Joint Exhibit 87

EPA OALJ Docket No. FIFRA-HQ-2022-0002

Data Evaluation Record on the Acute Toxicity of Tetrachloroterephthalic Acid (TPA) (Metabolite of DCPA) to Freshwater Invertebrates – *Daphnia magna*

PMRA Submission Number {.....} EPA MRID Number 49307519

Data Requirement:	PMRA Data Code	{.....}
	EPA DP Barcode	420874
	OECD Data Point	{.....}
	EPA MRID	49307519
	EPA Guideline	850.1010

Test material: **Tetrachloroterephthalic Acid (TPA) (Metabolite of DCPA)** **Purity:** 99.94%
Common name:
Chemical name: IUPAC:
CAS name:
CAS No.: 2136-79-0
Synonyms:

Primary Reviewer: Teresa Nelis **Signature:** Teresa Nelis
Environmental Scientist, CDM Smith/CSS-Dynamac JV **Date:** 10/11/2016

Secondary Reviewer: Moncie V. Wright **Signature:** Moncie V Wright
Environmental Scientist, CDM Smith/CSS-Dynamac JV **Date:** 10/28/2016

Primary Reviewer: Christina M. Wendel **Signature:**
EPA/OPP/EFED/ERB2/Biologist **Date:** 10/25/2021

Secondary Reviewer(s): Michael Wagman **Signature:**
EPA/OPP/EFED/ERB2/Senior Scientist **Date:** 11/12/2021

Reference/Submission No.: {.....}

Company Code	{.....}	[For PMRA]
Active Code	{.....}	[For PMRA]
Use Site Category:	{.....}	[For PMRA]
EPA PC Code	078701	

Date Evaluation Completed: 12-11-2021

CITATION: Wood, J. 2014. Tetrachloroterephthalic Acid (TPA): Acute Toxicity to Daphnia Magna (Revised per PRN 11-03 and 86-5). Study performed by Covance Laboratories Ltd, North Yorkshire, England. Laboratory Study Number: 1708/021. Study sponsored by AMVAC Chemical UK Ltd, Surrey, England. Study completed November 24, 2003.

This Data Evaluation Record may have been altered by the Environmental Fate and Effects Division subsequent to signing by CDM/CSS-Dynamac JV personnel.

EXECUTIVE SUMMARY:

The 48-hour acute toxicity of **Tetrachloroterephthalic Acid (TPA) (metabolite of DCPA)** to *Daphnia magna* was studied under static conditions. Daphnids were exposed to TPA at nominal concentrations of 0 (negative control) and 100 mg a.i./L for 48 hours. The mean-measured concentrations were <LOD (<0.002, control) and 103 mg a.i./L.

Mortality and sublethal effects were observed daily. No mortality or sublethal effects were observed in this experiment. The 48-hour EC_{50} was >103 mg a.i./L, and the observed NOAEC was 103 mg a.i./L based on the mean-measured concentrations.

Based on the results of this study, **TPA (metabolite of DCPA)** would be classified as practically non-toxic to *Daphnia magna* in accordance with the classification system of the U.S. EPA.

This study **is scientifically sound** and is classified as **acceptable**.

Results Synopsis

Test Organism Age (e.g., 1st instar): <24 hours old
Test Type (Flow-through, Static, Static Renewal): Static

48-hour EC_{50}: >103 mg a.i./L 95% C.I.: N/A
Probit Slope: N/A 95% C.I.: N/A
Observational NOAEC: 103 mg a.i./L

Endpoint(s) Affected: None

I. MATERIALS AND METHODS

GUIDELINE FOLLOWED: This study was conducted according to a protocol that meets the testing requirements of the U.S. Environmental Protection Agency's Ecological Effects Test Guideline OCSPP (*form*. OPPTS) 850.1010: *Aquatic Invertebrate Acute Toxicity Test, Freshwater Daphnids*, OECD Guideline for the testing of Chemicals No 202 Part 1 Daphnia sp., Acute Immobilisation Test, and JMAFF Guideline 2-7-2. The reviewer assessed the study methods and results according to U.S. EPA OCSPP 850.1010. The following deficiencies were noted:

