Data Evaluation Report on the Acute Dietary Toxicity of Pyrasulfotole to Avian Species {*Anas platyrhynchos*}

PMRA Submission Number 2006-2445 EPA MRID Number 468017-31

Data Requirement:	PMRA Data Code	9.6.2.4
	EPA DP Barcode	D328639
	OECD Data Point	IIA 8.1.2
	EPA MRID	468017-31
	EPA Guideline	850.2200

Test material: Pyrasulfotole Purity: 95.4%
Common name: AE 0317309
Chemical name: IUPAC: (5-hydroxy-1,3-dimethylpyrazol-4-yl)(2-mesyl-4-trifluoromethylphenyl)methanone
CAS name: Not reported
CAS No.: Not reported
Synonyms: Not reported

Primary Reviewer: Rebecca Bryan **Signature:** Rebecca L. Bryan
Staff Scientist, Dynamac Corporation **Date:** 5/18/06

Secondary Reviewer: Teri S. Myers **Signature:** Teri S Myers
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Primary Reviewer: Melissa Panger **Date:** 7-7-06
EPA

Secondary Reviewer: J.D. Whall (Officer No. 1268) **Date:** 11/21/06
PMRA J.D. Whall

Secondary Reviewer(s): David McAdam **Date:** 6 Nov 2006
Australian Government Department of the Environment and Heritage (DEH). D. McAdam

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

Company Code BCZ
Active Code PSA
Use Site Category: 13, 14**EPA PC Code** 000692

Date Evaluation Completed: 11-27-2006

CITATION: Stoughton, T.L. 2005. Technical AE0317309: A Subacute Dietary LC_{50} with Mallards. Unpublished study performed by Bayer Corporation, Agriculture Division, Research and Development Department, Environmental Research and Toxicology, Stilwell, Kansas. Study No. A9720801/EBAIM003. Study sponsored by Bayer CropScience, Research Triangle Park, NC. The final report issued October 14, 2005.

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.

Data Evaluation Report on the Acute Dietary Toxicity of Pyrasulfotole to Avian Species {*Anas platyrhynchos*}

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EXECUTIVE SUMMARY:

The acute dietary toxicity of Pyrasulfotole to 10-day old Mallards, *Anas platyrhychos,* was assessed over 8 days (5 days on treated feed and 3 day recovery period). Pyrasulfotole was administered to the birds in the diet at mean measured concentrations of 306, 605, 1275, 2657, and 5089 mg a.i./kg dw of diet (75, 143, 280, 597, and 1,067 mg a.i./kg bw daily dietary dose, respectively). The acute dietary LC_{50} was >5089 mg ai/kg diet. The NOAEC was 5089 mg a.i./kg diet. According to the US EPA classification, Pyrasulfotole would be classified as practically non-toxic to Mallards on an acute dietary basis.

There were no mortalities in the control or treatment groups. No treatment-related clinical signs of toxicity were observed during the study. There were no adverse effects on body weights or feed consumption during the exposure portion of the study. During the recovery period, there were statistically significant reductions in body weight gain (68-88%) at the 605, 1275, and 2657 mg a.i./kg diet treatment levels, but not the highest treatment level (5089 mg a.i./kg diet). Furthermore, there were no statistically significant differences in the body weight of the different treatment groups at the study termination. Therefore, the observed reduction in growth rate for the middle three treatment levels during the recovery period was not thought to adversely affect mallard growth.

Additionally, several birds in the 605, 1275, 2657, and 5089 treatment groups, after being necropsied, had visible yolk stalk diverticulums. The study author considered this a normal finding for mallards, however, the potential biological significance of this finding is unknown.

This toxicity study is classified as **ACCEPTABLE**, is scientifically sound, and does satisfy the guideline requirement for an acute dietary toxicity study with mallard ducks.

Results Synopsis

Test Organism Size/Age (Mean Weight): 10 days old, 155.0-163.4 g (treatment means), 136.8-172.3 g (range)

LC_{50}: >5089 mg ai/kg diet 95% C.I.: N/A

NOAEC: 5089 mg a.i./kg diet
Probit Slope: Not calculable 95% C.I.: N/A

Endpoint(s) affected: None

I. MATERIALS AND METHODS

GUIDELINE FOLLOWED: This study was based on procedures of EPA 71-2, OPPTS 850.2200 and OECD Guideline No. 205. The following deviations from U.S. Environmental Protection Agency Series 850-Ecological Effects Test Guidelines (*draft*), OPPTS Number 850.2200, *Avian dietary toxicity test* were noted:

No deviations were noted.

