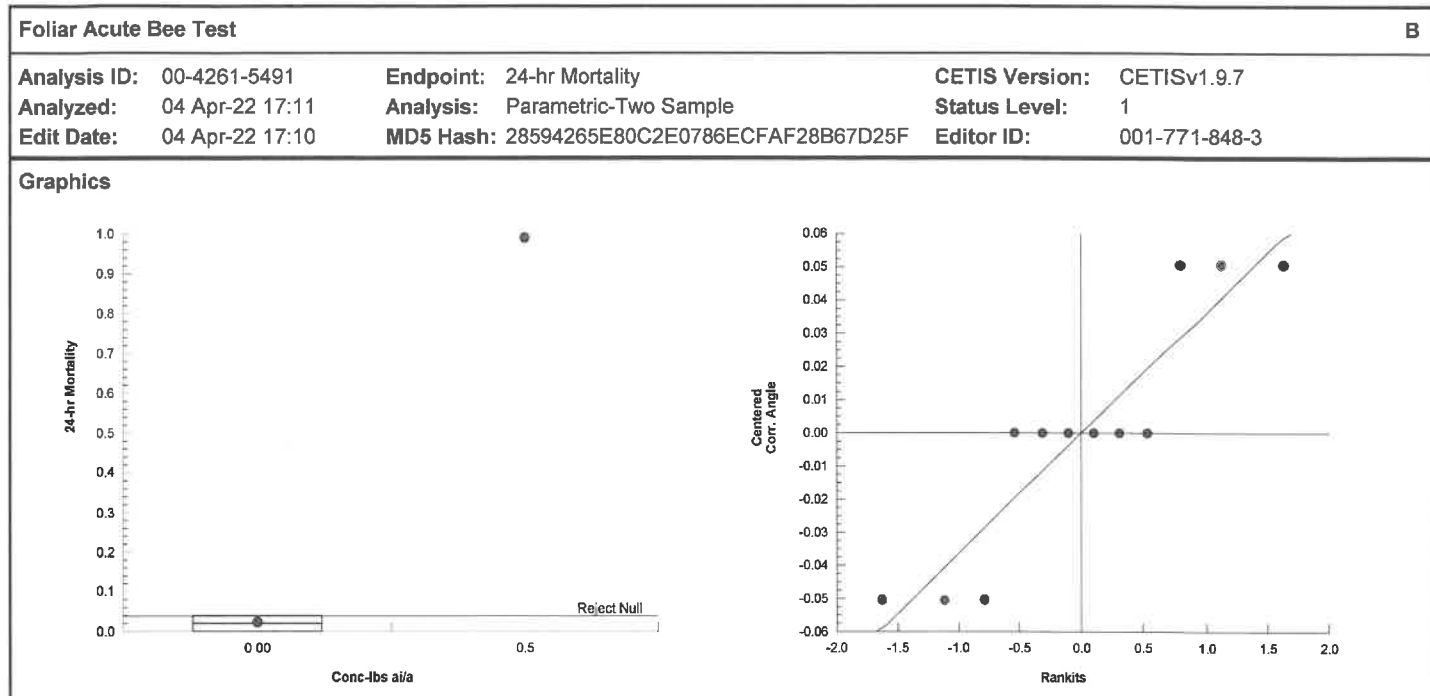
 Report Date: 04 Apr-22 17:12 (p 1 of 2)
 Test Code/ID: LabB_E_T1_6h / 03-8454-3196

Foliar Acute Bee Test										B	
Analysis ID: 00-4261-5491		Endpoint: 24-hr Mortality		CETIS Version: CETISv1.9.7							
Analyzed: 04 Apr-22 17:11		Analysis: Parametric-Two Sample		Status Level: 1							
Edit Date: 04 Apr-22 17:10		MD5 Hash: 28594265E80C2E0786ECFAF28B67D25F		Editor ID: 001-771-848-3							
Batch ID: 21-3420-9058		Test Type: Acute Bee Survival		Analyst: Alison Briden							
Start Date: 09 Jun-21		Protocol: OCSP 850.3030		Diluent: Not Applicable							
Ending Date: 10 Jun-21		Species: Apis Mellifera		Brine: Not Applicable							
Test Length: 24h		Taxon:		Source:		Age:					
Sample ID: 18-7772-5952		Code: LabB_E_T1_6h		Project: 36326							
Sample Date: 09 Jun-21		Material: Dimethoate		Source: Pacific EcoRisk							
Receipt Date: 09 Jun-21		CAS (PC):		Station: Lab B							
Sample Age: ---		Client:									
Comments: Post-application interval: 6h Eurofins alfalfa, Trial 1											
Data Transform		Alt Hyp		Comparison Result				PMSD			
Angular (Corrected)		C < T		0.5lbs ai/a failed 24-hr mortality endpoint				1.84%			
Unequal Variance t Two-Sample Test											
Control	vs	Conc-lbs ai/a	Test Stat	Critical	MSD	DF	P-Type	P-Value	Decision(α:5%)		
Control		0.5*	58.3	2.02	0.046	5	CDF	<1.0E-05	Significant Effect		
ANOVA Table											
Source	Sum Squares		Mean Square		DF	F Stat	P-Value	Decision(α:5%)			
Between	5.22614		5.22614		1	3400	<1.0E-05	Significant Effect			
Error	0.0153593		0.0015359		10						
Total	5.2415				11						
ANOVA Assumptions Tests											
Attribute	Test		Test Stat	Critical	P-Value	Decision(α:1%)					
Variance	Variance Ratio F Test		8.65E+12	14.9	<1.0E-05	Unequal Variances					
Distribution	Shapiro-Wilk W Normality Test		0.828	0.802	0.0199	Normal Distribution					
24-hr Mortality Summary											
Conc-lbs ai/a	Code	Count	Mean	95% LCL	95% UCL	Median	Min	Max	Std Err	CV%	%Effect
0	00	6	0.020	0.000	0.043	0.020	0.000	0.040	0.009	109.54%	0.00%
0.5		6	1.000	1.000	1.000	1.000	1.000	1.000	0.000	0.00%	100.00%
Angular (Corrected) Transformed Summary											
Conc-lbs ai/a	Code	Count	Mean	95% LCL	95% UCL	Median	Min	Max	Std Err	CV%	%Effect
0	00	6	0.151	0.093	0.209	0.151	0.100	0.201	0.023	36.76%	100.00%
0.5		6	1.470	1.470	1.470	1.470	1.470	1.470	0.000	0.00%	10.25%
24-hr Mortality Detail											
Conc-lbs ai/a	Code	Rep 1	Rep 2	Rep 3	Rep 4	Rep 5	Rep 6				
0	00	0.000	0.000	0.040	0.000	0.040	0.040				
0.5		1.000	1.000	1.000	1.000	1.000	1.000				
Angular (Corrected) Transformed Detail											
Conc-lbs ai/a	Code	Rep 1	Rep 2	Rep 3	Rep 4	Rep 5	Rep 6				
0	00	0.100	0.100	0.201	0.100	0.201	0.201				
0.5		1.470	1.470	1.470	1.470	1.470	1.470				
24-hr Mortality Binomials											
Conc-lbs ai/a	Code	Rep 1	Rep 2	Rep 3	Rep 4	Rep 5	Rep 6				
0	00	0/25	0/25	1/25	0/25	1/25	1/25				
0.5		25/25	25/25	25/25	25/25	25/25	25/25				

001-771-848-3

CETIS™ v1.9.7.7

Analyst: QA: 



CETIS Analytical Report

Report Date: 04 Apr-22 17:14 (p 1 of 2)

Test Code/ID: LabB_E_T1_24h / 13-1487-7154

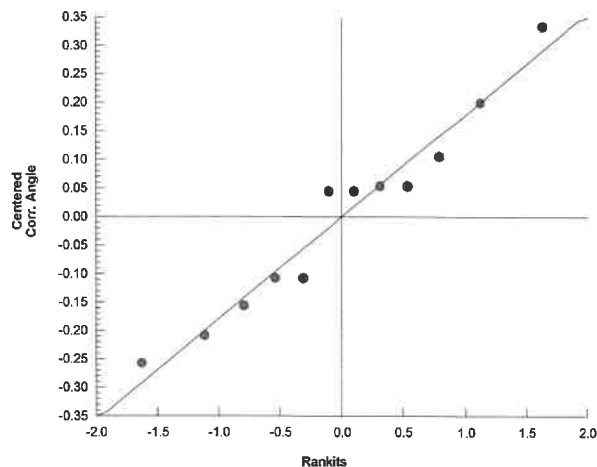
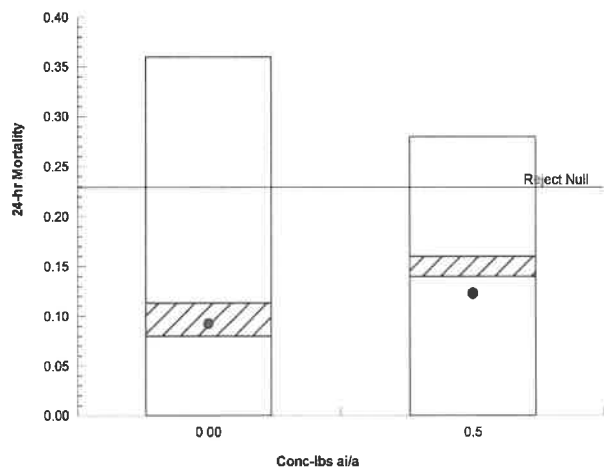
Foliar Acute Bee Test										B	
Analysis ID: 01-2053-7142		Endpoint: 24-hr Mortality		CETIS Version: CETISv1.9.7							
Analyzed: 04 Apr-22 17:14		Analysis: Parametric-Two Sample		Status Level: 1							
Edit Date: 04 Apr-22 17:14		MD5 Hash: 1F6D18CF061EE1BE5C51FDAFC84E52D		Editor ID: 001-771-848-3							
Batch ID: 13-4578-0981		Test Type: Acute Bee Survival		Analyst: Alison Briden							
Start Date: 10 Jun-21		Protocol: OCSPP 850.3030		Diluent: Not Applicable							
Ending Date: 11 Jun-21		Species: Apis Mellifera		Brine: Not Applicable							
Test Length: 24h		Taxon:		Source:		Age:					
Sample ID: 10-0009-8607		Code: LabB_E_T1_24h		Project: 36326							
Sample Date: 09 Jun-21		Material: Dimethoate		Source: Pacific EcoRisk							
Receipt Date: 09 Jun-21		CAS (PC):		Station: Lab B							
Sample Age: 24h		Client:									
Comments: Post-application interval: +24h Eurofins alfalfa, Trial 1											
Data Transform		Alt Hyp		Comparison Result				PMSD			
Angular (Corrected)		C < T		0.5lbs ai/a passed 24-hr mortality endpoint				13.06%			
Equal Variance t Two-Sample Test											
Control	vs	Conc-lbs ai/a	Test Stat	Critical	MSD	DF	P-Type	P-Value	Decision(α:5%)		
Control		0.5	0.464	1.81	0.19	10	CDF	0.3264	Non-Significant Effect		
ANOVA Table											
Source	Sum Squares		Mean Square		DF	F Stat	P-Value	Decision(α:5%)			
Between	0.0071023		0.0071023		1	0.215	0.6528	Non-Significant Effect			
Error	0.330422		0.0330422		10						
Total	0.337524				11						
ANOVA Assumptions Tests											
Attribute	Test		Test Stat	Critical	P-Value	Decision(α:1%)					
Variance	Variance Ratio F Test		1.24	14.9	0.8215	Equal Variances					
Distribution	Shapiro-Wilk W Normality Test		0.956	0.802	0.7300	Normal Distribution					
24-hr Mortality Summary											
Conc-lbs ai/a	Code	Count	Mean	95% LCL	95% UCL	Median	Min	Max	Std Err	CV%	%Effect
0	00	6	0.113	0.000	0.250	0.080	0.000	0.360	0.053	114.73%	0.00%
0.5		6	0.140	0.031	0.249	0.160	0.000	0.280	0.042	73.96%	3.01%
Angular (Corrected) Transformed Summary											
Conc-lbs ai/a	Code	Count	Mean	95% LCL	95% UCL	Median	Min	Max	Std Err	CV%	%Effect
0	00	6	0.309	0.108	0.510	0.278	0.100	0.644	0.078	61.86%	100.00%
0.5		6	0.358	0.177	0.538	0.412	0.100	0.558	0.070	48.07%	86.39%
24-hr Mortality Detail											
Conc-lbs ai/a	Code	Rep 1	Rep 2	Rep 3	Rep 4	Rep 5	Rep 6				
0	00	0.120	0.000	0.040	0.040	0.360	0.120				
0.5		0.000	0.040	0.200	0.160	0.280	0.160				
Angular (Corrected) Transformed Detail											
Conc-lbs ai/a	Code	Rep 1	Rep 2	Rep 3	Rep 4	Rep 5	Rep 6				
0	00	0.354	0.100	0.201	0.201	0.644	0.354				
0.5		0.100	0.201	0.464	0.412	0.558	0.412				
24-hr Mortality Binomials											
Conc-lbs ai/a	Code	Rep 1	Rep 2	Rep 3	Rep 4	Rep 5	Rep 6				
0	00	3/25	0/25	1/25	1/25	9/25	3/25				
0.5		0/25	1/25	5/25	4/25	7/25	4/25				

