Data Evaluation Report on the Acute Dietary Toxicity of AE 0172747 Technical to Mallard duck (*Anas platyrhynchos*)

PMRA Submission Number {.....} EPA MRID Number 466955-02

Data Requirement:	PMRA Data Code	{.....}
	EPA DP Barcode	D325337
	OECD Data Point	{.....}
	EPA MRID	466955-02
	EPA Guideline	850.2200

PC: 012801

Test material: AE 0172747 **Purity:** 94.0% (w:w)

Common name AE 0172747

Chemical name: IUPAC 2-[2-Chloro-4-mesyl-3-((2,2,2-trifluoroethoxy)methyl)benzoyl]cyclohexane-1,3-dione

CAS name 2-[2-Chloro-4-(methylsulfonyl)-3-[(2,2,2-trifluoroethoxy)methyl]-benzoyl]-1,3-cyclohexanedione

CAS No. 335-104-84-2

Synonyms Bayer AE 0172747

Primary Reviewer: Christie E. Padova **Signature:** Christie E. Padova
Staff Scientist, Dynamac Corporation **Date:** 5/29/06

Secondary Reviewer: Teri S. Myers **Signature:** Teri S Myers
Senior Scientist, Cambridge Environmental Inc. **Date:** 6/2/06

Primary Reviewer: **Date:** {.....}
EPA/OPP/EFED/ERB –

Secondary Reviewer(s): Jeannette Martinez **Date:** 7/25/06
{EPA/OECD/PMRA}

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

Company Code	{.....}	[For PMRA]
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Use Site Category:	{.....}	[For PMRA]
EPA PC Code	{.....}	012801

Date Evaluation Completed: dd-mm-yyyy

CITATION: Gallagher, S.P., *et al.* 2002. A Dietary LC50 Study with the Mallard: AE 0172747; Substance Technical; Product Code: AE 0172747 00 1C94 0002. Unpublished study performed by Wildlife International, Ltd., Easton, MD. Laboratory Project No. 149-183. Study submitted by Bayer CropScience GmbH, Frankfurt am Main, Germany. Study initiated August 28, 2002 and submitted November 22, 2002.

DISCLAIMER: This document provides guidance for EPA and PMRA reviewers on how to complete a data evaluation record after reviewing a scientific study concerning the acute dietary toxicity of a pesticide to avian species. It is not intended to prescribe conditions to any external party for conducting this study nor to establish absolute criteria regarding the assessment of whether the study is scientifically sound and whether the study satisfies any applicable data requirements. Reviewers are expected to review and to determine for each study, on a case-by-case basis, whether it is scientifically sound and provides sufficient information to satisfy applicable data requirements. Studies that fail to meet any of the conditions may be accepted, if appropriate; similarly, studies that meet all of the conditions may be rejected, if appropriate. In sum, the reviewer is to take into account the totality of factors related to the test methodology and results in determining the acceptability of the study.

EXECUTIVE SUMMARY:

The acute dietary toxicity of AE 0172747 Technical to 10-day old mallard duck (*Anas platyrhynchos*) was assessed over 8 days. AE 0172747 Technical was administered to the birds in the diet at nominal concentrations of 0 (control), 562, 1000, 1780, 3160, and 5620 mg ai/kg diet. Mean-measured concentrations were <50 (<LOD, control), 580, 1040, 1890, 3220, and 5790 mg ai/kg diet, respectively. The 8-day acute dietary LC_{50} was >5790 mg ai/kg diet. Based on treatment-related effects on body weight gain (the most sensitive endpoint) at all treatment levels during the exposure phase, the 8-day NOAEC of AE 0172747 Technical was <580 mg ai/kg diet. According to the US EPA classification, AE 0172747 Technical would be classified as practically non-toxic to mallard duck (*Anas platyrhynchos*) on an acute dietary basis.