COMPLIANCE: Signed and dated GLP, Quality Assurance and No Data Confidentiality statements were provided. The test was conducted according to the US EPA-FIFRA Good Laboratory Practice (40 CFR Part 160).

A. MATERIALS:

1. Test Material Pyrasulfotole (AE 0317309)

Description: Light brown powder

Lot No./Batch No. : OP 1-4

Purity: 95.4%

Stability of Compound Under Test Conditions: Stability of the test material was determined in the 313 and 5000 mg a.i./kg feed after one day. The recoveries were 96-97% of the initial concentrations.

Storage Conditions of Test Chemicals: Stored under ambient conditions.

Physicochemical properties of Pyrasulfotole

Parameter	Value	Comment
Molecular weight	362.3 g/mol	
Water Solubility (g/L) at 20°C	4.2 at pH 4 69.1 at pH 7 49.0 at pH 9	Very soluble
Vapor Pressure/Volatility	2.7×10^{-7} Pa at 20°C 6.8×10^{-7} Pa at 25°C	Non-volatile
UV Absorption	water $\lambda_{max} = 264$ 0.1M HCl $\lambda_{max} = 241$ 0.1M NaOH $\lambda_{max} = 216$	Not likely to undergo photolysis.
Pka	4.2 ± 0.15	
log K_{ow} at 23°C	0.276 at pH 4 -1.362 at pH 7 -1.58 at pH 9	Not likely to bioaccumulate
Stability of compound at room temperature, if provided		No significant degradation over 12 months at ambient temperatures.

Data obtained from pyrasulfatole chemistry review of Submission 2006-2445.

2. Test organism:

Species (common and scientific names): Mallard duck, *Anas platyrhychos*
(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): 155.0-163.4 g (treatment means), 136.8-172.3 g (range)

Source: Whistling Wings, Hanover, Illinois.

B. STUDY DESIGN:

1. Experimental Conditions

a. Range-finding Study: No range-finding study was reported.

b. Definitive Study:

Table 1: Experimental Parameters

Parameter	Details	Remarks *Criteria*
Acclimation Period: Conditions: (same as test or not) Feeding: Health: (any mortality observed)	 6 days Same as test Teklad Bayer Starter Ration and local tap water were provided *ad libitum*. Birds that appeared healthy were used for testing. There was one mortality during acclimation.	
Pen size and construction materials	Galvanized steel brooders (91L× 81W× 25H cm).	*Recommended pen size is about 35 x 100 x 24 cm*
Test duration	5 days with treated feed, and 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:	 313, 625, 1250, 2500, and 5000 mg a.i./kg 306, 605, 1275, 2657, and 5089 mg a.i./kg	*Five or six test concentrations should be used in a geometric scale, unless the LC_{50} > 5000 mg ai/kg diet.*

Parameter	Details	Remarks *Criteria*
Solvent/vehicle, if used type: amount:	N/A	*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 chemical and feed were added into mixing bowl and blended for 5 minutes in laboratory mixer. The premix was added to additional feed and mixed in laboratory mixers for a total of 20 minutes.	*The control group should be tested with a diet containing the maximum amount of vehicle used in treated diets.*
Feed withholding period	None	
Stability and homogeneity of test material in the diet determined (Yes/No)	Yes	See below
Number of birds per replicate/groups for negative control: for vehicle control: for treated:	 10 N/A 10	*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:	 1 N/A 1	
Test conditions temperature: relative humidity(%): photoperiod:	 22EC for room temperature and 32-38EC for brooder temperatures. 55% 14 hours light/10 hours dark	***Recommended brooder temperature is*** *about 35EC (95EF)* ***Recommended room temperature is*** *22-27EC (71-81EF)* ***Recommended relative humidity is*** *30-80%* ***Recommended photoperiod*** *is a minimum of 14 hours of light.*
Reference chemical, if used	None	

