Data Evaluation Report on the Acute Oral Toxicity of Hallcomid M-8-10 on Avian Species
Colinus virginianus
PMRA Submission Number EPA MRID Number 45369711

Data Requirement:

PMRA DATA CODE	{.....}
EPA DP Barcode	D284964
OECD Data Point	{.....}
EPA MRID	45369711
EPA Guideline	§71-1

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Test material: Hallcomid M-8-10 **Purity:** 94.4%
Common name: Hallcomid M-8-10
Chemical name: IUPAC: Not reported
CAS name: Mixture N,N-dimethylcapramid and N,N-dimethylcaprylamid
CAS No.: Not reported
Synonyms: Not reported

Primary Reviewer: Dana Worcester
Staff Scientist, Dynamac Corporation
Signature:
Date: 6/9/03

QC Reviewer: Teri Myers
Staff Scientist, Dynamac Corporation
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Primary Reviewer: **Date:**
{EPA/OECD/PMRA}

Secondary Reviewer(s): **Date:**
{EPA/OECD/PMRA}

Reference/Submission No.

Company Code:
Active Code:
EPA PC Code: 999999

Date Evaluation Completed:

CITATION: Grau, R. 1994. Hallcomid M-8-10 (Technical Grade) Acute Oral Toxicity Test to Bobwhite Quail. Unpublished study performed and sponsored by Bayer AG, Leverkusen, Germany and submitted by The C.P. Hall Company, Chicago, IL. Report Number: VB-024. Experimental start date August 24, 1993 and experimental termination September 7, 1993. Final report issued July 25, 1994.

EXECUTIVE SUMMARY:

The acute oral toxicity of Hallcomid M-8-10 to 19-week-old Northern Bobwhite quail (*Colinus virginianus*) was assessed over 14 days. Hallcomid M-8-10 was initially administered to male and female birds by gelatin capsules at nominal concentrations of 200, 400, 800, 1600, and 3200 mg/kg bw. Measured concentrations were not reported.

Mortality was 100, 50, 0, 0, and 0% in the 200, 400, 800, 1600, and 3200 mg/kg bw treatment groups, respectively. The acute oral LD_{50} is 1600 mg/kg bw, which categorizes Hallcomid M-8-10 as slightly toxic to Northern Bobwhite quail on an acute oral basis. In the 1600 and 800 mg/kg bw treatment groups, 100 and 50% of the birds exhibited treatment-related signs of toxicity. The TEC was 570 mg/kg bw. After 14 days, no treatment-related abnormalities were observed upon necropsy.

This toxicity study is scientifically sound and fulfills the guideline requirements for an acute toxicity study using the Northern Bobwhite quail (§71-1). This study is classified as Core.

Results Synopsis

Test Organism Size/Age : 19-week, 163-252 g (combined sexes)

LD_{50}: 1600 mg/kg bw
95% C.I.: 800-3200 mg/kg bw
NOEL: 400 mg/kg bw
Probit Slope: N/A
LOEL: 800 mg/kg bw
TEC: 570 mg/kg bw
Endpoint(s) Affected: Mortality and clinical signs of toxicity

I. MATERIALS AND METHODS

GUIDELINE FOLLOWED: The protocol followed procedures of the United States Environmental Protection Agency Section 71-1. The following deviations were noted:

1. The stability of the compound under test conditions was not reported.

2. The study was conducted using a photoperiod of that corresponded to natural daylight. EPA recommends using a photoperiod of 10 hours light/14 hours dark.

3. Birds were acclimated for 14 days. EPA recommends at least 15 days.

COMPLIANCE: Signed and dated GLP, Quality Assurance, and Data Confidentiality statements were provided. This study was conducted in accordance with OECD GLP.

A. MATERIALS:

1. Test Material Hallcomid M-8-10

Description: Colorless liquid

Lot No./Batch No.: 233290307

Purity: 94.4%

Stability of Compound Under Test Conditions: Not reported.

OECD requires water solubility, stability in water and light, pK_a, P_{ow}, and vapor pressure of the test compound. OECD requirements were not reported.

Storage conditions of test chemicals: Not reported

2. Test organism:

Species: Northern Bobwhite quail (*Colinus virginianus*)

Age at study initiation: 19 weeks

Weight at study initiation: Males: 165-227 g
Females: 163-254 g

Source: H. & E. Küberich Quail Farm, Germany.

B. STUDY DESIGN:

1. Experimental Conditions

a) Range-finding Study: Dose levels were based on a previously conducted range finding study.

b) Definitive Study:

Table 1. Experimental Parameters.