No mortality or clinical signs of toxicity were observed. However, a dose-responsive treatment-related reduction in body weight gain was observed in birds from all treatment levels compared to the control during the exposure period. Body weight changes from days 0-5 averaged 155 g for the control group, compared to 113, 62, 34, 34, and 10 g for the 562, 1000, 1780, 3160, and 5620 mg ai/kg diet. While there was some compensatory weight gain during the post-exposure observation period, when compared to the control group, mean body weights were still reduced in all treatment groups over the course of the study (days 0-8). A treatment-related effect in food consumption was observed in birds from the 1780, 3160, and 5620 mg ai/kg diet levels compared to controls during the exposure period. Food consumption averaged 106, 110, 97, 71, 80, and 83 g/bird/day for birds in the control, 562, 1000, 1780, 3160, and 5620 mg ai/kg diet levels, respectively.

This toxicity study is classified as scientifically sound and is thus acceptable and does satisfy the guideline requirement for an acute dietary toxicity study for the mallard duck.

Results Synopsis

Test Organism Size/Age(Mean Weight): 10-days old; 141-195 g

LC_{50}: >5790 mg ai/kg diet 95% C.I.: N/A
Probit Slope: N/A 95% C.I.: N/A

NOAEC: <580 mg ai/kg diet
LOAEC: 580 mg ai/kg diet

Endpoint(s) affected: body weight and food consumption
Most sensitive endpoint: body weight

I. MATERIALS AND METHODS

GUIDELINE FOLLOWED: The study protocol was based on procedures outlined in the U.S. EPA Ecological Effects Test Guidelines OPPTS No. 850.2200; U.S. EPA Pesticide Assessment Guidelines, §71-2; OECD Guideline for Testing of Chemicals, Number 205; and ASTM Standard E857-87. This study was submitted to fulfill the OPPTS 850.2200 guideline requirement. Deviations from OPPTS Guideline No. 850.2200 included:

1. Pre-test mortality was not reported.
2. It was unclear if a brooder temperature gradient was provided.
3. It was unclear if food consumption was measured frequently enough.
4. Although not required, statistical analyses were not performed on body weight data.
5. Although not required, necropsy of birds is recommended.

These deviations do not affect the scientific soundness of the study.

COMPLIANCE: Signed and dated GLP, Quality Assurance and Data Confidentiality statements were provided.

A. MATERIALS:

1. Test Material	AE 0172747 Technical
Description:	Beige powder
Lot No./Batch No. :	OP 2250027/PFI 0215
Purity:	94.0% (w:w)
Stability of Compound Under Test Conditions:	Verified for the 5-day exposure period (day 5 recoveries of 90-99% of day 0 values).
Storage Conditions of Test Chemicals:	Ambient conditions

Physicochemical properties of AE 0172747 Technical.

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 (common and scientific names): Mallard duck (*Anas platyrhynchos*)
(EPA recommends using either bobwhite quail or mallard duck.)

Age at study initiation: 10 days old
(EPA recommends: 10-14 days old)

Weight at study initiation (mean and range): 141-195 g

Source: Whistling Wings, Inc., Hanover, IL

B. STUDY DESIGN:

1. Experimental Conditions

a. Range-finding Study: None reported. The dietary concentrations were established based upon known toxicity data and information supplied by the Sponsor.

b. Definitive Study:

Table 1: Experimental Parameters

Parameter	Details	Remarks *Criteria*
Acclimation Period:	10 days (hatch until study initiation)	All birds were observed daily during acclimation, and any birds exhibiting abnormal behavior or physical injury during acclimation were not used for the study.
Conditions: (same as test or not)	Same as test	
Feeding:	Laboratory-formulated game bird ration and tap water, *ad libitum*	Water from the town of Easton public water supply was provided *ad libitum.*
Health: (any mortality observed)	The birds appeared to be in good health at test initiation.	