2. Observations:

Table 2: Observations

Parameters	Details	Remarks
Parameters measured (mortality/body weight/ mean feed consumption/ others)	- Mortality - Clinical signs of toxicity - Mean feed consumption (g/bird/day) - Mean body weight	
Indicate the stability and homogeneity of test chemical in the diet	Stability: Stability of the test material in feed was assessed in treated feed prepared at 313 and 5000 ppm from one day in the brooder during the test and from 14 days in the freezer. The brooder recoveries were 96-97% of the initial concentrations. The freezer recoveries were 93-99% of the initial concentrations. Homogeneity: Homogeneity of the test material in feed was assessed in treated feed prepared at 313 and 5000 ppm. Recoveries were 95-121% of the nominal concentrations.	
Indicate if the test material was regurgitated	No regurgitation was reported.	
Treatments on which necropsies were performed	All surviving birds were necropsied.	Several birds in the 605, 1275, 2657, and 5089 treatment groups had visible yolk stalk diverticulums; the author considered this a normal finding for mallards and it was not considered an adverse effect.
Observation intervals	Mortality and signs of toxicity: Determined three times on Day 0 and daily (1 to 2 times) thereafter. Feed consumption: Determined daily Body Weight: Days -3, 0, 5, and 8	
Were raw data included?	Yes	

II. RESULTS AND DISCUSSION:

A. MORTALITY:

There were no mortalities in the control or treatment groups. The NOAEC based on mortality was ≥5089 mg a.i./kg.

Table 3: Effect of Pyrasulfotole on Mortality of *Anas platyrhychos*

Treatment (mg ai/kg diet) measured (nominal) conc.		No. of birds per treatment	Cumulative mortality							
			day 1	day 2	day 3	day 4	day 5	day 6	day 7	day 8
Control		10	0	0	0	0	0	0	0	0
306 (313)		10	0	0	0	0	0	0	0	0
605 (625)		10	0	0	0	0	0	0	0	0
1275 (1250)		10	0	0	0	0	0	0	0	0
2657 (2500)		10	0	0	0	0	0	0	0	0
5089 (5000)		10	0	0	0	0	0	0	0	0
NOAEC		≥5089 mg a.i./kg								
LC_{50}		>5089 mg a.i./kg								
Reference chemical	mortality	N/A								
	LC_{50}	N/A								
	NOEC	N/A								

B. SUB-LETHAL TOXICITY ENDPOINTS:

No treatment-related clinical signs of toxicity were observed during the study. There were no adverse effects on body weights or feed consumption during the exposure portion of the study. Significant reductions in body weight gain were detected in the 605, 1275, and 2657 mg a.i./kg diet treatment groups. Because these effects were not dose dependent, and because body weight at the end of the study did not differ among treatment groups the NOAEC based on body weights and feed consumption was determined to be ≥5089 mg a.i./kg diet.

No treatment-related findings were observed in postmortem examinations. Several birds in the 605, 1275, 2657, and 5089 treatment groups had visible yolk stalk diverticulums; the author considered this a normal finding for mallards and it was not considered an adverse effect.

Table 4: Sublethal Effect of Pyrasulfotole on *Anas platyrhychos*

Treatment (mg ai/kg diet) measured (nominal) conc.		Observation				
		Mean body weight (g)			Food consumption (g/bird/day)	
		Day			Day	
		0	5	8	0-5	6-8
Control		155.0	273.4	316.5	46.4	43.1
306 (313)		155.4	279.9	324.5	53.2	48.7
605 (625)		159.5	277.4	288.8	51.8	31.5
1275 (1250)		155.5	281.2	294.8	48.0	38.2
2657 (2500)		163.4	289.7	294.3	50.9	34.6
5089 (5000)		160.0	299.0	336.2	48.1	50.9
NOAEC		≥5089 mg a.i./kg			≥5089 mg a.i./kg	
EC_{50}		Not determined			Not determined	
Reference chemical	NOEC	N/A				
	EC_{50}	N/A				

C. REPORTED STATISTICS:

The LC_{50} could not be calculated because there were no mortalities. The bodyweight and growth data were analyzed using the chi-square test for normality and the Levene's test for homogeneity of variance. The body weight treatment group data were compared to the control using Dunnett's one-tailed test ($p>0.05$). The statistical analyses on body weight were conducted using the TOXSTAT version 3.4 computer program. The study author-reported NOAEC was 5089 mg a.i./kg feed. Mean measured concentrations were used in all estimations. Feed consumption data were not analyzed statistically.