Parameter	Details	Remarks *Criteria*
Acclimation period: Conditions (same as test or not): Feeding: Health (any mortality observed):	14 days Same as test. Municipal water and LA 55 Laying Hen Ration provided *ad libitum*. Mortality was <5% prior to initiation. No deformed or injured birds were used.	*EPA recommends that birds be pre-conditioned to the test facilities for at least 15 days.* *OECD recommends that birds be pre-conditioned to the test facilities for at least 7 days.*
Pen size and construction materials	Cages were constructed of stainless steel wire; 18 x 23 x 13 cm.	*EPA requires: pens must conform to good husbandry practices and should not create crowding stress.* *OECD lists no criteria for pen construction other than stating that pens should be suitable for the captive rearing of that species.*
Test duration	14 Days	*EPA requires a day for dosing and at least 14 days observation.*

Parameter	Details	Remarks Criteria
Dose preparation Indicate method of confirmation of dose	For each dose, the test substance was weighed and placed in gelatin capsules. N/A	
Mode of dose administration	Gelatin capsules	*Gavage or gelatin capsule.*
Dose levels nominal: measured:	200, 400, 800, 1600, and 3200 mg/kg bw Not reported	*EPA requires a minimum of 5 treatment levels unless LD_{50} is demonstrated to be greater than 2000 mg ai/kg*
Solvent/vehicle, if used type: amount/bw:	None N/A	*EPA recommends that the test material be administered without a vehicle if possible. Maximum vehicle should not exceed 0.1 to 1.0% of body weight.*
Number of birds per groups/treatment for negative control: for solvent/vehicle control: for treated:	10 N/A 10	*EPA recommends 10 birds per treatment group and 10 birds for each control and vehicle group.*
No. of feed withholding days before dosing	Birds were fasted for 18 hours prior to dosing.	*EPA recommends that food should be withheld for at least 15 hours prior to dosing.*
Test conditions Temperature:	20 ± 2°C	*EPA recommends that a 10*

Parameter	Details	Remarks
		Criteria
Relative humidity: Photo-period:	30-90% Corresponded to natural daylight.	*hr light/14 hr dark photo-period.*
Reference chemical, if used name: concentrations tested:	None used.	

2. Observations:

Table 2: Observations.

Parameter	Details	Remarks/Criteria
Parameters measured		
Parameters measured (mortality/individual body weight at test initiation and termination/ mean feed consumption/others)	- Mortality - Clinical signs of toxicity - Individual body weight - Average feed consumption	*EPA recommends:* *Body weight measured at test initiation, on Day 14 and at end of the test if the test is extended beyond 14 days.* *Calculation of mortality.* *Mortality must NOT be more than 10% in controls.* *Feed consumption may be measured as average daily food consumption.*
Indicate if the test material was regurgitated	No regurgitation was reported.	*Regurgitation is an indication that the dose was rejected. The test may have to be repeated if the problem persists.*
Groups on which necropsies were performed	All surviving birds in the 800 and 1600 mg/kg bw treatment groups and on all birds that died.	*EPA recommends that gross necropsies be performed with inspections of the GI tract, liver, kidneys, heart, and spleen.*

Parameter	Details	Remarks/Criteria
Observation intervals	Mortality and signs of toxicity were determined hourly on day 1, once daily thereafter. Body Weight: Days -1, 7, 14 Feed consumption: individually 3 days, 7 days, and 4 days	
Were raw data included?	Raw data were included.	

II. RESULTS AND DISCUSSION:

A. MORTALITY:

Mortality was 100, 50, 0, 0, and 0% in the 200, 400, 800, 1600, and 3200 mg/kg bw treatment groups, respectively.

Table 3: Effect of Hallcomid M-8-10 on mortality of *Colinus virginianus*.

Treatment (mg/kg bw)		No. of birds	Cumulative mortality							
			day 1	day 2	day 5	day 7	day 9	day 11	day 13	day 14
Control		10	0	0	0	0	0	0	0	0
200		10	0	0	0	0	0	0	0	0
400		10	0	0	0	0	0	0	0	0
800		10	0	0	0	0	0	0	0	0
1600		10	5	5	5	5	5	5	5	5
3200		10	10	10	10	10	10	10	10	10
NOEL		400 mg/kg bw								
LD_{50}		1600 mg/kg bw								
Reference chemical	mortality	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A
	LD_{50}	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A
	NOEL	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A

B. SUB-LETHAL TOXICITY ENDPOINTS:

100 and 50% of the birds in the 1600 and 800 mg/kg bw treatment groups, respectively exhibited treatment-related signs of toxicity such as convulsions, diarrhea, ptosis, loss of equilibrium and apathy. No other birds displayed symptoms.

Weights of the 1600 mg/kg bw birds was significantly higher than the control at test termination. There were no other statistically significant weight differences between the control and treatment groups.

No gross abnormalities were detected in chicks examined at study termination.

Table 4: Sublethal effects of Hallcomid M-8-10 on *Colinus virginianus*.