Parameter	Details	Remarks *Criteria*
Pen size and construction materials	Vinyl-coated wire grid cages measuring 62 x 92 x 25.5 cm.	Birds were housed in groups of five. The floor area was approximately 1141 cm^2 per bird, fulfilling the minimum recommended size of at least 600 cm^2 per duckling. *Recommended pen size is about 35 x 100 x 24 cm*
Test duration	5 days with treated feed followed by 3 days with untreated feed	*Recommended test duration is 5 days with treated feed and at least 3 days observation with "clean" feed.*
Test concentrations nominal: measured:	0 (control), 562, 1000, 1780, 3160, and 5620 mg ai/kg diet <50 (<LOD, control), 580, 1040, 1890, 3220, and 5790 mg ai/kg diet	*Five or six test concentrations should be used in a geometric scale, unless the LC_{50} > 5000 mg ai/kg diet.*
Solvent/vehicle, if used type: amount:	Acetone Approx. 1.1% by weight	Reviewer-calculated using the density of acetone as 0.79 g/ml (CRC Handbook). *Recommended solvents include distilled water, corn oil, propylene glycol, 1% carboxymethylcellulose, or gum arabic. The solvent should not be more than 2%.*
Diet preparation and feeding	The appropriate amount of test substance was dissolved in acetone for approx. 3 minutes with a magnetic stir plate, a the solution was combined with 5000 g of basal ration and mixed for 15 minutes using a Hobart mixer. The remaining basal ration (approx. 4000 g) was added and the contents were mixed an additional 6 minutes. The prepared feed was transferred to paper feed bags. An amount of diet sufficient to last the 5-day exposure period was prepared on the day of test initiation.	The acetone was allowed to volatilize from the diets during the mixing procedure. *The control group should be tested with a diet containing the maximum amount of vehicle used in treated diets.*

Parameter	Details	Remarks *Criteria*
Feed withholding period	None	
Stability and homogeneity of test material in the diet determined (Yes/No)	Yes	
Number of birds per replicate/groups for negative control: for vehicle control: for treated:	 N/A 5 birds/replicate 5 birds/replicate	 *The recommended number of birds per replicate is a minimum of ten.*
Number of replicates/group (if used) for negative control: for vehicle control: for treated:	 N/A 6 replicates 2 replicates/level	
Test conditions temperature: relative humidity(%): photoperiod:	 Brooder: 30 ± 1°C Room: 23.8 ± 0.6°C Room: 68 ± 7% 16 hours light/8 hours dark	It is recommended that a temperature gradient in the pen of approximately 22 to 38°C is provided to allow young birds to seek a proper temperature. Light intensity averaged 206 lux (19 foot candles). ***Recommended brooder temperature is*** *about 35 °C (95 °F)* ***Recommended room temperature is*** *22-27 °C (71-81 °F)* ***Recommended relative humidity is*** *30-80%* ***Recommended photoperiod*** *is a minimum of 14 hours of light.*
Reference chemical, if used	N/A	

2. Observations:

Table 2: Observations

Parameters	Details	Remarks
Parameters measured (mortality/body weight/ mean feed consumption/ others)	- Mortality - Clinical signs of toxicity - Body weight - Food consumption	
Indicate the stability and homogeneity of	Stability: stability of the test	

Parameters	Details	Remarks
test chemical in the diet	material in avian diet was assessed after 5 days of feed-trough storage from all levels. Recoveries were 90-99% of initial concentrations (day 0). Homogeneity: homogeneity was assessed by collecting samples from the top, middle, and bottom left and right areas (six samples) from treated feed prepared at 562 and 5620 mg ai/kg diet. Coefficients of variation were 1.58 and 1.88%, respectively.	
Indicate if the test material was regurgitated	No regurgitation was indicated.	
Treatments on which necropsies were performed	None performed	Although not required, gross necropsies are recommended.
Observation intervals	Birds were observed four times on day 0, and twice daily thereafter for mortality and clinical signs of toxicity. Body weights were determined on days 0, 5, and 8. Feed consumption was calculated for days 0-5 (exposure) and 6-8 (recovery).	It was unclear if food consumption was measured frequently enough. Food consumption should be measured daily or every other day in the second highest and second lowest concentration and control pens. Food consumption should be estimated for the exposure period and post-exposure period for the remaining pens. In this study, food consumption was determined the same way for all test levels, by recording the change in the weight of the feed presented to the birds "over a given period of time", and was presented as average values for the exposure and post-exposure periods.
Were raw data included?	Yes	

II. RESULTS AND DISCUSSION:

A. MORTALITY:

No mortality was observed in the control or any treatment group. The 8-day acute dietary LC_{50} was >5620 mg ai/kg diet.

Table 3: Effect of AE 0172747 Technical on Mortality of Mallard Duck.