1. Health of the parental daphnids (*e.g.*, number of broods, presence of ephippia) was not described. This is an uncertainty and considered a study deficiency for both the draft OPPTS and final OCSPP 850.1010 guideline. However, this is considered a minor deficiency with the information provided in Appendix 1 in the study report, although not complete it provides some insight into the health and maintenance of the laboratory cultures of *D. magna* that were used in the study.
2. Juveniles for use in acute toxicity tests were collected from the third brood onwards from adults aged between 14 and 42 days. OPPTS guidance recommends that juveniles are collected from the fourth brood onward. However, OECD guidance and the final OCSPP 850.1010 guideline only recommends that juveniles used for testing are not first brood progeny. This is considered a minor deficiency.
3. Light intensity measurements were not reported, nor was the use of a transition period. It was only reported that a 16L:8D hour photoperiod was utilized (Appendix 1; pg. 25-26). Both sets of guidelines suggest that light intensity be reported. Both draft and final OPPTS and OCSPP 850.1010 guidelines recommends 30-minute transition periods between light and dark conditions, while OECD guidance does not address the use of transition period. This would be considered a minor deficiency.
4. The TOC, particulate matter, pesticides, metals, and residual chlorine content of the test water were not reported. Both draft and final OPPTS and OCSPP 850.1010 guidelines recommends specific maximum concentrations for each of these categories, while OECD guidance does not. However, this is considered a minor deficiency as the solubility (175.4 mg/L) of TPA in water would not result in an underestimation of toxicity; and the full preparation details of the dilution water were presented in Appendix 1 of the study report (pg. 25-26).

These deficiencies *do not* have an impact on the scientific integrity of this experiment.

COMPLIANCE: Signed and dated GLP, Quality Assurance, and Data Confidentiality statements were provided. This study was conducted in compliance with U.S. EPA Good Laboratory Practice regulations (40 CFR, Part 160), UK Statutory Instrument 1999 No. 3106, the OECD Principles on GLP ENV/MC/CHEM (98)17, and GLP Standards for Agricultural Chemicals 14-Seisan-7739, with no exceptions indicated.

A. MATERIALS:

1. Test material: **Tetrachloroterephthalic Acid (TPA) (metabolite of DCPA)**

Description: White powder

Lot No./Batch No.: 021101

Purity: 99.94%

Stability of compound under test conditions: Analytical verification performed at time 0 yielded a recovery of 107%. At 48 hours (test termination), recoveries averaged 104% of the nominal concentration, indicating the test material was stable.

Storage conditions of test chemicals: The test material was stored at room temperature (20ºC ± 10).

Physicochemical properties of TPA.

Parameter	Values	Comments
Water solubility at 20°C	Not reported.	
Vapor pressure	Not reported.	
UV absorption	Not reported.	
pKa	Not reported.	
Kow	Not reported.	

(OECD recommends water solubility, stability in water and light, pKa, Pow, and vapor pressure of test compound)

2. Test organism:

Species: *Daphnia magna*
(Strain not reported)
(EPA preferred species is *Daphnia magna;* OECD preferred species is *Daphnia magna* or any other suitable *Daphnia* species)

Age at test initiation: <24 hours old
(EPA recommends that Daphnids are in their first instar (#24 hrs old) and that all organisms are approximately the same size and age; OECD requires age <24 hrs old)

Source: In-house laboratory cultures maintained at Covance Laboratories, Ltd., originally obtained from Sheffield University, Sheffield, UK, from the third or later brood of a given parent.
(EPA requires that all organisms are from the same source. Daphnids from ephippia-producing cultures should not be used; Daphnids should be from the fourth or later brood of a given parent)