Foliar Acute Bee Test

B

Analysis ID: 01-2053-7142	Endpoint: 24-hr Mortality	CETIS Version: CETISv1.9.7
Analyzed: 04 Apr-22 17:14	Analysis: Parametric-Two Sample	Status Level: 1
Edit Date: 04 Apr-22 17:14	MD5 Hash: 1F6D18CF061EE1BE5C51FDAFC84E52D	Editor ID: 001-771-848-3

Graphics



CETIS Analytical Report

Report Date: 07 Apr-22 10:23 (p 1 of 1)
Test Code/ID: LabB_E_T1_RT25 / 12-7789-2849

Foliar Acute Bee Test										B	
Analysis ID: 12-4338-5529		Endpoint: 24-hr Mortality RT25			CETIS Version: CETISv1.9.7						
Analyzed: 07 Apr-22 10:23		Analysis: Linear Interpolation (ICPIN)			Status Level: 1						
Edit Date: 07 Apr-22 10:23		MD5 Hash: 5108EB43A43F62FFA6D4EA8E54E409D8			Editor ID: 001-771-848-3						
Batch ID: 12-3533-8156		Test Type: Acute Bee Survival			Analyst: Alison Briden						
Start Date: 09 Jun-21		Protocol: OCSP 850.3030			Diluent: Not Applicable						
Ending Date: 11 Jun-21		Species: Apis Mellifera			Brine: Not Applicable						
Test Length: 48h		Taxon:			Source: Age:						
Sample ID: 13-2271-2168		Code: LabB_E_T1_RT25			Project: 36326						
Sample Date: 09 Jun-21		Material: Dimethoate			Source: Pacific EcoRisk						
Receipt Date: 09 Jun-21		CAS (PC):			Station: Lab B						
Sample Age: ---		Client:									
Comments: RT25, Eurofins alfalfa, Trial 1											
Linear Interpolation Options											
X Transform	Y Transform	Seed	Resamples	Exp 95% CL	Method						
Linear	Linear	681219	1	Yes	Two-Point Interpolation						
Point Estimates											
Level	T-hrs	95% LCL	95% UCL								
IC10	7.9	---	---								
IC15	8.85	---	---								
IC20	9.8	---	---								
IC25	10.8	---	---								
IC40	13.6	---	---								
IC50	15.5	---	---								
24-hr Mortality RT25 Summary											
		Calculated Variate							Isotonic Variate		
T-hrs	Code	Count	Mean	Median	Min	Max	CV%	%Effect	Mean	%Effect	
0		1	100	100	100	100	---	---	100		
6		1	100	100	100	100	---	---	100		
24		1	5.3	5.3	5.3	5.3	---	---	5.3		
24-hr Mortality RT25 Detail											
T-hrs	Code	Rep 1									
0		100									
6		100									
24		5.3									
Graphics											

Test Item:	T=Dimethoate 400 EC Formulation	Application Date: 9 Jun 2021 @ 10:47
Bee Colony Used:	20-A-10	Treatment Rate: T=0.5 lb ai/Ac=560.4 g a.i./ha
		Crop: Alfalfa
* Corrected Mortality= (% T - % C)/(100 - % C) * 100		

Residual Timepoint:	6 Hours After Application
Harvest Time:	06/09/2021 @ 16:34
Exposure Time:	06/09/2021 @ C = 17:17, T= 17:22

	Date:		9-Jun-21	10-Jun-21	24 Hr.		
Treatment	Cage No.	No. Bees	Number of Dead Bees		Cumulative Total	% Cumulative Mortality	% Corrected Mortality
			≤4hr	24 hr			
C (Untreated Water Spray Alfalfa)	1	25	NR	0	3	2.0	NA
	2	25	NR	0			
	3	25	NR	1			
	4	25	NR	0			
	5	25	NR	1			
	6	25	NR	1			
Total		150	0	3			
% Cumulative Mortality			0.0	2.0			
T (Dimethoate 400 EC Treated Alfalfa)	1	25	NR	25	150	100.0	100
	2	25	NR	25			
	3	25	NR	25			
	4	25	NR	25			
	5	25	NR	25			
	6	25	NR	25			
Total		150	0	150			
% Cumulative Mortality			0.0	100.0			

2 affected bees @ 4-hour assessment

Residual Timepoint:	24 Hours After Application
Harvest Time:	06/10/2021 @ 10:34
Exposure Time:	06/10/2021 @ C = 11:22, T= 11:22

	Date:		10-Jun-21	11-Jun-21	24 Hr.		
Treatment	Cage No.	No. Bees	Number of Dead Bees		Cumulative Total	% Cumulative Mortality	% Corrected Mortality
			≤4hr	24 hr			
C (Untreated Water Spray Alfalfa)	1	25	0	3	17	11.3	NA
	2	25	0	0			
	3	25	0	1			
	4	25	0	1			
	5	25	0	9			
	6	25	0	3			
Total		150	0	17			
% Cumulative Mortality			0.0	11.3			
T (Dimethoate 400 EC Treated Alfalfa)	1	25	0	0	21	14.0	3
	2	25	0	1			
	3	25	0	5			
	4	25	0	4			
	5	25	0	7			
	6	25	0	4			
Total		150	0	21			
% Cumulative Mortality			0.0	14.0			

Appendix I

Summary of Statistics for the Toxicity of Facility A September Alfalfa Application Tested By Lab A



CETIS Analytical Report

 Report Date: 01 Apr-22 13:46 (p 1 of 2)
 Test Code/ID: LabA_S_T2_6h / 01-0531-2399

Foliar Acute Bee Test										A	
Analysis ID: 08-4598-8093		Endpoint: 24-hr Mortality		CETIS Version: CETISv1.9.7							
Analyzed: 01 Apr-22 13:46		Analysis: Parametric-Two Sample		Status Level: 1							
Edit Date: 01 Apr-22 13:45		MD5 Hash: 819999C83AD013A3B6E6BE91CA68FE6F		Editor ID: 001-771-848-3							
Batch ID: 09-3332-1075		Test Type: Acute Bee Survival		Analyst: Alison Briden							
Start Date: 16 Sep-21		Protocol: OCSP 850.3030		Diluent: Not Applicable							
Ending Date: 17 Sep-21		Species: Apis Mellifera		Brine: Not Applicable							
Test Length: 24h		Taxon:		Source:		Age:					
Sample ID: 01-6125-6495		Code: LabA_S_T2_6h		Project: 36326							
Sample Date: 16 Sep-21		Material: Dimethoate		Source: Pacific EcoRisk							
Receipt Date: 16 Sep-21		CAS (PC):		Station: Lab A							
Sample Age: ---		Client:									
Comments: Post-application interval: +6h Smithers alfalfa, Trial 2											
Data Transform		Alt Hyp		Comparison Result				PMSD			
Angular (Corrected)		C < T		0.5lbs ai/a passed 24-hr mortality endpoint				8.61%			
Equal Variance t Two-Sample Test											
Control	vs	Conc-lbs ai/a	Test Stat	Critical	MSD	DF	P-Type	P-Value	Decision(α:5%)		
Control		0.5	1.19	1.81	0.167	10	CDF	0.1315	Non-Significant Effect		
ANOVA Table											
Source	Sum Squares		Mean Square		DF	F Stat	P-Value	Decision(α:5%)			
Between	0.0356603		0.0356603		1	1.41	0.2629	Non-Significant Effect			
Error	0.253394		0.0253394		10						
Total	0.289054				11						
ANOVA Assumptions Tests											
Attribute	Test		Test Stat	Critical	P-Value	Decision(α:1%)					
Variance	Variance Ratio F Test		8.94	14.9	0.0312	Equal Variances					
Distribution	Shapiro-Wilk W Normality Test		0.836	0.802	0.0249	Normal Distribution					
24-hr Mortality Summary											
Conc-lbs ai/a	Code	Count	Mean	95% LCL	95% UCL	Median	Min	Max	Std Err	CV%	%Effect
0	00	6	0.033	0.002	0.065	0.040	0.000	0.080	0.012	90.33%	0.00%
0.5		6	0.107	0.000	0.263	0.080	0.000	0.400	0.061	139.64%	7.59%
Angular (Corrected) Transformed Summary											
Conc-lbs ai/a	Code	Count	Mean	95% LCL	95% UCL	Median	Min	Max	Std Err	CV%	%Effect
0	00	6	0.182	0.107	0.257	0.201	0.100	0.287	0.029	39.26%	100.00%
0.5		6	0.291	0.067	0.515	0.287	0.100	0.685	0.087	73.39%	62.52%
24-hr Mortality Detail											
Conc-lbs ai/a	Code	Rep 1	Rep 2	Rep 3	Rep 4	Rep 5	Rep 6				
0	00	0.000	0.040	0.080	0.000	0.040	0.040				
0.5		0.080	0.400	0.080	0.000	0.080	0.000				
Angular (Corrected) Transformed Detail											
Conc-lbs ai/a	Code	Rep 1	Rep 2	Rep 3	Rep 4	Rep 5	Rep 6				
0	00	0.100	0.201	0.287	0.100	0.201	0.201				
0.5		0.287	0.685	0.287	0.100	0.287	0.100				
24-hr Mortality Binomials											
Conc-lbs ai/a	Code	Rep 1	Rep 2	Rep 3	Rep 4	Rep 5	Rep 6				
0	00	0/25	1/25	2/25	0/25	1/25	1/25				
0.5		2/25	10/25	2/25	0/25	2/25	0/25				

CETIS Analytical Report

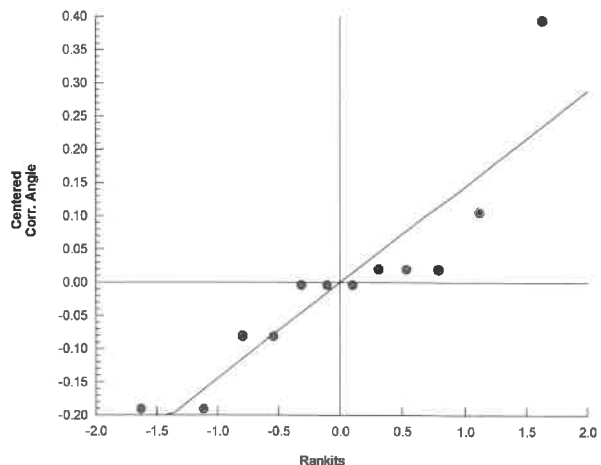
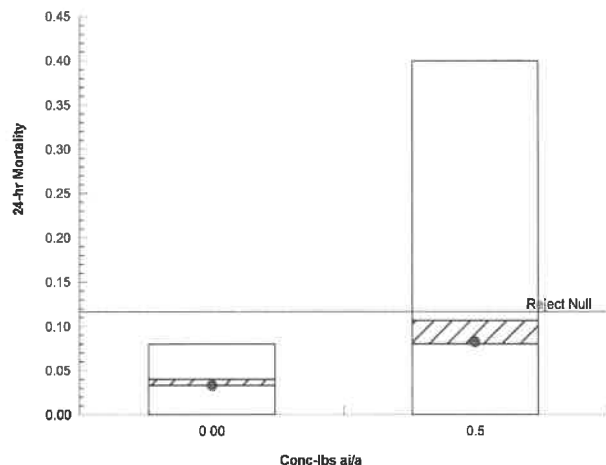
Report Date: 01 Apr-22 13:46 (p 2 of 2)
 Test Code/ID: LabA_S_T2_6h / 01-0531-2399

Foliar Acute Bee Test

A

Analysis ID: 08-4598-8093	Endpoint: 24-hr Mortality	CETIS Version: CETISv1.9.7
Analyzed: 01 Apr-22 13:46	Analysis: Parametric-Two Sample	Status Level: 1
Edit Date: 01 Apr-22 13:45	MD5 Hash: 819999C83AD013A3B6E6BE91CA68FE6F	Editor ID: 001-771-848-3