Mean Body Weight, g					
Treatment, mg/kg bw		Males		Females	
		Day -1	Day 14	Day -1	Day 14
Control		175.4 ± 10.5	168.4 ± 13.7	195.2 ± 22.8	192.4 ± 19.4
200		187.4 ± 12.6	184.0 ± 12.4	183.6 ± 9.4	182.0 ± 8.9
400		197.4 ± 19.6	189.2 ± 15.3	197.0 ± 17.8	192.2 ± 17.0
800		184.4 ± 10.5	183.4 ± 7.2	189.6 ± 6.5	188.4 ± 5.4
1600		196.6 ± 13.6	202.5 ± 7.8	205.4 ± 27.0	218.7 ± 28.0
3200		199.2 ± 16.1	N/A	201.6 ± 32.6	N/A
NOEL		400 mg/kg bw			
EC_{50}		1600 mg/kg bw			
Reference chemical	effect: NOEL: LD_{50}:	N/A	N/A	N/A	N/A

Mean Feed Consumption, g/bird/day			
Treatment, mg/kg bw	Combined Sexes		
	Days 0-3	Days 3-7	Days 7-14
Control	19.1	13.8	17.1
200	16.8	11.7	16.4
400	23.0	12.0	15.9
800	15.3	12.3	17.5
1600	11.2	11.8	23.7
3200	N/A	N/A	N/A
NOEL	400 mg/kg bw		
EC_{50}	1600 mg/kg bw		

Reference chemical	effect NOEL LD_{50}	N/A	N/A	N/A

C. REPORTED STATISTICS:

Bartlett's test were employed to evaluate the homogenity of the variances. ANOVA and one-tailed Bonferroni's tests were used to compare the treatment groups to the control. The LD_{50} was calculated using a computer program using either moving average, binomial probability or probit. All other statistical analyses were conducted using [R]Satgraphics plus for Windows.

D. VERIFICATION OF STATISTICAL RESULTS:

Statistical Method: The NOEL and LOEL were determined visually and were based on sublethal effects at the 800 mg/kg bw level. The LD_{50} was estimated using the binomial method via TOXANAL statistical software.

LD_{50}: 1600 mg/kg bw 95% C.I.: 800-3200 mg/kg bw
NOEL: 400 mg/kg bw Probit Slope: N/A
LOEL: 800 mg/kg bw
Endpoint(s) Affected: Mortality and clinical signs of toxicity

E. STUDY DEFICIENCIES:

None of the deviations from US EPA guidelines impacted the classification of the study.

F. REVIEWER'S COMMENTS:

The reviewer's conclusions were identical to the study author. The 14-day acute oral LD_{50} is 1600 mg/kg bw, which categorizes Hallcomid M-8-10 Technical as slightly toxic to Northern Bobwhite quail on an acute oral basis.

G. CONCLUSIONS:

This toxicity study is scientifically sound and fulfills the guideline requirements for an acute toxicity study using the Northern Bobwhite quail (§71-1). This study is classified as Core.

LD_{50}: 1600 mg/kg bw 95% C.I.: 800-3200 mg/kg bw
NOEL: 400 mg/kg bw Probit Slope: N/A

LOEL: 800 mg/kg bw
TEC: 570 mg/kg bw
Endpoint(s) Affected: Mortality and clinical signs of toxicity

III. REFERENCES:

Stephan, C.E. 1982. U.S.E.P.A., Environmental Research Laboratory, Duluth, MN. Personal Communication to Dr. Lowell Bahner, Chairman, ASTM Task Group on Calculating LC50.

Stephan, C.E. 1977. Methods for Calculating and LC50. In: Aquatic Toxicology and Hazard Evaluation, ASTM STP 634. F.L. Mayer and J.L. Hamelink, eds. American Society for Testing Materials, Philadelphia, PA. 65-84.

[R]Satgraphics plus for Windows User Manual 1.0, Manugistics, Inc, Rockville, MD 20852, USA, 1994.

APPENDIX I. OUTPUT OF REVIEWER'S STATISTICAL VERIFICATION:

	EXPOSED	DEAD	DEAD	PROB.(PERCENT)
3200	10	10	100	9.765625E-02
1600	10	5	50	62.30469
800	10	0	0	9.765625E-02
400	10	0	0	9.765625E-02
200	10	0	0	9.765625E-02

THE BINOMIAL TEST SHOWS THAT 800 AND 3200 CAN BE USED AS STATISTICALLY SOUND CONSERVATIVE 95 PERCENT CONFIDENCE LIMITS, BECAUSE THE ACTUAL CONFIDENCE LEVEL ASSOCIATED WITH THESE LIMITS IS GREATER THAN 95 PERCENT.

AN APPROXIMATE LC50 FOR THIS SET OF DATA IS 1600

WHEN THERE ARE LESS THAN TWO CONCENTRATIONS AT WHICH THE PERCENT DEAD IS BETWEEN 0 AND 100, NEITHER THE MOVING AVERAGE NOR THE PROBIT METHOD CAN GIVE ANY STATISTICALLY SOUND RESULTS.