B. STUDY DESIGN:

1. Experimental Conditions

a. Range-finding study A 48-hour static range-finding study was conducted with a negative control and concentrations of 0.1, 1.0, 10 and 100 mg a.i./L. Twenty daphnids (four replicates, five daphnids per replicate, <24 hours old) were exposed to each control and treatment group. After 48 hours of exposure, immobilization was 0% in the control and in all the treatment groups. One daphnia that was floating on the surface at 24 hours in the 100 mg a.i./L nominal treatment group re-submerged by 48 hours.

b. Definitive Study

Table 1: Experimental Parameters

Parameter	Details	Remarks *Criteria*
Acclimation Period:	Daphnids were cultured for 14 to 42 days prior to testing under similar conditions to those used for testing.	New cultures were started with *D. magna* <24 hours old at a density of about 20/L. *D. magna* were cultured in 600 mL glass beakers containing 500 mL of standard hard water. Water in the glass beakers were renewed weekly. All juveniles used to initiate a test were <24 hours old. *The recommended acclimation period is a minimum of 7 days. Organisms should not feed during the study.* *Pretest mortality should be <3% 48 hours prior to testing.*
Conditions: (same as test or not)	Similar to test: ASTM standard hard water (prepared with analysis grade reagents and reverse-osmosis water), 16L:8D photoperiod, dissolved oxygen ranged from 98 to 100%, pH ranged from 7.5 to 7.8, and temperature ranged from 20.2 to 20.7ºC).	
Feeding:	Daphnids were fed a concentrated suspension of *Chlorella vulgaris* at 5 x 10^5 cells/mL (≤2 weeks in age), and seaweed extract (Marinure, Glenside Organics, Scotland) daily.	
Health: (any mortality observed)	The adults used to produce offspring for this study were 14 to 42 days old. Juveniles collected for use in the study were collected from the third brood onwards.	
Duration of the test	48 hours	*EPA requires 96 hours, except daphnids which are 48 hours.*
Test condition Static/flow-through	Static	*The recommended flow rates are 5 - 10 volume additions/24 hours; meter systems should be calibrated before and after the study and checked twice daily during the test period.*
Type of dilution system for flow-through method.	N/A	
Renewal rate for static renewal	N/A	
Aeration, if any	None provided.	
Test vessel Material: *(glass/stainless steel)*	Glass beakers	*EPA requires: small organisms in 3.9 L (1 gallon) wide mouth jars with 2-3 L of solution or daphnids and midge larvae in 250 ml jars w/ 200 ml fill*
Size: Fill volume:	400 mL 200 mL	

Parameter	Details	Remarks *Criteria*
Source of dilution water	ASTM standard hard water (prepared with analysis grade reagents and reverse-osmosis water). The final concentrations of the following constituents in dilution water were as follows: Sodium hydrogen carbonate: 192 mg/L Calcium sulphate dihydrate: 120 mg/L Magnesium sulphate heptahydrate: 246 mg/L Potassium chloride: 8 mg/L The dilution water had a hardness ranging from 165-170 mg/L as $CaCO_3$, alkalinity ranging from 77-84 mg/L as $CaCO_3$, a pH ranging from7.5-7.8, and a dissolved oxygen % saturation ranging from 98-100%, and a culture temperature ranging from 20.2-20.7°C.	Full preparation details were presented in Appendix 1 of the study report (pg. 25-26). *Recommended source of dilution water is soft, reconstituted water or water from a natural, uncontaminated source. EPA does not recommend the use of dechlorinated tap water; however, its use may be supportable if the biological responses for the organisms and chemical analyses of residual chlorine meet conditions in the Agency's 850.1010 guidelines for dilution water (http://www.epa.gov/opptsfrs/OPPTS_Harmonized/850_Ecological_Effects_Test_Guidelines/Draft/850.1010Opdf). Dilution water should be intensely aerated before the study.*
<u>Water parameters</u> Hardness pH Dissolved oxygen Temperature Total Organic Carbon Particulate matter Metals Pesticides Chlorine	 163 to 168 mg/L as $CaCO_3$ 7.3 to 8.3 98 to 100% 20.1 to 20.6ºC Not reported Not reported Not reported Not reported Not reported	Alkalinity as $CaCO_3$: 88 to 94 mg/L *<u>Hardness</u>:* *EPA recommends 40 - 48 mg/L as $CaCO_3$ (OECD recommends 140 - 250 mg/L)* *<u>pH:</u>* *EPA recommends: 7.2 - 7.6 (OECD recommends pH of 6-9); measured at start and end of test in control, high, medium, and low test concentrations* *<u>Temperature</u>:* *EPA recommends: 20ºC for Daphnia (measured hourly) in at least one test vessel or if water baths are used, every 6 hr, may not vary > 1ºC;* OECD *recommends* range of 18-22EC (±1EC) *<u>Dissolved oxygen</u>:* *EPA recommends: Measured at start and every 48 hours thereafter in control, high, medium, and low test concentrations.* *Static: 60-100% during 1st 48 hr and 40-100% during 2nd 48 hr* *Flow-through: 60-100% at all times*