Graphics



CETIS Analytical Report

Report Date: 01 Apr-22 13:51 (p 1 of 2)
 Test Code/ID: LabA_S_T2_24h / 07-3950-6538

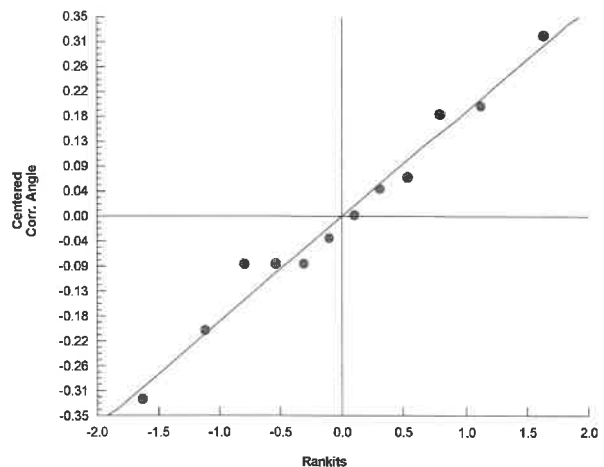
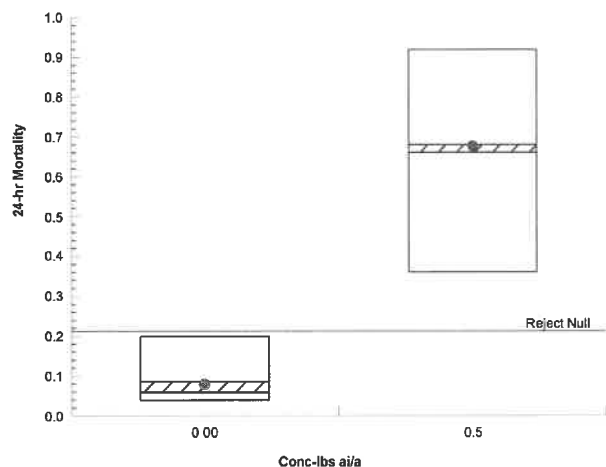
Foliar Acute Bee Test												A
Analysis ID: 00-3640-4168		Endpoint: 24-hr Mortality				CETIS Version: CETISv1.9.7						
Analyzed: 01 Apr-22 13:50		Analysis: Parametric-Two Sample				Status Level: 1						
Edit Date: 01 Apr-22 13:48		MD5 Hash: 0A914FD738A5F1BECDB2BE7319FACC9				Editor ID: 001-771-848-3						
Batch ID: 09-5301-9669		Test Type: Acute Bee Survival				Analyst: Alison Briden						
Start Date: 17 Sep-21		Protocol: OCSPP 850.3030				Diluent: Not Applicable						
Ending Date: 18 Sep-21		Species: Apis Mellifera				Brine: Not Applicable						
Test Length: 24h		Taxon:				Source: Age:						
Sample ID: 19-2685-8016		Code: LabA_S_T2_24h				Project: 36326						
Sample Date: 17 Sep-21		Material: Dimethoate				Source: Pacific EcoRisk						
Receipt Date: 17 Sep-21		CAS (PC):				Station: Lab A						
Sample Age: ---		Client:										
Comments: Post-application interval: +24h Smithers Alfalfa, Trial 2												
Data Transform		Alt Hyp		Comparison Result						PMSD		
Angular (Corrected)		C < T		0.5lbs ai/a failed 24-hr mortality endpoint						13.75%		
Equal Variance t Two-Sample Test												
Control	vs	Conc-lbs ai/a	Test Stat	Critical	MSD	DF	P-Type	P-Value	Decision(α:5%)			
Control		0.5*	6.36	1.81	0.194	10	CDF	4.1E-05	Significant Effect			
ANOVA Table												
Source	Sum Squares		Mean Square		DF	F Stat	P-Value	Decision(α:5%)				
Between	1.39025		1.39025		1	40.4	8.3E-05	Significant Effect				
Error	0.34411		0.034411		10							
Total	1.73436				11							
ANOVA Assumptions Tests												
Attribute	Test		Test Stat	Critical	P-Value	Decision(α:1%)						
Variance	Variance Ratio F Test		4.97	14.9	0.1031	Equal Variances						
Distribution	Shapiro-Wilk W Normality Test		0.98	0.802	0.9836	Normal Distribution						
24-hr Mortality Summary												
Conc-lbs ai/a	Code	Count	Mean	95% LCL	95% UCL	Median	Min	Max	Std Err	CV%	%Effect	
0	00	6	0.087	0.019	0.154	0.060	0.040	0.200	0.026	73.94%	0.00%	
0.5		6	0.660	0.437	0.883	0.680	0.360	0.920	0.087	32.24%	62.77%	
Angular (Corrected) Transformed Summary												
Conc-lbs ai/a	Code	Count	Mean	95% LCL	95% UCL	Median	Min	Max	Std Err	CV%	%Effect	
0	00	6	0.285	0.172	0.397	0.244	0.201	0.464	0.044	37.71%	100.00%	
0.5		6	0.965	0.714	1.220	0.970	0.644	1.280	0.098	24.79%	29.49%	
24-hr Mortality Detail												
Conc-lbs ai/a	Code	Rep 1	Rep 2	Rep 3	Rep 4	Rep 5	Rep 6					
0	00	0.040	0.080	0.120	0.200	0.040	0.040					
0.5		0.640	0.920	0.360	0.720	0.840	0.480					
Angular (Corrected) Transformed Detail												
Conc-lbs ai/a	Code	Rep 1	Rep 2	Rep 3	Rep 4	Rep 5	Rep 6					
0	00	0.201	0.287	0.354	0.464	0.201	0.201					
0.5		0.927	1.280	0.644	1.010	1.160	0.765					
24-hr Mortality Binomials												
Conc-lbs ai/a	Code	Rep 1	Rep 2	Rep 3	Rep 4	Rep 5	Rep 6					
0	00	1/25	2/25	3/25	5/25	1/25	1/25					
0.5		16/25	23/25	9/25	18/25	21/25	12/25					

Foliar Acute Bee Test

A

Analysis ID:	00-3640-4168	Endpoint:	24-hr Mortality	CETIS Version:	CETISv1.9.7
Analyzed:	01 Apr-22 13:50	Analysis:	Parametric-Two Sample	Status Level:	1
Edit Date:	01 Apr-22 13:48	MD5 Hash:	0A914FD738A5F1BECDB2BE7319FACC9	Editor ID:	001-771-848-3

Graphics



CETIS Analytical Report

Report Date: 01 Apr-22 13:55 (p 1 of 2)
Test Code/ID: LabA_S_T2_48h / 15-4302-0623

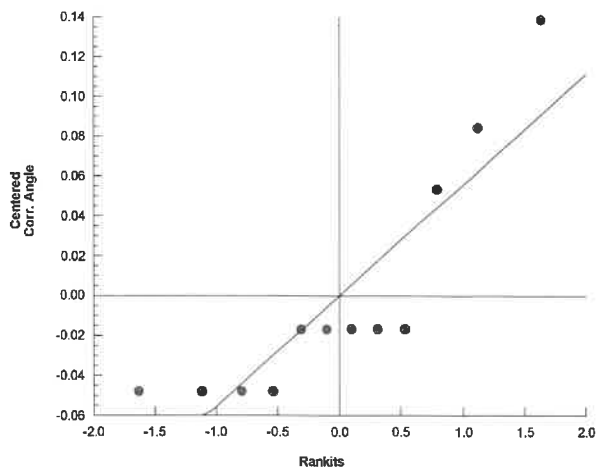
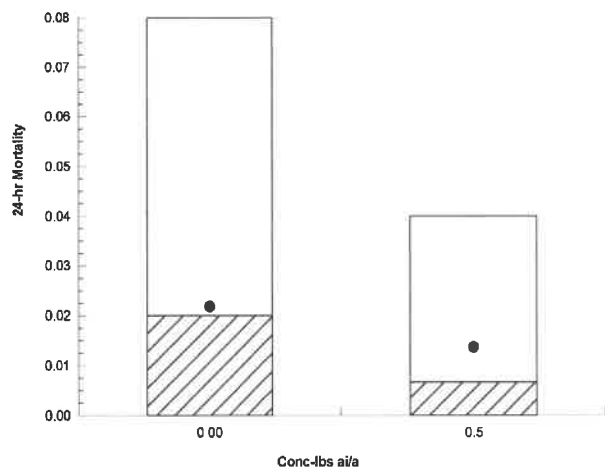
Foliar Acute Bee Test												A
Analysis ID: 17-2202-1259		Endpoint: 24-hr Mortality				CETIS Version: CETISv1.9.7						
Analyzed: 01 Apr-22 13:55		Analysis: Nonparametric-Two Sample				Status Level: 1						
Edit Date: 01 Apr-22 13:54		MD5 Hash: 20D5857939520302C21E45E1B120C2FC				Editor ID: 001-771-848-3						
Batch ID: 05-7471-4530		Test Type: Acute Bee Survival				Analyst: Alison Briden						
Start Date: 18 Sep-21		Protocol: OCSPP 850.3030				Diluent: Not Applicable						
Ending Date: 19 Sep-21		Species: Apis Mellifera				Brine: Not Applicable						
Test Length: 24h		Taxon:				Source: Age:						
Sample ID: 08-3860-0765		Code: LabA_S_T2_48h				Project: 36326						
Sample Date: 18 Sep-21		Material: Dimethoate				Source: Pacific EcoRisk						
Receipt Date: 18 Sep-21		CAS (PC):				Station: Lab A						
Sample Age: ---		Client:										
Comments: Post-application interval: +48h Smithers Alfalfa, Trial 1												
Data Transform		Alt Hyp		Comparison Result						PMSD		
Angular (Corrected)		C < T		0.5lbs ai/a passed 24-hr mortality endpoint						2.57%		
Wilcoxon Rank Sum Two-Sample Test												
Control	vs	Conc-lbs ai/a	Test Stat	Critical	Ties	DF	P-Type	P-Value	Decision(α:5%)			
Control		0.5	42.5	---	2	10	Exact	0.9091	Non-Significant Effect			
ANOVA Table												
Source	Sum Squares		Mean Square		DF	F Stat	P-Value	Decision(α:5%)				
Between	0.0029013		0.0029013		1	0.729	0.4131	Non-Significant Effect				
Error	0.0397851		0.0039785		10							
Total	0.0426864				11							
ANOVA Assumptions Tests												
Attribute	Test		Test Stat	Critical	P-Value	Decision(α:1%)						
Variance	Variance Ratio F Test		3.66	14.9	0.1806	Equal Variances						
Distribution	Shapiro-Wilk W Normality Test		0.768	0.802	0.0042	Non-Normal Distribution						
24-hr Mortality Summary												
Conc-lbs ai/a	Code	Count	Mean	95% LCL	95% UCL	Median	Min	Max	Std Err	CV%	%Effect	
0	00	6	0.020	0.000	0.055	0.000	0.000	0.080	0.014	167.33%	0.00%	
0.5		6	0.007	0.000	0.024	0.000	0.000	0.040	0.007	244.95%	-1.36%	
Angular (Corrected) Transformed Summary												
Conc-lbs ai/a	Code	Count	Mean	95% LCL	95% UCL	Median	Min	Max	Std Err	CV%	%Effect	
0	00	6	0.148	0.065	0.231	0.100	0.100	0.287	0.032	53.37%	100.00%	
0.5		6	0.117	0.074	0.160	0.100	0.100	0.201	0.017	35.30%	126.57%	
24-hr Mortality Detail												
Conc-lbs ai/a	Code	Rep 1	Rep 2	Rep 3	Rep 4	Rep 5	Rep 6					
0	00	0.000	0.000	0.080	0.000	0.040	0.000					
0.5		0.000	0.000	0.040	0.000	0.000	0.000					
Angular (Corrected) Transformed Detail												
Conc-lbs ai/a	Code	Rep 1	Rep 2	Rep 3	Rep 4	Rep 5	Rep 6					
0	00	0.100	0.100	0.287	0.100	0.201	0.100					
0.5		0.100	0.100	0.201	0.100	0.100	0.100					
24-hr Mortality Binomials												
Conc-lbs ai/a	Code	Rep 1	Rep 2	Rep 3	Rep 4	Rep 5	Rep 6					
0	00	0/25	0/25	2/25	0/25	1/25	0/25					
0.5		0/25	0/25	1/25	0/25	0/25	0/25					