Treatment, mg ai/kg diet Mean-measured (and nominal) conc.		No. of birds per treatment	Cumulative mortality				
			day 1	day 2	day 3	day 4	day 8
Control		30	0	0	0	0	0
580 (562)		10	0	0	0	0	0
1040 (1000)		10	0	0	0	0	0
1890 (1780)		10	0	0	0	0	0
3220 (3160)		10	0	0	0	0	0
5790 (5620)		10	0	0	0	0	0
NOAEC		5620 mg ai/kg diet (nominal)					
LC_{50}		>5620 mg ai/kg diet (nominal)					
Reference chemical	mortality	N/A					
	LC_{50}	N/A					
	NOEC	N/A					

B. SUB-LETHAL TOXICITY ENDPOINTS:

All birds in all treatment groups were normal in appearance and behavior throughout the test.

Based on visual inspection of the data, a dose-responsive treatment-related reduction in body weight gain was observed in birds from all treatment levels compared to the control during the exposure period. Body weight changes from days 0-5 averaged 155 g for the control group, compared to 113, 62, 34, 34, and 10 g for the 562, 1000, 1780, 3160, and 5620 mg ai/kg diet levels, respectively. While there was some compensatory weight gain during the post-exposure observation period, when compared to the control group, mean body weights were still reduced in all treatment groups over the 8-day study period. The NOAEC for body weight effects was <562 mg ai/kg diet.

Based on visual inspection of the data, a treatment-related effect in food consumption was observed in birds from the 1780, 3160, and 5620 mg ai/kg diet levels compared to controls during the exposure period. Food consumption during exposure averaged 106, 110, 97, 71, 80, and 83 g/bird/day for birds in the control, 562, 1000, 1780, 3160, and 5620 mg ai/kg diet levels, respectively. The NOAEC for food consumption was 1000 mg ai/kg diet.

Table 4: Sublethal Effect of AE 0172747 Technical on Mallard Duck.

Treatment, mg ai/kg diet Mean-measured (and nominal) conc.		Observation							
		Body weight change, g			Food consumption, g/bird/day		Other Endpoints		
		days 0-5	days 5-8	days 0- 8	days 0-5	days 6-8	day x1	day xn	% affected
Control		155	98	252	106	168	N/A		
580 (562)		113[(a)]	105	218	110	174	N/A		
1040 (1000)		62[(a)]	120	182	97	186	N/A		
1890 (1780)		34[(a)]	117	151	71[(a)]	177	N/A		
3220 (3160)		34[(a)]	121	154	80[(a)]	176	N/A		
5790 (5620)		10[(a)]	122	132	83[(a)]	179	N/A		
NOAEC		<580 mg ai/kg diet			1000 mg ai/kg diet		N/A		
EC_{50}		Not reported			Not reported				
Reference chemical	NOAEC	N/A							
	EC_{50}	N/A							

[(a)] Visually determined to be a treatment-related reduction.

C. REPORTED STATISTICS:

As no mortality occurred, statistical analyses were not necessary to determine the 8-day LC_{50}. Body weight and food consumption effects were determined from visual inspection of the data. Results were provided in terms of nominal concentrations.

D. VERIFICATION OF STATISTICAL RESULTS:

Statistical Method: The reviewer statistically analyzed the results for body weight and food consumption. Data for both endpoints satisfied the assumptions of normality and homogeneity of variances. The NOAEC values were determined using ANOVA, followed by William's (body weight) or Bonferroni's t-test (food consumption). These analyses were conducted using Toxstat statistical software. There was no mortality, so the LC_{50} was visually determined.

LC_{50}: >5790 mg ai/kg diet 95% C.I.: N/A

NOAEC: 1040 mg ai/kg diet

Probit Slope: N/A 95% C.I.: N/A

Endpoints affected: Body weight change (total; days 0-8)

E. STUDY DEFICIENCIES:

There were no study deficiencies.

F. REVIEWER'S COMMENTS:

The reviewer's results differed from the study authors'. The reviewer's statistical analysis did not detect any significant differences for food consumption and body weight was not significantly reduced at the lowest treatment level. Because the study authors' conclusions (based on visual analysis) are toxicologically more conservative (and, likely, biologically relevant), they are reported in the Executive Summary and Conclusions section.

The estimated dietary intake was 277, 500, 698, 1397, and 2760 mg ai/kg bw/day for the 562, 1000, 1780, 3160, and 5620 mg ai/kg diet levels, respectively.