Parameter	Details	Remarks *Criteria*
Number of replicates Negative control: Solvent control: Treatments:	4 N/A 4	*EPA requires 2 or more containers for each treatment group; individuals must be randomly assigned to test vessels* *OECD recommends 4 groups of 5 animals for each test concentration and the controls*
Number of organisms per replicate Negative control: Solvent control: Treatments:	5 N/A 5	20 daphnids per concentration and control. *EPA/OECD requires 5 treatment levels plus one or more control groups; no more than 10% or 5% of control organisms should die during a static or flow-through study, respectively* *EPA requires a minimum of 20 daphnids in 2 or more containers per treatment; however, if a limit test is conducted, it must be shown that the LC_{50}/EC_{50} is >100 mg/L by exposing∃ 30 organisms to ≥100 mg/L or greater. Biomass loading rate for static ≤ 0.8 g/L at ≤ 17°C and #0.5 g/L at > 17°C; flow-through: # 10 g/L at ≤ 17^{B}C and ≤5 g/L at > 17^{B}C.* *OECD recommends a minimum of 20 animals, preferably with 4 groups of 5 animals for each test concentration. There should be at least 2ml of test solution for each animal.*
Treatment concentrations Nominal: Measured:	0 (negative control), 100 mg a.i./L <LOD (<0.002, control), 103 mg a.i./L	This study was conducted as a limit test. The negative control medium was prepared using ASTM only *Treatment concentrations should include a geometric series of at least five concentrations plus a control with each recommended concentration being at least 60% of the next higher one. The variability of measured concentrations between replicates of the same concentration should not exceed 1.5.* *OECD recommends that the highest test concentration should result in 100% immobilization and not be ≥1 g/L, while the lowest concentration should have no observable effect.*

Parameter	Details	Remarks *Criteria*
Solvent (type, percentage, if used)	N/A	*Solvents should not exceed 0.5 ml/L for static tests or 0.1 ml/L for flow-though tests. OECD recommends that the solvent not exceed 100 mg/L.*
Lighting	Provided by fluorescent bulbs at a photoperiod of 16L:8D; transition period and light intensity measurements not reported.	*EPA-recommended photoperiod is 16 hours of light and 8 hours of dark with a 15-30 minute transition period.* *OECD: optional light-dark cycle or complete darkness.*
Stability of chemical in the test system	The test material was stable; recovery was 103 to 104% of nominal at 48 hours.	
Recovery of chemical Level of Quantitation (LOQ) Level of Detection (LOD)	Test solutions were sampled in every treatment vessel at 0 and 48 hours. Test solutions were analyzed for TPA using HPLC with UV detection (220 nm). Not reported 0.002 mg a.i./L	Samples (duplicate 20 mL volumes) of test solutions for analysis were taken from each test vessel for chemical analysis of TPA at the start and end of the exposure period. At the start of exposure and at the end of exposure period the test solutions were clear and colorless.
Positive control {if used, indicate the chemical and concentrations}	N/A	
Other parameters, if any	None	

2. Observations:

Table 2: Observations

Criteria	Details	Remarks
Parameters measured including sublethal effects	Immobility and sublethal effects	Considered immobile if, when the contents of the test vessel were briefly swirled, they did not swim during a 15-second period of observation.
Observation intervals	0, 24, and 48 hours	
Were raw data included?	Yes	
Other observations, if any	None	

II. RESULTS AND DISCUSSION

A. MORTALITY:

No immobility occurred in the control or treatment levels throughout the study. The study author reported an EC_{50} value of >100 mg a.i./L, based on the nominal concentrations.