Foliar Acute Bee Test

A

Analysis ID: 17-2202-1259	Endpoint: 24-hr Mortality	CETIS Version: CETISv1.9.7
Analyzed: 01 Apr-22 13:55	Analysis: Nonparametric-Two Sample	Status Level: 1
Edit Date: 01 Apr-22 13:54	MD5 Hash: 20D5857939520302C21E45E1B120C2FC	Editor ID: 001-771-848-3

Graphics



CETIS Analytical Report

Report Date: 07 Apr-22 09:56 (p 1 of 2)
 Test Code/ID: LabA_S_T2_RT25 / 20-8317-6789

Foliar Acute Bee Test						A				
Analysis ID: 15-5831-3519		Endpoint: 24-hr Mortality RT25		CETIS Version: CETISv1.9.7						
Analyzed: 07 Apr-22 9:56		Analysis: Linear Interpolation (ICPIN)		Status Level: 1						
Edit Date: 07 Apr-22 9:55		MD5 Hash: 0355172035B586CE18088DEBC81BE2CF		Editor ID: 001-771-848-3						
Batch ID: 01-5306-3882		Test Type: Acute Bee Survival		Analyst: Alison Briden						
Start Date: 16 Sep-21		Protocol: OCSPP 850.3030		Diluent: Not Applicable						
Ending Date: 19 Sep-21		Species: Apis Mellifera		Brine: Not Applicable						
Test Length: 72h		Taxon:		Source: Age:						
Sample ID: 04-3308-6674		Code: LabA_S_T2_RT25		Project: 36326						
Sample Date: 16 Sep-21		Material: Dimethoate		Source: Pacific EcoRisk						
Receipt Date: 16 Sep-21		CAS (PC):		Station: Lab A						
Sample Age: ---		Client:								
Comments: RT25, Smithers alfalfa, Trial 2										
Linear Interpolation Options										
X Transform	Y Transform	Seed	Resamples	Exp 95% CL	Method					
Linear	Linear	190361	1	Yes	Two-Point Interpolation					
Point Estimates										
Level	T-hrs	95% LCL	95% UCL							
IC10	1.41	---	---							
IC15	2.12	---	---							
IC20	2.82	---	---							
IC25	3.53	---	---							
IC40	5.65	---	---							
IC50	27.2	---	---							
24-hr Mortality RT25 Summary										
		Calculated Variate							Isotonic Variate	
T-hrs	Code	Count	Mean	Median	Min	Max	CV%	%Effect	Mean	%Effect
0		1	100	100	100	100	---	---	100	
6		1	16	16	16	16	---	---	57.5	
24		1	99	99	99	99	---	---	57.5	
48		1	1	1	1	1	---	---	1	
24-hr Mortality RT25 Detail										
T-hrs	Code	Rep 1								
0		100								
6		16								
24		99								
48		1								
Graphics										

Test Name:	T=Dimethoate 400 EC Formulation	Application Date:	14 Oct 2021 @ 10:06
Res Colony Used:	15A-12	Exposure Rate:	T = 0.5 lb ai/ha = 500.4 g a.i./ha
		Crop:	Alfalfa
* Corrected Mortality = $(N - T) \cdot \% C \cdot 100$			

Residual Timepoint: 6 Hours After Application
Harvest Time: 09/16/2021 @ 16:08
Exposure Time: 09/16/2021 @ 17:09

	Date:		16-Sep-21	17-Sep-21	24 Hr.		
Treatment	Cage No.	No. Bees	Number of Dead Bees		Cumulative Total	% Cumulative Mortality	% Corrected Mortality
			24hr	24 hr			
C (Untreated Water Spray Alfalfa)	1	25	0	0	5	3.3	NA
	2	25	0	1			
	3	25	0	2			
	4	25	0	0			
	5	25	0	1			
	6	25	0	1			
Total		150	0	5			
% Cumulative Mortality			0.0	3.3			
T (Dimethoate 400 EC Treated Alfalfa)	1	25	0	2	16	10.7	7.6
	2	25	0	10			
	3	25	0	2			
	4	25	0	0			
	5	25	0	2			
	6	25	0	0			
Total		150	0	16			
% Cumulative Mortality			0.0	10.7			

Residual Timepoint: 24 Hours After Application
Harvest Time: 09/17/2021, 1010
Exposure Time: 09/17/2021 @ 10:55

		Date:	17-Sep-21	18-Sep-21	24 Hr.		
Treatment	Cage No.	No. Bees	Number of Dead Bees		Cumulative Total	% Cumulative Mortality	% Corrected Mortality
			17hr	24 hr			
C (Untreated Water Spray Alfalfa)	1	25	0	1	13	8.7	NA
	2	25	0	2			
	3	25	2	3			
	4	25	0	5			
	5	25	0	1			
	6	25	1	1			
Total		150	3	13			
% Cumulative Mortality			2.0	8.7			
T (Dimethoate 400 EC Treated Alfalfa)	1	25	1	16	99	66.0	62.8
	2	25	4	23			
	3	25	2	9			
	4	25	0	18			
	5	25	2	21			
	6	25	0	12			
Total		150	9	69			
% Cumulative Mortality			6.0	46.0			

Residual Timepoint: 48 Hours After Application
Harvest Time: 09/18/2021, 1105
Exposure Time: 09/18/2021 @ 12:03

		Date:	18-Sep-21	19-Sep-21	24 Hr.		
Treatment	Cage No.	No. Bees	Number of Dead Bees		Cumulative Total	% Cumulative Mortality	% Corrected Mortality
			≤4hr	24 hr			
C (Untreated Water Spray Alfalfa)	1	25	0	0	3	2.0	NA
	2	25	0	0			
	3	25	0	2			
	4	25	0	0			
	5	25	0	1			
	6	25	0	0			
Total		150	0	3			
% Cumulative Mortality			0.0	2.0			
T (Dimethoate 400 EC Treated Alfalfa)	1	25	0	0	1	0.7	-8.8
	2	25	0	0			
	3	25	0	1			
	4	25	0	0			
	5	25	0	0			
	6	25	0	0			
Total		150	0	1			
% Cumulative Mortality			0.0	0.7			

Post-application interval: +6hr	≤ 4 - Hour Observations		24 - Hour Observations	
Location: Eurofins	Test Concentration		Test Concentration	
Replicate	Control	T1	Control	T1
	Observation	Observation	Observation	Observation
1	All N	All N	All N	All dead
2	All N	All N	All N	All dead
3	All N	All N	All N	All dead
4	All N	All N	All N	All dead
5	All N	All N	All N	All dead
6	All N	All N	All N	All dead
Total	All N	All N	All N	All dead

Note: bees appear to be having a somewhat more difficult time climbing wall of cages than normal recorded 15 Sep 2021 by AW

Post-application interval: +24hr	≤ 4 - Hour Observations		24 - Hour Observations	
Location: Eurofins	Test Concentration		Test Concentration	
Replicate	Control	T1	Control	T1
	Observation	Observation	Observation	Observation
1	All N	All N	All N	All N
2	All N	All N	All N	All N
3	All N	All N	All N	All N
4	All N	All N	All N	All N
5	All N	All N	All N	All N
6	All N	All N	All N	All N
Total	All N	All N	All N	All N

Post-application interval: +6hr	≤ 4 - Hour Observations	24 - Hour Observations
---------------------------------	-------------------------	------------------------

Location: Smithers	Test Concentration		Test Concentration	
Replicate	Control	T1	Control	T1
	Observation	Observation	Observation	Observation
1	All N	All N	All N	All N
2	All N	All N	All N	All N
3	All N	All N	All N	All N
4	All N	All N	All N	All N
5	All N	All N	All N	All N
6	All N	All N	All N	All N
Total	All N	All N	All N	All N

Post-application interval: +24hr	≤ 4 - Hour Observations		24 - Hour Observations	
Location: Smithers	Test Concentration		Test Concentration	
Replicate	Control	T1	Control	T1
	Observation	Observation	Observation	Observation
1	All N	All N	All N	All N
2	All N	All N	All N	All N
3	All N	All N	All N	All N
4	All N	All N	All N	All N
5	All N	All N	All N	All N
6	All N	All N	All N	All N
Total	All N	All N	All N	All N

Post-application interval: +48hr	≤ 4 - Hour Observations	24 - Hour Observations
Location: Smithers	Test Concentration	Test Concentration

Appendix J

Summary of Statistics for the Toxicity of Facility A September Alfalfa Application Tested By Lab B



CETIS Analytical Report

Report Date: 04 Apr-22 17:19 (p 1 of 2)
Test Code/ID: LabB_S_T2_6h / 12-5140-4977

Foliar Acute Bee Test											B	
Analysis ID: 00-7902-9565			Endpoint: 24-hr Mortality			CETIS Version: CETISv1.9.7						
Analyzed: 04 Apr-22 17:19			Analysis: Parametric-Two Sample			Status Level: 1						
Edit Date: 04 Apr-22 17:19			MD5 Hash: E86B695062C644847B33B2E439A71F9F			Editor ID: 001-771-848-3						
Batch ID: 12-0671-8943			Test Type: Acute Bee Survival			Analyst: Alison Briden			Age:			
Start Date: 16 Sep-21			Protocol: OCSPP 850.3030			Diluent: Not Applicable						
Ending Date: 17 Sep-21			Species: Apis Mellifera			Brine: Not Applicable						
Test Length: 24h			Taxon:			Source:						
Sample ID: 05-2378-3786			Code: LabB_S_T2_6h			Project: 36326						
Sample Date: 16 Sep-21			Material: Dimethoate			Source: Pacific EcoRisk						
Receipt Date: 16 Sep-21			CAS (PC):			Station: Lab B						
Sample Age: ---			Client:									
Comments: Post-application interval: +6h Smithers alfalfa, Trial 2												
Data Transform		Alt Hyp		Comparison Result						PMSD		
Angular (Corrected)		C < T		0.5lbs ai/a failed 24-hr mortality endpoint						4.86%		
Equal Variance t Two-Sample Test												
Control	vs	Conc-lbs ai/a	Test Stat	Critical	MSD	DF	P-Type	P-Value	Decision(α:5%)			
Control		0.5*	8.02	1.81	0.111	10	CDF	<1.0E-05	Significant Effect			
ANOVA Table												
Source	Sum Squares		Mean Square		DF	F Stat	P-Value	Decision(α:5%)				
Between	0.717998		0.717998		1	64.4	1.1E-05	Significant Effect				
Error	0.111531		0.0111531		10							
Total	0.829529				11							
ANOVA Assumptions Tests												
Attribute	Test		Test Stat	Critical	P-Value	Decision(α:1%)						
Variance	Variance Ratio F Test		2.71	14.9	0.2983	Equal Variances						
Distribution	Shapiro-Wilk W Normality Test		0.937	0.802	0.4568	Normal Distribution						
24-hr Mortality Summary												
Conc-lbs ai/a	Code	Count	Mean	95% LCL	95% UCL	Median	Min	Max	Std Err	CV%	%Effect	
0	00	6	0.027	0.000	0.061	0.020	0.000	0.080	0.013	122.47%	0.00%	
0.5		6	0.373	0.244	0.502	0.340	0.240	0.560	0.050	32.97%	35.62%	
Angular (Corrected) Transformed Summary												
Conc-lbs ai/a	Code	Count	Mean	95% LCL	95% UCL	Median	Min	Max	Std Err	CV%	%Effect	
0	00	6	0.165	0.084	0.246	0.151	0.100	0.287	0.032	47.01%	100.00%	
0.5		6	0.654	0.520	0.788	0.622	0.512	0.846	0.052	19.51%	25.22%	
24-hr Mortality Detail												
Conc-lbs ai/a	Code	Rep 1	Rep 2	Rep 3	Rep 4	Rep 5	Rep 6					
0	00	0.000	0.000	0.080	0.040	0.000	0.040					
0.5		0.560	0.280	0.320	0.480	0.240	0.360					
Angular (Corrected) Transformed Detail												
Conc-lbs ai/a	Code	Rep 1	Rep 2	Rep 3	Rep 4	Rep 5	Rep 6					
0	00	0.100	0.100	0.287	0.201	0.100	0.201					
0.5		0.846	0.558	0.601	0.765	0.512	0.644					
24-hr Mortality Binomials												
Conc-lbs ai/a	Code	Rep 1	Rep 2	Rep 3	Rep 4	Rep 5	Rep 6					
0	00	0/25	0/25	2/25	1/25	0/25	1/25					
0.5		14/25	7/25	8/25	12/25	6/25	9/25					