Concurrent with the sample analysis, procedural recoveries were determined. In avian feed treated with AE 0172747 Technical at 100, 2000, and 6000 mg ai/kg diet, mean recoveries ranged from 100-103%.

In-life dates were August 29 to September 6, 2002.

G. CONCLUSIONS:

This study is scientifically sound. In addition, no significant deviations from OPPTS 850.2200 guidance were observed, and therefore, this study is classified as ACCEPTABLE.

LC_{50}: >5790 mg ai/kg diet 95% C.I.: N/A
Probit Slope: N/A 95% C.I.: N/A

NOAEC: <580 mg ai/kg diet
LOAEC: 580 mg ai/kg diet

Endpoint(s) affected: body weight and food consumption
Most sensitive endpoint: body weight

III. REFERENCES:

U.S. Environmental Protection Agency. 1996. Series 850 - Ecological Effects Test Guidelines (*draft*), OPPTS Number 850.2200: *Avian Dietary Toxicity Test.*

U.S. Environmental Protection Agency. 1982. *Pesticide Assessment Guidelines, FIFRA Subdivision E, Hazard Evaluation: Wildlife and Aquatic Organisms,* subsection 71-2. Environmental Protection Agency. Office of Pesticide Programs. Washington, D.C.

Organization for Economic Cooperation and Development. 1984. *Avian Dietary Toxicity Test.* OECD Guideline for Testing of Chemicals. Guideline 205. Paris.

American Society for Testing and Materials. 1987. *Standard Practice for Conducting Subacute Dietary Toxicity Tests with Avian Species.* ASTM Standard E857-87. Annual Book of ASTM Standards, Vol. 11.04. Philadelphia, PA.

National Research Council. 1996. *Guide for the Care and Use of Laboratory Animals.* Washington, DC. National Academy Press. 125 pp.

Stephan, C.E. 1977. Methods for Calculating an LC50. Pages 65-84 In *Aquatic Toxicology and Hazard Evaluations.* American Society for Testing and Materials, Pub. No. STP 634. Philadelphia, PA.

Finney, D.J. 1971. *Statistical Methods in Biological Assay,* 2nd ed., Griffin Press, London.

Thompson, W.R. 1947. *Bacteriological Reviews*, Vol. 2, No. 2: 115-145.

Stephan, C.E. 1978. U.S. EPA, Environmental Research Laboratory, Duluth, MN. Personal Communication.

APPENDIX I. OUTPUT OF REVIEWER'S STATISTICAL VERIFICATION:

total body weight change
File: 5502w Transform: NO TRANSFORMATION

ANOVA TABLE

SOURCE	DF	SS	MS	F
Between	5	34934.604	6986.921	13.938
Within (Error)	10	5012.833	501.283	
Total	15	39947.438		

Critical F value = 3.33 (0.05,5,10)
Since F > Critical F REJECT Ho:All groups equal

total body weight change
File: 5502w Transform: NO TRANSFORMATION

BONFERRONI T-TEST - TABLE 1 OF 2 Ho:Control<Treatment

GROUP	IDENTIFICATION	TRANSFORMED MEAN	MEAN CALCULATED IN ORIGINAL UNITS	T STAT	SIG
1	control	250.667	250.667		
2	562	218.500	218.500	1.760	
3	1000	181.500	181.500	3.784	*
4	1780	151.000	151.000	5.452	*
5	3160	154.500	154.500	5.261	*
6	5620	132.000	132.000	6.491	*

Bonferroni T table value = 2.76 (1 Tailed Value, P=0.05, df=10,5)

total body weight change
File: 5502w Transform: NO TRANSFORMATION

BONFERRONI T-TEST - TABLE 2 OF 2 Ho:Control<Treatment

GROUP	IDENTIFICATION	NUM OF REPS	Minimum Sig Diff (IN ORIG. UNITS)	% of CONTROL	DIFFERENCE FROM CONTROL
1	control	6			
2	562	2	50.528	20.2	32.167
3	1000	2	50.528	20.2	69.167
4	1780	2	50.528	20.2	99.667
5	3160	2	50.528	20.2	96.167
6	5620	2	50.528	20.2	118.667