Table 3: Effect of TPA on Mortality of *Daphnia magna*[1]

Treatment Nominal [Mean-Measured] (mg a.i./L)	No. of Organisms	Observation Period					
		0 hour		24 hours		48 hours	
		No Dead	% Immobility	No Dead	% Immobility	No Dead	% Immobility
<LOD* (Control; dilution water only)	20	0	0	0	0	0	0
100 [103]	20	0	0	0	0	0	0
EC_{50}	>100 mg a.i./L						
Positive control, if used. mortality: LC_{50}:	N/A						

[1] Data were obtained from Table 3 and 4 on page 23 of the study report.
* Level of Detection (LOD) = 0.002 mg a.i./L

B. SUB-LETHAL TOXICITY ENDPOINTS:

No sublethal effects were observed in the control or any of the treatment groups that were tested.

Table 4: Effect of TPA on Sublethal Effects – *Daphnia magna*

Treatment Nominal [Mean-Measured] (mg a.i./L)	Observation period		
	0 hour	24 hours	48 hours
	% affected	% affected	% affected
<LOD* (Control; dilution water only)	0	0	0
100 [103]	0	0	0
EC_{50}	N/A		
Positive control, if used % sublethal effect: EC_{50}:	N/A		

* Level of Detection (LOD) = 0.002 mg a.i./L

C. REPORTED STATISTICS:

No concentration tested resulted in ≥50% mortality, so the EC_{50} value was empirically estimated. Nominal concentrations were used for the analysis.

D. VERIFICATION OF STATISTICAL RESULTS:

Statistical Method: The mortality data and mean-measured concentrations of the test material were entered into the program CETIS, (Version 1.8.7.12) with backend settings implemented by EFED on 10/20/15. The reviewer visually determined the EC_{50} due to a complete lack of mortality in this study. Mean-measured concentrations were used.

48-hour LC_{50}: >103 mg a.i./L 95% C.I.: N/A
Probit Slope: N/A

E. STUDY DEFICIENCIES:

There were no major study deficiencies. Minor deficiencies included the following: the health of parental daphnids (*e.g.*, number of broods, presence of ephippia) was not fully described, juveniles for use in acute toxicity tests were collected from the third brood onwards from adults aged between 14 and 42 days, light intensity measurements were not reported, nor was the use of a transition period, and the TOC, particulate matter, pesticides, metals, and residual chlorine content of the test water were not reported. All were considered to be minor deficiencies.

F. REVIEWER'S COMMENTS:

The reviewer's and the study author's results were in general agreement; there was no toxicity in this study. However, the reviewer's results are reported using the mean-measured concentration, and therefore reported in the Executive Summary and Conclusions sections of this DER.

The study was conducted following the draft OPPTS 850.1010 (1996) guideline, OECD 202 guideline, and JMAFF 2-7-2 Guideline, and the reviewer considered both the draft and the final OCSPP 850.1010 (2016) guideline and OECD 202 guideline in their evaluation of the data.

The in-life phase of the study was initiated on May 12, 2003 and the definitive test was completed on April 2, 2003.

G. CONCLUSIONS:

This study **is scientifically sound** and is classified as **acceptable.** There was no mortality or sublethal effects observed in this study. The 48-hour EC_{50} value for Tetrachloroterephthalic Acid (TPA) (metabolite of DCPA) was >103 mg a.i./L, and the observed NOAEC was 103 mg a.i./L, based on the mean-measured concentration.

III. REFERENCES:

None.