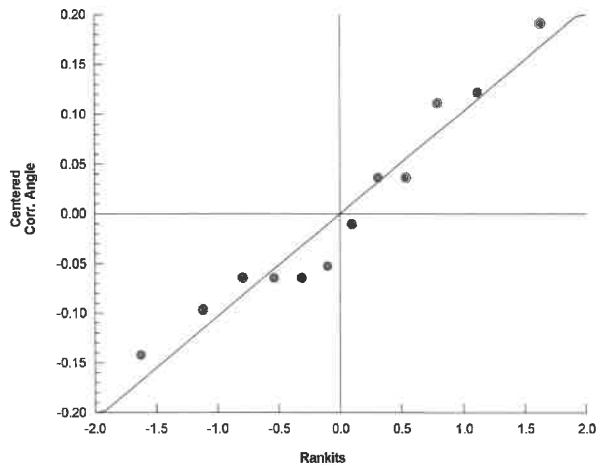
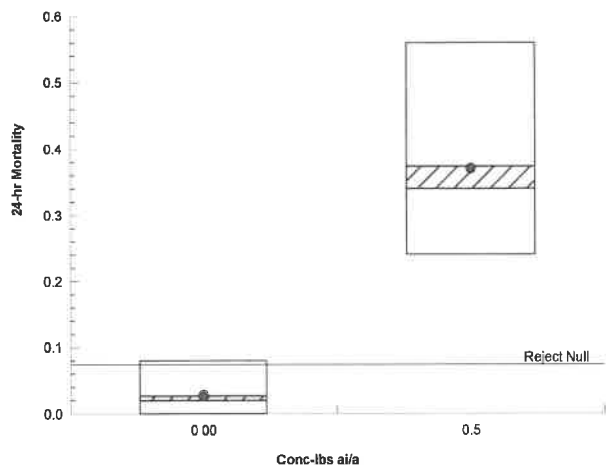
Foliar Acute Bee Test

B

Analysis ID: 00-7902-9565 Endpoint: 24-hr Mortality
 Analyzed: 04 Apr-22 17:19 Analysis: Parametric-Two Sample
 Edit Date: 04 Apr-22 17:19 MD5 Hash: E86B695062C644847B33B2E439A71F9F

CETIS Version: CETISv1.9.7
 Status Level: 1
 Editor ID: 001-771-848-3

Graphics



CETIS Analytical Report

 Report Date: 06 Apr-22 10:49 (p 1 of 2)
 Test Code/ID: LabB_S_T2_24h / 20-5545-6423

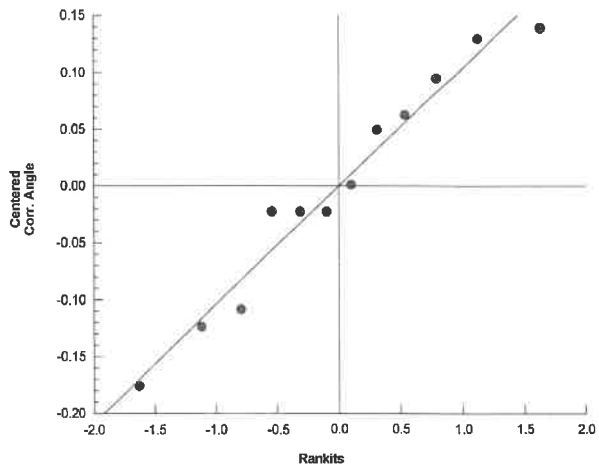
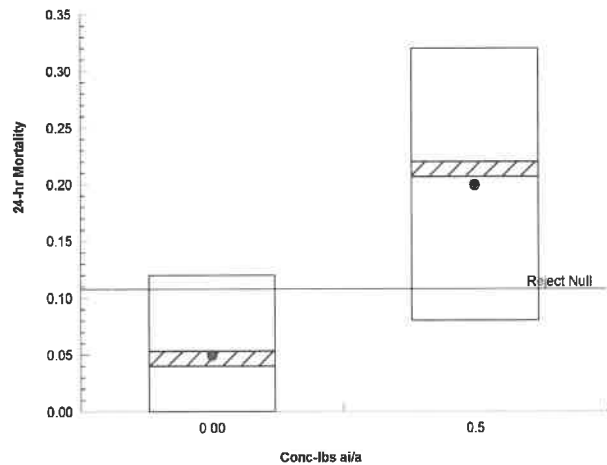
Foliar Acute Bee Test										B	
Analysis ID: 06-6036-1153		Endpoint: 24-hr Mortality		CETIS Version: CETISv1.9.7							
Analyzed: 04 Apr-22 17:21		Analysis: Parametric-Two Sample		Status Level: 1							
Edit Date: 04 Apr-22 17:21		MD5 Hash: 403CC6BCD4CE37EF53935BC9497677CF		Editor ID: 001-771-848-3							
Batch ID: 15-4550-7016		Test Type: Acute Bee Survival		Analyst: Alison Briden							
Start Date: 17 Sep-21		Protocol: OCSPP 850.3030		Diluent: Not Applicable							
Ending Date: 18 Sep-21		Species: Apis Mellifera		Brine: Not Applicable							
Test Length: 24h		Taxon:		Source:		Age:					
Sample ID: 03-5892-4841		Code: LabB_S_T2_24h		Project: 36326							
Sample Date: 16 Sep-21		Material: Dimethoate		Source: Pacific EcoRisk							
Receipt Date: 16 Sep-21		CAS (PC):		Station: Lab B							
Sample Age: 24h		Client:									
Comments: Post-application interval: +24h Smithers alfalfa, Trial 2											
Data Transform		Alt Hyp		Comparison Result				PMSD			
Angular (Corrected)		C < T		0.5lbs ai/a failed 24-hr mortality endpoint				5.75%			
Equal Variance t Two-Sample Test											
Control	vs	Conc-lbs ai/a	Test Stat	Critical	MSD	DF	P-Type	P-Value	Decision(α:5%)		
Control		0.5*	3.92	1.81	0.11	10	CDF	0.0014	Significant Effect		
ANOVA Table											
Source	Sum Squares		Mean Square		DF	F Stat	P-Value	Decision(α:5%)			
Between	0.170466		0.170466		1	15.3	0.0029	Significant Effect			
Error	0.111106		0.0111106		10						
Total	0.281572				11						
ANOVA Assumptions Tests											
Attribute	Test		Test Stat	Critical	P-Value	Decision(α:1%)					
Variance	Variance Ratio F Test		1.95	14.9	0.4807	Equal Variances					
Distribution	Shapiro-Wilk W Normality Test		0.952	0.802	0.6715	Normal Distribution					
24-hr Mortality Summary											
Conc-lbs ai/a	Code	Count	Mean	95% LCL	95% UCL	Median	Min	Max	Std Err	CV%	%Effect
0	00	6	0.053	0.010	0.097	0.040	0.000	0.120	0.017	77.46%	0.00%
0.5		6	0.207	0.109	0.304	0.220	0.080	0.320	0.038	44.84%	16.20%
Angular (Corrected) Transformed Summary											
Conc-lbs ai/a	Code	Count	Mean	95% LCL	95% UCL	Median	Min	Max	Std Err	CV%	%Effect
0	00	6	0.224	0.133	0.315	0.201	0.100	0.354	0.035	38.71%	100.00%
0.5		6	0.462	0.335	0.590	0.488	0.287	0.601	0.050	26.21%	48.46%
24-hr Mortality Detail											
Conc-lbs ai/a	Code	Rep 1	Rep 2	Rep 3	Rep 4	Rep 5	Rep 6				
0	00	0.120	0.040	0.000	0.040	0.080	0.040				
0.5		0.280	0.080	0.200	0.120	0.240	0.320				
Angular (Corrected) Transformed Detail											
Conc-lbs ai/a	Code	Rep 1	Rep 2	Rep 3	Rep 4	Rep 5	Rep 6				
0	00	0.354	0.201	0.100	0.201	0.287	0.201				
0.5		0.558	0.287	0.464	0.354	0.512	0.601				
24-hr Mortality Binomials											
Conc-lbs ai/a	Code	Rep 1	Rep 2	Rep 3	Rep 4	Rep 5	Rep 6				
0	00	3/25	1/25	0/25	1/25	2/25	1/25				
0.5		7/25	2/25	5/25	3/25	6/25	8/25				

Foliar Acute Bee Test

B

Analysis ID: 06-6036-1153	Endpoint: 24-hr Mortality	CETIS Version: CETISv1.9.7
Analyzed: 04 Apr-22 17:21	Analysis: Parametric-Two Sample	Status Level: 1
Edit Date: 04 Apr-22 17:21	MD5 Hash: 403CC6BCD4CE37EF53935BC9497677CF	Editor ID: 001-771-848-3

Graphics



CETIS Analytical Report

Report Date: 07 Apr-22 09:58 (p 1 of 1)
Test Code/ID: LabB_S_T2_RT25 / 18-8342-4814

Foliar Acute Bee Test										B	
Analysis ID: 08-3804-5558		Endpoint: 24-hr Mortality RT25			CETIS Version: CETISv1.9.7						
Analyzed: 07 Apr-22 9:58		Analysis: Linear Interpolation (ICPIN)			Status Level: 1						
Edit Date: 07 Apr-22 9:57		MD5 Hash: 9C86420B687DBD02C612A5783D8DFEBE			Editor ID: 001-771-848-3						
Batch ID: 17-6057-6551		Test Type: Acute Bee Survival			Analyst: Alison Briden						
Start Date: 16 Sep-21		Protocol: OCSP 850.3030			Diluent: Not Applicable						
Ending Date: 18 Sep-21		Species: Apis Mellifera			Brine: Not Applicable						
Test Length: 48h		Taxon:			Source: Age:						
Sample ID: 01-7224-3844		Code: LabB_S_T2_RT25			Project: 36326						
Sample Date: 16 Sep-21		Material: Dimethoate			Source: Pacific EcoRisk						
Receipt Date: 16 Sep-21		CAS (PC):			Station: Lab B						
Sample Age: ---		Client:									
Comments: RT25, Smithers alfalfa, Trial 2											
Linear Interpolation Options											
X Transform	Y Transform	Seed	Resamples	Exp 95% CL	Method						
Linear	Linear	1714822	1	Yes	Two-Point Interpolation						
Point Estimates											
Level	T-hrs	95% LCL	95% UCL								
IC10	0.932	---	---								
IC15	1.4	---	---								
IC20	1.86	---	---								
IC25	2.33	---	---								
IC40	3.73	---	---								
IC50	4.66	---	---								
24-hr Mortality RT25 Summary											
		Calculated Variate							Isotonic Variate		
T-hrs	Code	Count	Mean	Median	Min	Max	CV%	%Effect	Mean	%Effect	
0		1	100	100	100	100	---	---	100		
6		1	35.6	35.6	35.6	35.6	---	---	35.6		
24		1	16.2	16.2	16.2	16.2	---	---	16.2		
24-hr Mortality RT25 Detail											
T-hrs	Code	Rep 1									
0		100									
6		35.6									
24		16.2									
Graphics											

Test Item:	T = Dimethoate 400 EC Formulation	Application Date: 16 Sept 2021 @ 10:08
Bee Colony Used:	20-A-10	Treatment Rate: T = 0.5 lb ai/Ac = 560.4 g a.i./ha
		Crop: Alfalfa
* Corrected Mortality= (% T - % C)/(100 - % C) * 100		

Residual Timepoint:	6 Hours After Application
Harvest Time:	09/16/2021 @ 16:08
Exposure Time:	09/16/2021 @ C = 17:32, T=17:40

	Date:		16-Sep-21	17-Sep-21	24 Hr.		
Treatment	Cage No.	No. Bees	Number of Dead Bees		Cumulative Total	% Cumulative Mortality	% Corrected Mortality
			≤4hr	24 hr			
C (Untreated Water Spray Alfalfa)	1	25	NR	0	4	2.7	NA
	2	25	NR	0			
	3	25	NR	2			
	4	25	NR	1			
	5	25	NR	0			
	6	25	NR	1			
Total		150	0	4			
% Cumulative Mortality			0.0	2.7			
T (Dimethoate 400 EC Treated Alfalfa)	1	25	NR	14	56	37.3	35.6
	2	25	NR	7			
	3	25	NR	8			
	4	25	NR	12			
	5	25	NR	6			
	6	25	NR	9			
Total		150	0	56			
% Cumulative Mortality			0.0	37.3			

3 moribund @ 24 hour assessment
2 moribund @ 24 hour assessment
1 moribund, 3 affected @ 24 hour assessment
2 moribund @ 24 hour assessment
3 moribund, 1 affected @ 24 hour assessment
4 moribund @ 24 hour assessment

Residual Timepoint:	24 Hours After Application
Harvest Time:	09/17/2021, 1010
Exposure Time:	09/17/2021 @ C = 11:32, T=11:38