total body weight change
File: 5502w Transform: NO TRANSFORMATION

WILLIAMS TEST (Isotonic regression model) TABLE 1 OF 2

GROUP	IDENTIFICATION	N	ORIGINAL MEAN	TRANSFORMED MEAN	ISOTONIZED MEAN
1	control	6	250.667	250.667	250.667
2	562	2	218.500	218.500	218.500
3	1000	2	181.500	181.500	181.500
4	1780	2	151.000	151.000	152.750
5	3160	2	154.500	154.500	152.750
6	5620	2	132.000	132.000	132.000

total body weight change
File: 5502w Transform: NO TRANSFORMATION

WILLIAMS TEST (Isotonic regression model) TABLE 2 OF 2

IDENTIFICATION	ISOTONIZED MEAN	CALC. WILLIAMS	SIG P=.05	TABLE WILLIAMS	DEGREES OF FREEDOM
control	250.667				
562	**218.500**	**1.760**		**1.81**	**k= 1, v=10**
1000	181.500	3.784	*	1.91	k= 2, v=10
1780	152.750	5.356	*	1.94	k= 3, v=10
3160	152.750	5.356	*	1.96	k= 4, v=10
5620	132.000	6.491	*	1.97	k= 5, v=10

s = 22.389
Note: df used for table values are approximate when v > 20.

food consumption
File: 5502f Transform: NO TRANSFORMATION

ANOVA TABLE

SOURCE	DF	SS	MS	F
Between	5	2986.417	597.283	2.067
Within (Error)	10	2889.333	288.933	
Total	15	5875.750		

Critical F value = 3.33 (0.05,5,10)
Since F < Critical F FAIL TO REJECT Ho:All groups equal

food consumption
File: 5502f Transform: NO TRANSFORMATION

BONFERRONI T-TEST - TABLE 1 OF 2 Ho:Control<Treatment

GROUP	IDENTIFICATION	TRANSFORMED MEAN	MEAN CALCULATED IN ORIGINAL UNITS	T STAT	SIG
1	control	105.333	105.333		
2	562	110.000	110.000	-0.336	
3	1000	97.000	97.000	0.600	
4	1780	71.000	71.000	2.474	
5	3160	80.000	80.000	1.825	
6	5620	83.000	83.000	1.609	

Bonferroni T table value = 2.76 (1 Tailed Value, P=0.05, df=10,5)

food consumption
File: 5502f Transform: NO TRANSFORMATION

BONFERRONI T-TEST - TABLE 2 OF 2 Ho:Control<Treatment

GROUP	IDENTIFICATION	NUM OF REPS	Minimum Sig Diff (IN ORIG. UNITS)	% of CONTROL	DIFFERENCE FROM CONTROL
1	control	6			
2	562	2	38.361	36.4	-4.667
3	1000	2	38.361	36.4	8.333
4	1780	2	38.361	36.4	34.333
5	3160	2	38.361	36.4	25.333
6	5620	2	38.361	36.4	22.333

food consumption
File: 5502f Transform: NO TRANSFORMATION

WILLIAMS TEST (Isotonic regression model) TABLE 1 OF 2

GROUP	IDENTIFICATION	N	ORIGINAL MEAN	TRANSFORMED MEAN	ISOTONIZED MEAN
1	control	6	105.333	105.333	106.500
2	562	2	110.000	110.000	106.500
3	1000	2	97.000	97.000	97.000
4	1780	2	71.000	71.000	78.000
5	3160	2	80.000	80.000	78.000
6	5620	2	83.000	83.000	78.000

food consumption
File: 5502f Transform: NO TRANSFORMATION

WILLIAMS TEST (Isotonic regression model) TABLE 2 OF 2

IDENTIFICATION	ISOTONIZED MEAN	CALC. WILLIAMS	SIG P=.05	TABLE WILLIAMS	DEGREES OF FREEDOM
control	106.500				
562	106.500	0.084		1.81	k= 1, v=10
1000	97.000	0.600		1.91	k= 2, v=10
1780	78.000	1.969	*	1.94	k= 3, v=10
3160	78.000	1.969	*	1.96	k= 4, v=10
5620	78.000	1.969		1.97	k= 5, v=10

s = 16.998
Note: df used for table values are approximate when v > 20.