D. VERIFICATION OF STATISTICAL RESULTS:

Statistical Method: With the exception of the 605 mg ai/kg diet treatment group, body weight gain was higher in the treatment groups than in the control group during the exposure period; the reduction in the 605 mg ai/kg diet group was less than 1%. As a result, the toxicity values for body weight gain during the exposure period could be visually determined. Body weight gain during the recovery period was analyzed using ANOVA, followed by Dunnett's test, after confirming that data satisfied the assumptions of normality and homogeneity of variances; this analysis was conducted using Toxstat statistical software.

LC_{50}: >5089 mg ai/kg diet 95% C.I.: N/A

NOAEC: 5089 mg a.i./kg diet

Probit Slope: Not calculable 95% C.I.: N/A

E. STUDY DEFICIENCIES:

There were no study deficiencies.

F. REVIEWERS' COMMENTS:

Results of the reviewers' statistical verification and conclusions were similar to the study author's. Both the reviewers and study author detected significant reductions in body weight gain in the 605, 1275, and 2657 mg ai/kg diet treatment groups during the recovery period. The study author dismissed these substantial reductions in weight gain (68-88%) because they were not dose dependent. The observed reductions in body weight gain during the recovery phase coincided with a reduction in feed consumption at those treatment levels. It is possible that there was some food avoidance occurring at these middle doses, however it is unlikely to be treatment related as food consumption and growth were not affected during the actual exposure period. The reviewers therefore agree with the study author that the observed decrease in growth rate at the middle concentrations is not a biologically significant sublethal effect, and suggest that the NOAEC is 5089 mg a.i./kg diet.

G. CONCLUSIONS:

This study is scientifically sound and is classified as **ACCEPTABLE**. The NOAEC was 5089 mg a.i./kg diet and the LC_{50} was >5089 mg a.i./kg diet, the highest treatment group, which categorizes pyrasulfotole as practically non-toxic to Mallard ducks on an acute dietary basis.

LC_{50}: >5089 mg ai/kg diet 95% C.I.: N/A
NOAEC: 5089 mg a.i./kg diet
Endpoint(s) affected: None

III. REFERENCES:

American Society of Testing and Materials (ASTM). 1993. Standard Practice for Conducting Subacute Dietary Tests with Avian Species. ASTM Standard E857-87.

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APPENDIX I. OUTPUT OF REVIEWER'S STATISTICAL VERIFICATION:

body weight gain (recovery)
File: 1731b Transform: NO TRANSFORMATION

ANOVA TABLE

SOURCE	DF	SS	MS	F
Between	5	1927.783	385.557	4.839
Within (Error)	54	4302.440	79.675	
Total	59	6230.223		

Critical F value = 2.45 (0.05,5,40)
Since F > Critical F REJECT Ho:All groups equal

body weight gain (recovery)
File: 1731b Transform: NO TRANSFORMATION

DUNNETTS TEST - TABLE 1 OF 2 Ho:Control<Treatment

GROUP	IDENTIFICATION	TRANSFORMED MEAN	MEAN CALCULATED IN ORIGINAL UNITS	T STAT	SIG
1	control	15.603	15.603		
2	306	15.781	15.781	-0.045	
3	605	4.078	4.078	2.887	*
4	1275	5.021	5.021	2.651	*
5	2657	1.866	1.866	3.441	*
6	5089	12.517	12.517	0.773	

Dunnett table value = 2.31 (1 Tailed Value, P=0.05, df=40,5)

body weight gain (recovery)
File: 1731b Transform: NO TRANSFORMATION

DUNNETTS 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	10			
2	306	10	9.221	59.1	-0.178
3	605	10	9.221	59.1	11.525
4	1275	10	9.221	59.1	10.582
5	2657	10	9.221	59.1	13.737
6	5089	10	9.221	59.1	3.086

body weight gain (recovery)
File: 1731b Transform: NO TRANSFORMATION

WILLIAMS TEST (Isotonic regression model) TABLE 1 OF 2

GROUP	IDENTIFICATION	N	ORIGINAL MEAN	TRANSFORMED MEAN	ISOTONIZED MEAN
1	control	10	15.603	15.603	15.692
2	306	10	15.781	15.781	15.692
3	605	10	4.078	4.078	5.871
4	1275	10	5.021	5.021	5.871
5	2657	10	1.866	1.866	5.871
6	5089	10	12.517	12.517	5.871

body weight gain (recovery)
File: 1731b 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	15.692				
306	15.692	0.022		1.68	k= 1, v=54
605	5.871	2.438	*	1.76	k= 2, v=54
1275	5.871	2.438	*	1.79	k= 3, v=54
2657	5.871	2.438	*	1.80	k= 4, v=54
5089	5.871	2.438	*	1.80	k= 5, v=54

s = 8.926
Note: df used for table values are approximate when v > 20.