	Date:		17-Sep-21	18-Sep-21	24 Hr.		
Treatment	Cage No.	No. Bees	Number of Dead Bees		Cumulative Total	% Cumulative Mortality	% Corrected Mortality
			≤4hr	24 hr			
C (Untreated Water Spray Alfalfa)	1	25	0	3	8	5.3	NA
	2	25	0	1			
	3	25	0	0			
	4	25	0	1			
	5	25	0	2			
	6	25	0	1			
Total		150	0	8			
% Cumulative Mortality			0.0	5.3			
T (Dimethoate 400 EC Treated Alfalfa)	1	25	0	7	31	20.7	16.2
	2	25	0	2			
	3	25	1	5			
	4	25	0	3			
	5	25	1	6			
	6	25	1	8			
Total		150	3	31			
% Cumulative Mortality			2.0	20.7			

1 moribund @ 24 hours
1 affected at 4 hours, 3 affected at 24 hours
3 affected at 4 Hours, 6 affected at 24 hours
4 affected, 1 moribund @ 24 hours
2 affected @ 4 hours, 2 moribund @ 24 hours
2 affected, 1 moribund @ 4 hours, 2 affected at 24 hours

Appendix K

Summary of Statistics for the Toxicity of Facility B September Alfalfa Application Tested By Lab A



CETIS Analytical Report

Report Date: 01 Apr-22 14:00 (p 1 of 2)
 Test Code/ID: LabA_E_T2_6h / 11-4783-4464

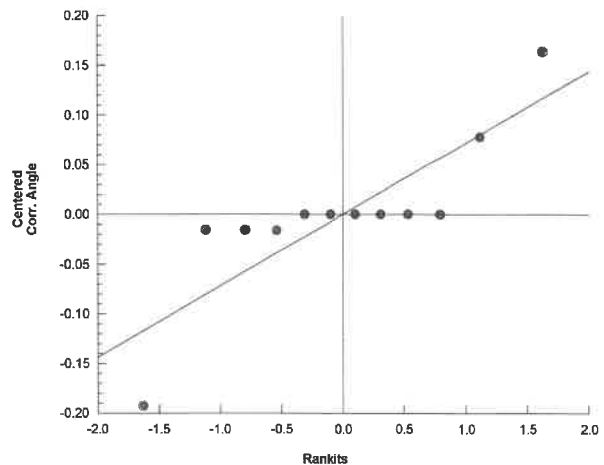
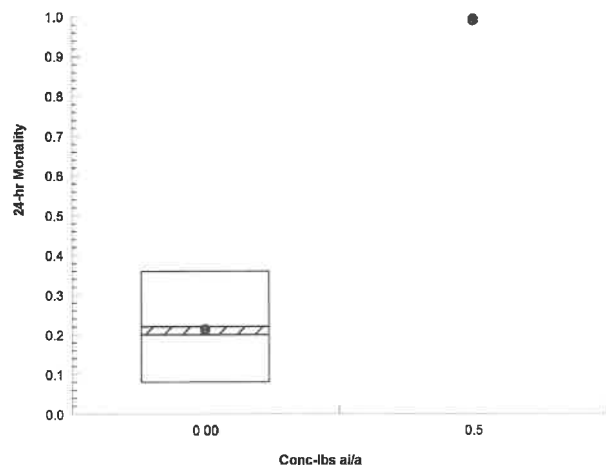
Foliar Acute Bee Test										A	
Analysis ID: 11-3370-3725		Endpoint: 24-hr Mortality		CETIS Version: CETISv1.9.7							
Analyzed: 01 Apr-22 13:59		Analysis: Nonparametric-Two Sample		Status Level: 1							
Edit Date: 01 Apr-22 13:58		MD5 Hash: D5338866BD7D21A5F905562C8DF82FF7		Editor ID: 001-771-848-3							
Batch ID: 20-2884-9407		Test Type: Acute Bee Survival		Analyst: Alison Briden							
Start Date: 14 Sep-21		Protocol: OCSP 850.3030		Diluent: Not Applicable							
Ending Date: 15 Sep-21		Species: Apis Mellifera		Brine: Not Applicable							
Test Length: 24h		Taxon:		Source:		Age:					
Sample ID: 16-0872-9766		Code: LabA_E_T2_6h		Project: 36326							
Sample Date: 14 Sep-21		Material: Dimethoate		Source: Pacific EcoRisk							
Receipt Date: 14 Sep-21		CAS (PC):		Station: Lab A							
Sample Age: ---		Client:									
Comments: Post-application interval: +6h Eurofins Alfalfa, Trial 2											
Data Transform		Alt Hyp		Comparison Result				PMSD			
Angular (Corrected)		C < T		0.5lbs ai/a failed 24-hr mortality endpoint				8.89%			
Wilcoxon Rank Sum Two-Sample Test											
Control	vs	Conc-lbs ai/a	Test Stat	Critical	Ties	DF	P-Type	P-Value	Decision(α:5%)		
Control		0.5*	21	---	0	10	Exact	0.0011	Significant Effect		
ANOVA Table											
Source	Sum Squares		Mean Square		DF	F Stat	P-Value	Decision(α:5%)			
Between	2.94523		2.94523		1	415	<1.0E-05	Significant Effect			
Error	0.0708992		0.0070899		10						
Total	3.01613				11						
ANOVA Assumptions Tests											
Attribute	Test		Test Stat	Critical	P-Value	Decision(α:1%)					
Variance	Variance Ratio F Test		3.99E+13	14.9	<1.0E-05	Unequal Variances					
Distribution	Shapiro-Wilk W Normality Test		0.765	0.802	0.0039	Non-Normal Distribution					
24-hr Mortality Summary											
Conc-lbs ai/a	Code	Count	Mean	95% LCL	95% UCL	Median	Min	Max	Std Err	CV%	%Effect
0	00	6	0.220	0.122	0.318	0.200	0.080	0.360	0.038	42.64%	0.00%
0.5		6	1.000	1.000	1.000	1.000	1.000	1.000	0.000	0.00%	100.00%
Angular (Corrected) Transformed Summary											
Conc-lbs ai/a	Code	Count	Mean	95% LCL	95% UCL	Median	Min	Max	Std Err	CV%	%Effect
0	00	6	0.480	0.355	0.605	0.464	0.287	0.644	0.049	24.82%	100.00%
0.5		6	1.470	1.470	1.470	1.470	1.470	1.470	0.000	0.00%	32.63%
24-hr Mortality Detail											
Conc-lbs ai/a	Code	Rep 1	Rep 2	Rep 3	Rep 4	Rep 5	Rep 6				
0	00	0.080	0.200	0.360	0.280	0.200	0.200				
0.5		1.000	1.000	1.000	1.000	1.000	1.000				
Angular (Corrected) Transformed Detail											
Conc-lbs ai/a	Code	Rep 1	Rep 2	Rep 3	Rep 4	Rep 5	Rep 6				
0	00	0.287	0.464	0.644	0.558	0.464	0.464				
0.5		1.470	1.470	1.470	1.470	1.470	1.470				
24-hr Mortality Binomials											
Conc-lbs ai/a	Code	Rep 1	Rep 2	Rep 3	Rep 4	Rep 5	Rep 6				
0	00	2/25	5/25	9/25	7/25	5/25	5/25				
0.5		25/25	25/25	25/25	25/25	25/25	25/25				

Foliar Acute Bee Test

A

Analysis ID: 11-3370-3725
Analyzed: 01 Apr-22 13:59
Edit Date: 01 Apr-22 13:58Endpoint: 24-hr Mortality
Analysis: Nonparametric-Two Sample
MD5 Hash: D5338866BD7D21A5F905562C8DF82FF7CETIS Version: CETISv1.9.7
Status Level: 1
Editor ID: 001-771-848-3

Graphics



CETIS Analytical Report

 Report Date: 01 Apr-22 14:03 (p 1 of 2)
 Test Code/ID: LabA_E_T2_24h / 05-4092-5307

Foliar Acute Bee Test										A	
Analysis ID: 03-6045-0243		Endpoint: 24-hr Mortality		CETIS Version: CETISv1.9.7							
Analyzed: 01 Apr-22 14:03		Analysis: Parametric-Two Sample		Status Level: 1							
Edit Date: 01 Apr-22 14:02		MD5 Hash: CA25446150CCE6AA1F8AAEE8A900BA02		Editor ID: 001-771-848-3							
Batch ID: 07-6888-2474		Test Type: Acute Bee Survival		Analyst: Alison Briden							
Start Date: 15 Sep-21		Protocol: OCSPP 850.3030		Diluent: Not Applicable							
Ending Date: 16 Sep-21		Species: Apis Mellifera		Brine: Not Applicable							
Test Length: 24h		Taxon:		Source:		Age:					
Sample ID: 05-3681-6480		Code: LabA_E_T2_24h		Project: 36326							
Sample Date: 15 Sep-21		Material: Dimethoate		Source: Pacific EcoRisk							
Receipt Date: 15 Sep-21		CAS (PC):		Station: Lab A							
Sample Age: ---		Client:									
Comments: Post-application interval: +24h Eurofins Alfalfa, Trial 2											
Data Transform		Alt Hyp		Comparison Result						PMSD	
Angular (Corrected)		C < T		0.5lbs ai/a passed 24-hr mortality endpoint						6.04%	
Equal Variance t Two-Sample Test											
Control	vs	Conc-lbs ai/a	Test Stat	Critical	MSD	DF	P-Type	P-Value	Decision(α:5%)		
Control		0.5	-1.23	1.81	0.103	10	CDF	0.8767	Non-Significant Effect		
ANOVA Table											
Source	Sum Squares		Mean Square		DF	F Stat	P-Value	Decision(α:5%)			
Between	0.0147223		0.0147223		1	1.52	0.2465	Non-Significant Effect			
Error	0.0971724		0.0097172		10						
Total	0.111895				11						
ANOVA Assumptions Tests											
Attribute	Test		Test Stat	Critical	P-Value	Decision(α:1%)					
Variance	Variance Ratio F Test		3.05	14.9	0.2470	Equal Variances					
Distribution	Shapiro-Wilk W Normality Test		0.942	0.802	0.5289	Normal Distribution					
24-hr Mortality Summary											
Conc-lbs ai/a	Code	Count	Mean	95% LCL	95% UCL	Median	Min	Max	Std Err	CV%	%Effect
0	00	6	0.087	0.019	0.154	0.080	0.000	0.160	0.026	73.94%	0.00%
0.5		6	0.047	0.015	0.078	0.040	0.000	0.080	0.012	64.52%	-4.38%
Angular (Corrected) Transformed Summary											
Conc-lbs ai/a	Code	Count	Mean	95% LCL	95% UCL	Median	Min	Max	Std Err	CV%	%Effect
0	00	6	0.283	0.156	0.410	0.287	0.100	0.412	0.049	42.74%	100.00%
0.5		6	0.213	0.140	0.286	0.201	0.100	0.287	0.028	32.55%	132.90%
24-hr Mortality Detail											
Conc-lbs ai/a	Code	Rep 1	Rep 2	Rep 3	Rep 4	Rep 5	Rep 6				
0	00	0.080	0.160	0.080	0.040	0.000	0.160				
0.5		0.000	0.040	0.080	0.040	0.040	0.080				
Angular (Corrected) Transformed Detail											
Conc-lbs ai/a	Code	Rep 1	Rep 2	Rep 3	Rep 4	Rep 5	Rep 6				
0	00	0.287	0.412	0.287	0.201	0.100	0.412				
0.5		0.100	0.201	0.287	0.201	0.201	0.287				
24-hr Mortality Binomials											
Conc-lbs ai/a	Code	Rep 1	Rep 2	Rep 3	Rep 4	Rep 5	Rep 6				
0	00	2/25	4/25	2/25	1/25	0/25	4/25				
0.5		0/25	1/25	2/25	1/25	1/25	2/25				

CETIS Analytical Report

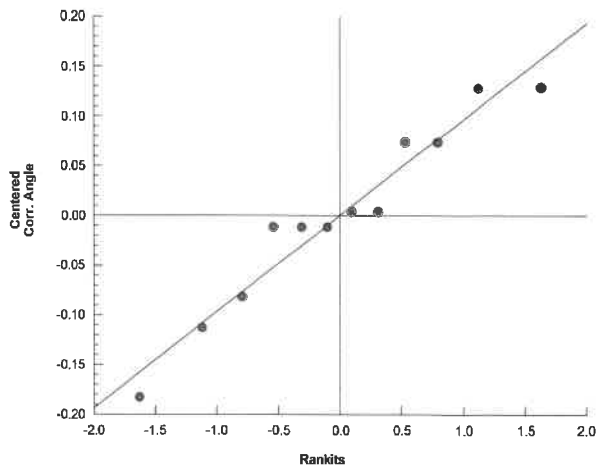
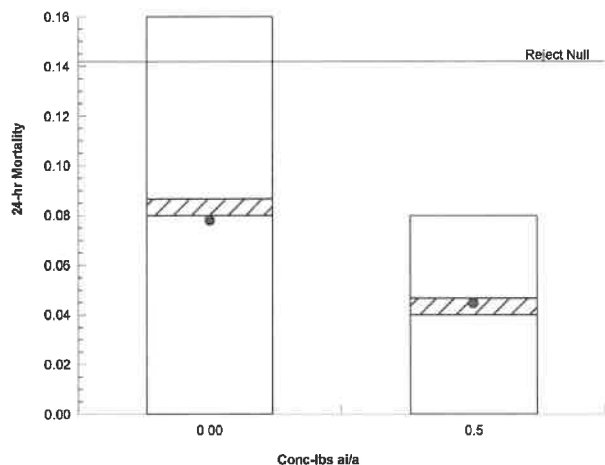
Report Date: 01 Apr-22 14:03 (p 2 of 2)
 Test Code/ID: LabA_E_T2_24h / 05-4092-5307

Foliar Acute Bee Test

A

Analysis ID: 03-6045-0243	Endpoint: 24-hr Mortality	CETIS Version: CETISv1.9.7
Analyzed: 01 Apr-22 14:03	Analysis: Parametric-Two Sample	Status Level: 1
Edit Date: 01 Apr-22 14:02	MD5 Hash: CA25446150CCE6AA1F8AAEE8A900BA02	Editor ID: 001-771-848-3

Graphics



CETIS Analytical Report

Report Date: 07 Apr-22 10:01 (p 1 of 1)
Test Code/ID: LabA_E_T2_RT25 / 08-9411-3943

Foliar Acute Bee Test										A	
Analysis ID: 02-9099-4484			Endpoint: 24-hr Mortality RT25			CETIS Version: CETISv1.9.7					
Analyzed: 07 Apr-22 10:01			Analysis: Linear Interpolation (ICPIN)			Status Level: 1					
Edit Date: 07 Apr-22 10:01			MD5 Hash: 70D35DC01752B36FAEC99666798AFA58			Editor ID: 001-771-848-3					
Batch ID: 10-2067-6662			Test Type: Acute Bee Survival			Analyst: Alison Briden					
Start Date: 14 Sep-21			Protocol: OCSPP 850.3030			Diluent: Not Applicable					
Ending Date: 16 Sep-21			Species: Apis Mellifera			Brine: Not Applicable					
Test Length: 48h			Taxon:			Source:			Age:		
Sample ID: 00-1221-4786			Code: LabA_E_T2_RT25			Project: 36326					
Sample Date: 14 Sep-21			Material: Dimethoate			Source: Pacific EcoRisk					
Receipt Date: 14 Sep-21			CAS (PC):			Station: Lab A					
Sample Age: ---			Client:								
Comments: RT25, Eurofins alfalfa, Trial 2											
Linear Interpolation Options											
X Transform	Y Transform	Seed	Resamples	Exp 95% CL	Method						
Linear	Linear	1413836	1	Yes	Two-Point Interpolation						
Point Estimates											
Level	T-hrs	95% LCL	95% UCL								
IC10	7.89	---	---								
IC15	8.83	---	---								
IC20	9.78	---	---								
IC25	10.7	---	---								
IC40	13.6	---	---								
IC50	15.4	---	---								
24-hr Mortality RT25 Summary											
			Calculated Variate						Isotonic Variate		
T-hrs	Code	Count	Mean	Median	Min	Max	CV%	%Effect	Mean	%Effect	
0		1	100	100	100	100	---	---	100		
6		1	100	100	100	100	---	---	100		
24		1	4.7	4.7	4.7	4.7	---	---	4.7		
24-hr Mortality RT25 Detail											
T-hrs	Code	Rep 1									
0		100									
6		100									
24		4.7									
Graphics											

Test Item:	T = Dimethoate 400 EC Formulation	Application Date: 14 Sep 2021 @ 09:25
Bee Colony Used:	21-A-04	Treatment Rate: T = 0.5 lb a.i./Ac = 560.4 g a.i./ha
		Crop: Alfalfa
* Corrected Mortality= (% T - % C)/(100 - % C) * 100		

Residual Timepoint:	6 Hours After Application
Harvest Time:	09/14/2021 @ 15:16
Exposure Time:	09/14/2021 @ C = 16:09, T = 16:17

	Date:		14-Sep-21	15-Sep-21	24 Hr.		
Treatment	Cage No.	No. Bees	Number of Dead Bees		Cumulative Total	% Cumulative Mortality	% Corrected Mortality
			≤4hr	24 hr			
C (Untreated Water Spray Alfalfa)	1	25	0	2	33	22.0	NA
	2	25	0	5			
	3	25	0	9			
	4	25	0	7			
	5	25	0	5			
	6	25	0	5			
Total		150	0	33			
% Cumulative Mortality			0.0	22.0			
T (Dimethoate 400 EC Treated Alfalfa)	1	25	0	25	150	100.0	100
	2	25	0	25			
	3	25	0	25			
	4	25	0	25			
	5	25	0	25			
	6	25	0	25			
Total		150	0	150			
% Cumulative Mortality			0.0	100.0			

Residual Timepoint:	24 Hours After Application
Harvest Time:	09/15/2021 @ 09:13
Exposure Time:	09/15/2021 @ C = 10:24, T = 10:32

	Date:		15-Sep-21	16-Sep-21	24 Hr.		
Treatment	Cage No.	No. Bees	Number of Dead Bees		Cumulative Total	% Cumulative Mortality	% Corrected Mortality
			≤4hr	24 hr			
C (Untreated Water Spray Alfalfa)	1	25	0	2	13	8.7	NA
	2	25	0	4			
	3	25	0	2			
	4	25	0	1			
	5	25	0	0			
	6	25	0	4			
Total		150	0	13			
% Cumulative Mortality			0.0	8.7			
T (Dimethoate 400 EC Treated Alfalfa)	1	25	0	0	7	4.7	-4.4
	2	25	0	1			
	3	25	0	2			
	4	25	0	1			
	5	25	0	1			
	6	25	0	2			
Total		150	0	7			
% Cumulative Mortality			0.0	4.7			

Post-application interval: +6hr	≤ 4 - Hour Observations		24 - Hour Observations	
Location: Eurofins	Test Concentration		Test Concentration	
Replicate	Control	T1	Control	T1
	Observation	Observation	Observation	Observation
1	All N	All N	All N	All dead
2	All N	All N	All N	All dead
3	All N	All N	All N	All dead
4	All N	All N	All N	All dead
5	All N	All N	All N	All dead
6	All N	All N	All N	All dead
Total	All N	All N	All N	All dead

Note: bees appear to be having a somewhat more difficult time climbing wall of cages than normal recorded 15 Sep 2021 by AW

Post-application interval: +24hr	≤ 4 - Hour Observations		24 - Hour Observations	
Location: Eurofins	Test Concentration		Test Concentration	
Replicate	Control	T1	Control	T1
	Observation	Observation	Observation	Observation
1	All N	All N	All N	All N
2	All N	All N	All N	All N
3	All N	All N	All N	All N
4	All N	All N	All N	All N
5	All N	All N	All N	All N
6	All N	All N	All N	All N
Total	All N	All N	All N	All N

Appendix L

Summary of Statistics for the Toxicity of Facility B September Alfalfa Application Tested By Lab B



CETIS Analytical Report

Report Date: 04 Apr-22 17:25 (p 1 of 2)
Test Code/ID: LabB_E_T2_6h / 14-0138-9934

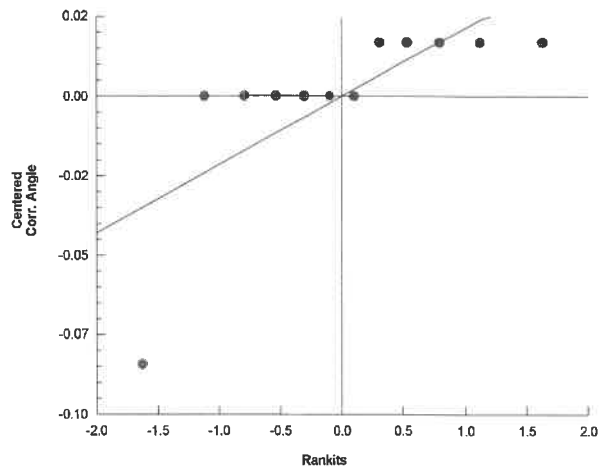
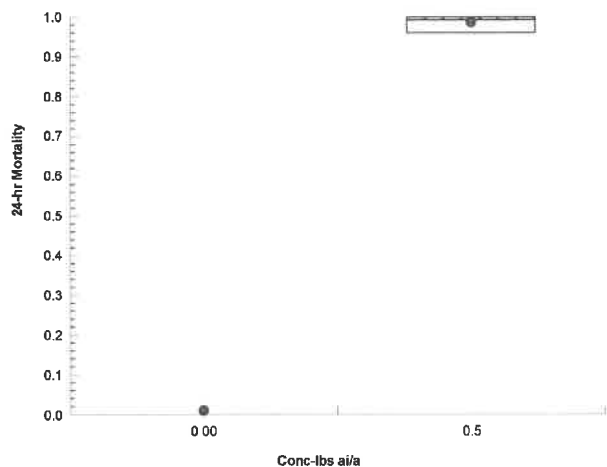
Foliar Acute Bee Test											B
Analysis ID: 10-6538-7459		Endpoint: 24-hr Mortality				CETIS Version: CETISv1.9.7					
Analyzed: 04 Apr-22 17:25		Analysis: Nonparametric-Two Sample				Status Level: 1					
Edit Date: 04 Apr-22 17:25		MD5 Hash: F11267F7180C622FC4546EB6CF10D877				Editor ID: 001-771-848-3					
Batch ID: 12-9844-6651		Test Type: Acute Bee Survival				Analyst: Alison Briden					
Start Date: 14 Sep-21		Protocol: OCSPP 850.3030				Diluent: Not Applicable					
Ending Date: 15 Sep-21		Species: Apis Mellifera				Brine: Not Applicable					
Test Length: 24h		Taxon:				Source:		Age:			
Sample ID: 12-2343-7350		Code: LabB_E_T2_6h				Project: 36326					
Sample Date: 14 Sep-21		Material: Dimethoate				Source: Pacific EcoRisk					
Receipt Date: 14 Sep-21		CAS (PC):				Station: Lab B					
Sample Age: ---		Client:									
Data Transform		Alt Hyp		Comparison Result					PMSD		
Angular (Corrected)		C < T		0.5lbs ai/a failed 24-hr mortality endpoint					1.70%		
Wilcoxon Rank Sum Two-Sample Test											
Control	vs	Conc-lbs ai/a	Test Stat	Critical	Ties	DF	P-Type	P-Value	Decision(α:5%)		
Control		0.5*	21	---	0	10	Exact	0.0011	Significant Effect		
ANOVA Table											
Source	Sum Squares		Mean Square		DF	F Stat	P-Value	Decision(α:5%)			
Between	5.49667		5.49667		1	6440	<1.0E-05	Significant Effect			
Error	0.0085329		0.0008533		10						
Total	5.5052				11						
ANOVA Assumptions Tests											
Attribute	Test				Test Stat	Critical	P-Value	Decision(α:1%)			
Variance	Variance Ratio F Test				6.15E+14	14.9	<1.0E-05	Unequal Variances			
Distribution	Shapiro-Wilk W Normality Test				0.561	0.802	5.2E-05	Non-Normal Distribution			
24-hr Mortality Summary											
Conc-lbs ai/a	Code	Count	Mean	95% LCL	95% UCL	Median	Min	Max	Std Err	CV%	%Effect
0	00	6	0.000	0.000	0.000	0.000	0.000	0.000	0.000	---	0.00%
0.5		6	0.993	0.976	1.000	1.000	0.960	1.000	0.007	1.64%	99.33%
Angular (Corrected) Transformed Summary											
Conc-lbs ai/a	Code	Count	Mean	95% LCL	95% UCL	Median	Min	Max	Std Err	CV%	%Effect
0	00	6	0.100	0.100	0.100	0.100	0.100	0.100	0.000	0.00%	100.00%
0.5		6	1.450	1.410	1.500	1.470	1.370	1.470	0.017	2.84%	6.89%
24-hr Mortality Detail											
Conc-lbs ai/a	Code	Rep 1	Rep 2	Rep 3	Rep 4	Rep 5	Rep 6				
0	00	0.000	0.000	0.000	0.000	0.000	0.000				
0.5		1.000	1.000	1.000	1.000	1.000	0.960				
Angular (Corrected) Transformed Detail											
Conc-lbs ai/a	Code	Rep 1	Rep 2	Rep 3	Rep 4	Rep 5	Rep 6				
0	00	0.100	0.100	0.100	0.100	0.100	0.100				
0.5		1.470	1.470	1.470	1.470	1.470	1.370				
24-hr Mortality Binomials											
Conc-lbs ai/a	Code	Rep 1	Rep 2	Rep 3	Rep 4	Rep 5	Rep 6				
0	00	0/25	0/25	0/25	0/25	0/25	0/25				
0.5		25/25	25/25	25/25	25/25	25/25	24/25				