body weight gain (recovery)
6
10
10
10
10
10
10
control
-0.43
18.43
16.31
18.59
8.44
29.64
24.53
-0.16
25.86
14.82
306
2.21
17.68
19.91
27.32
10.89
12.15
-2.02
24.77
12.26
32.64
605
-2.04
1.65
16.18
8.5
3.25
6.96
4.63
3.1
-3.17
1.72
1275
-4.54
10.15
12.19
8.88
21.41
4.82
-0.43
-8.37
8.19
-2.09
2657
-7.68
-9.14
-16.44
4.99

17.1
1.74
2.45
10.48
10.68
4.48
5089
17.6
19.07
5.83
12.38
1.04
12.83
13.73
15.78
7.59
19.32

				% body weight gain	
	d0	d5	d8	d 0-5	d 5-8
control	165.5	254.7	253.6	53.90	-0.43
	154.9	287.1	340	85.35	18.43
	160.7	312.6	363.6	94.52	16.31
	140.8	245.3	290.9	74.22	18.59
	152.1	273.6	296.7	79.88	8.44
	136.8	248.3	321.9	81.51	29.64
	144.4	280.1	348.8	93.98	24.53
	164.3	248	247.6	50.94	-0.16
	161.3	284.2	357.7	76.19	25.86
	169.1	299.6	344	77.17	14.82
306	162.3	276.4	282.5	70.30	2.21
	151.9	281.6	331.4	85.39	17.68
	148.2	276.3	331.3	86.44	19.91
	162.5	286.6	364.9	76.37	27.32
	149.8	246	272.8	64.22	10.89
	158	278.3	312.1	76.14	12.15
	161.9	271.9	266.4	67.94	-2.02
	161	310.1	386.9	92.61	24.77
	158.7	302.5	339.6	90.61	12.26
	139.7	269.3	357.2	92.77	32.64
605	159.3	288.9	283	81.36	-2.04
	165	297.6	302.5	80.36	1.65
	155.6	266.4	309.5	71.21	16.18
	156.7	284.7	308.9	81.68	8.50
	155.9	252	260.2	61.64	3.25
	157.5	284.6	304.4	80.70	6.96
	170.7	315	329.6	84.53	4.63
	152	277	285.6	82.24	3.10
	159.5	240	232.4	50.47	-3.17
	162.5	267.5	272.1	64.62	1.72
1275	168.2	299.7	286.1	78.18	-4.54
	151.9	270.9	298.4	78.34	10.15
	152.3	280.5	314.7	84.18	12.19
	150.8	262.3	285.6	73.94	8.88
	154.2	286.8	348.2	85.99	21.41
	143.3	275.8	289.1	92.46	4.82
	162.9	302.2	300.9	85.51	-0.43
	161.6	287.8	263.7	78.09	-8.37
	147.9	263.6	285.2	78.23	8.19
	161.4	282.1	276.2	74.78	-2.09
2657	154.9	289.2	267	86.70	-7.68
	180.8	289.9	263.4	60.34	-9.14
	171.5	298.6	249.5	74.11	-16.44
	163.2	306.9	322.2	88.05	4.99
	157.5	262.6	307.5	66.73	17.10
	167.3	292.9	298	75.07	1.74

	172.3	322.7	330.6	87.29	2.45
	160.6	293	323.7	82.44	10.48
	149.5	252.9	279.9	69.16	10.68
	156.3	287.8	300.7	84.13	4.48
5089	150	276.7	325.4	84.47	17.60
	152	271.7	323.5	78.75	19.07
	171	291.6	308.6	70.53	5.83
	165.1	303	340.5	83.53	12.38
	159.7	298.4	301.5	86.85	1.04
	147.9	291.5	328.9	97.09	12.83
	168.8	329.2	374.4	95.02	13.73
	160.9	304.2	352.2	89.06	15.78
	168.2	316.1	340.1	87.93	7.59
	156.6	307.4	366.8	96.30	19.32