Foliar Acute Bee Test

B

Analysis ID: 10-6538-7459	Endpoint: 24-hr Mortality	CETIS Version: CETISv1.9.7
Analyzed: 04 Apr-22 17:25	Analysis: Nonparametric-Two Sample	Status Level: 1
Edit Date: 04 Apr-22 17:25	MD5 Hash: F11267F7180C622FC4546EB6CF10D877	Editor ID: 001-771-848-3

Graphics



CETIS Analytical Report

Report Date: 04 Apr-22 17:28 (p 1 of 2)
 Test Code/ID: LabB_E_T2_24h / 15-8141-0064

Foliar Acute Bee Test											B
Analysis ID: 12-5918-0734		Endpoint: 24-hr Mortality		CETIS Version: CETISv1.9.7							
Analyzed: 04 Apr-22 17:27		Analysis: Parametric-Two Sample		Status Level: 1							
Edit Date: 04 Apr-22 17:27		MD5 Hash: 345B3665E557DEA91931B14B33F28BF9		Editor ID: 001-771-848-3							
Batch ID: 15-4552-4752		Test Type: Acute Bee Survival		Analyst: Alison Briden							
Start Date: 15 Sep-21		Protocol: OCSP 850.3030		Diluent: Not Applicable							
Ending Date: 16 Sep-21		Species: Apis Mellifera		Brine: Not Applicable							
Test Length: 24h		Taxon:		Source: Age:							
Sample ID: 03-5151-2715		Code: LabB_E_T2_24h		Project: 36326							
Sample Date: 14 Sep-21		Material: Dimethoate		Source: Pacific EcoRisk							
Receipt Date: 14 Sep-21		CAS (PC):		Station: Lab B							
Sample Age: 24h		Client:									
Comments: Post-application interval: +6h Eurofins alfalfa, Trial 2											
Data Transform	Alt Hyp	Comparison Result								PMSD	
Angular (Corrected)	C < T	0.5lbs ai/a failed 24-hr mortality endpoint								4.39%	
Equal Variance t Two-Sample Test											
Control	vs	Conc-lbs ai/a	Test Stat	Critical	MSD	DF	P-Type	P-Value	Decision(α:5%)		
Control		0.5*	4.85	1.81	0.095	10	CDF	0.0003	Significant Effect		
ANOVA Table											
Source	Sum Squares		Mean Square		DF	F Stat	P-Value	Decision(α:5%)			
Between	0.192361		0.192361		1	23.5	0.0007	Significant Effect			
Error	0.0818159		0.0081816		10						
Total	0.274177				11						
ANOVA Assumptions Tests											
Attribute	Test		Test Stat	Critical	P-Value	Decision(α:1%)					
Variance	Variance Ratio F Test		1.34	14.9	0.7533	Equal Variances					
Distribution	Shapiro-Wilk W Normality Test		0.9	0.802	0.1595	Normal Distribution					
24-hr Mortality Summary											
Conc-lbs ai/a	Code	Count	Mean	95% LCL	95% UCL	Median	Min	Max	Std Err	CV%	%Effect
0	00	6	0.040	0.002	0.078	0.040	0.000	0.080	0.015	89.44%	0.00%
0.5		6	0.193	0.112	0.275	0.180	0.120	0.320	0.032	40.15%	15.97%
Angular (Corrected) Transformed Summary											
Conc-lbs ai/a	Code	Count	Mean	95% LCL	95% UCL	Median	Min	Max	Std Err	CV%	%Effect
0	00	6	0.196	0.108	0.284	0.201	0.100	0.287	0.034	42.60%	100.00%
0.5		6	0.449	0.348	0.551	0.438	0.354	0.601	0.040	21.56%	43.64%
24-hr Mortality Detail											
Conc-lbs ai/a	Code	Rep 1	Rep 2	Rep 3	Rep 4	Rep 5	Rep 6				
0	00	0.040	0.000	0.000	0.040	0.080	0.080				
0.5		0.200	0.160	0.320	0.120	0.120	0.240				
Angular (Corrected) Transformed Detail											
Conc-lbs ai/a	Code	Rep 1	Rep 2	Rep 3	Rep 4	Rep 5	Rep 6				
0	00	0.201	0.100	0.100	0.201	0.287	0.287				
0.5		0.464	0.412	0.601	0.354	0.354	0.512				
24-hr Mortality Binomials											
Conc-lbs ai/a	Code	Rep 1	Rep 2	Rep 3	Rep 4	Rep 5	Rep 6				
0	00	1/25	0/25	0/25	1/25	2/25	2/25				
0.5		5/25	4/25	8/25	3/25	3/25	6/25				

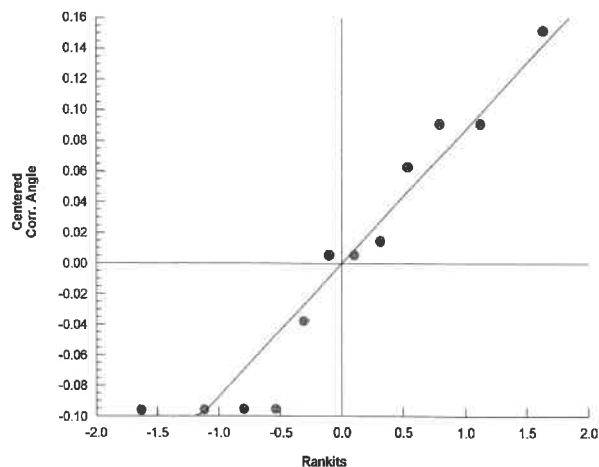
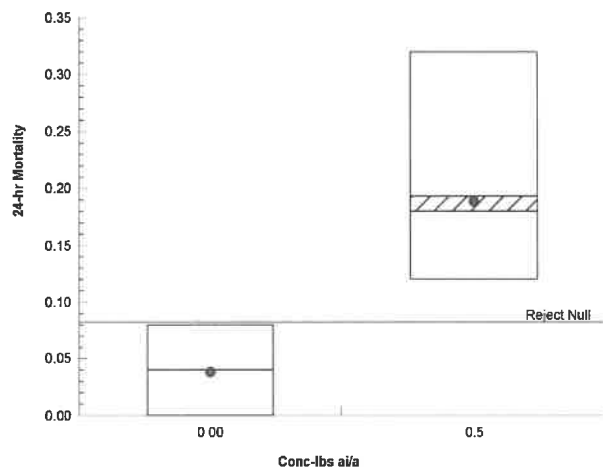
Foliar Acute Bee Test

B

Analysis ID: 12-5918-0734 Endpoint: 24-hr Mortality
Analyzed: 04 Apr-22 17:27 Analysis: Parametric-Two Sample
Edit Date: 04 Apr-22 17:27 MD5 Hash: 345B3665E557DEA91931B14B33F28BF9

CETIS Version: CETISv1.9.7
Status Level: 1
Editor ID: 001-771-848-3

Graphics



CETIS Analytical Report

Report Date: 07 Apr-22 10:19 (p 1 of 1)
Test Code/ID: LabB_E_T2_RT25 / 02-8372-5060

Foliar Acute Bee Test										B
Analysis ID: 06-4912-4016		Endpoint: 24-hr Mortality RT25		CETIS Version: CETISv1.9.7						
Analyzed: 07 Apr-22 10:18		Analysis: Linear Interpolation (ICPIN)		Status Level: 1						
Edit Date: 07 Apr-22 10:18		MD5 Hash: D18521EA6FDF04CC0461F1A5C1564861		Editor ID: 001-771-848-3						
Batch ID: 10-2218-9687		Test Type: Acute Bee Survival		Analyst: Alison Briden						
Start Date: 14 Sep-21		Protocol: OCSP 850.3030		Diluent: Not Applicable						
Ending Date: 16 Sep-21		Species: Apis Mellifera		Brine: Not Applicable						
Test Length: 48h		Taxon:		Source:		Age:				
Sample ID: 04-2414-9694		Code: LabB_E_T2_RT25		Project: 36326						
Sample Date: 14 Sep-21		Material: Dimethoate		Source: Pacific EcoRisk						
Receipt Date: 14 Sep-21		CAS (PC):		Station: Lab B						
Sample Age: ---		Client:								
Comments: RT25, Eurofins alfalfa, Trial 2										
Linear Interpolation Options										
X Transform	Y Transform	Seed	Resamples	Exp 95% CL	Method					
Linear	Linear	1497058	1	Yes	Two-Point Interpolation					
Point Estimates										
Level	T-hrs	95% LCL	95% UCL							
IC10	8.01	---	---							
IC15	9.09	---	---							
IC20	10.2	---	---							
IC25	11.3	---	---							
IC40	14.5	---	---							
IC50	16.7	---	---							
24-hr Mortality RT25 Summary										
		Calculated Variate							Isotonic Variate	
T-hrs	Code	Count	Mean	Median	Min	Max	CV%	%Effect	Mean	%Effect
0		1	100	100	100	100	---	---	100	
6		1	99.3	99.3	99.3	99.3	---	---	99.3	
24		1	16	16	16	16	---	---	16	
24-hr Mortality RT25 Detail										
T-hrs	Code	Rep 1								
0		100								
6		99.3								
24		16								
Graphics										

Test Item:	T = Dimethoate 400 EC Formulation	Application Date: 14 Sep 2021 @ 09:25
Bee Colony Used:	21-A-04	Treatment Rate: T = 0.5 lb ai / Ac = 560.4 g a.i. / ha
		Crop: Alfalfa
* Corrected Mortality = $(\% T - \% C) / (100 - \% C) * 100$		

Residual Timepoint:	6 Hours After Application
Harvest Time:	09/14/2021 @ 15:16
Exposure Time:	09/14/2021 @ C = 16:09, T = 16:17

	Date:		14-Sep-21	15-Sep-21	24 Hr.		
Treatment	Cage No.	No. Bees	Number of Dead Bees		Cumulative Total	% Cumulative Mortality	% Corrected Mortality
			≤4hr	24 hr			
C (Untreated Water Spray Alfalfa)	1	25	0	0	0	0.0	NA
	2	25	0	0			
	3	25	0	0			
	4	25	0	0			
	5	25	0	0			
	6	25	0	0			
Total		150	0	0			
% Cumulative Mortality			0.0	0.0			
T (Dimethoate 400 EC Treated Alfalfa)	1	25	1	25	149	99.3	99.3
	2	25	0	25			
	3	25	0	25			
	4	25	1	25			
	5	25	0	25			
	6	25	0	24			
Total		150	2	149			
% Cumulative Mortality			1.3	99.3			

1 apathetic bee @ 4 and 24 hour assessments

Residual Timepoint:	24 Hours After Application
Harvest Time:	09/15/2021 @ 09:13
Exposure Time:	09/15/2021 @ C = 10:24, T = 10:32

	Date:		15-Sep-21	16-Sep-21	24 Hr.		
Treatment	Cage No.	No. Bees	Number of Dead Bees		Cumulative Total	% Cumulative Mortality	% Corrected Mortality
			≤4hr	24 hr			
C (Untreated Water Spray Alfalfa)	1	25	0	1	6	4.0	NA
	2	25	0	0			
	3	25	0	0			
	4	25	0	1			
	5	25	0	2			
	6	25	0	2			
Total		150	0	6			
% Cumulative Mortality			0.0	4.0			
T (Dimethoate 400 EC Treated Alfalfa)	1	25	0	5	29	19.3	16
	2	25	0	4			
	3	25	0	8			
	4	25	0	3			
	5	25	0	3			
	6	25	0	6			
Total		150	0	29			
% Cumulative Mortality			0.0	19.3			

3 affected @ 24 hour assessment
2 affected @ 24 hour assessment
1 affected @ 24 hour assessment

2 affected @ 24 hour assessment
1 moribund @ 24 hour assessment