VOLUME____OF____OF SUBMISSION

ABAMECTIN EC (A8612AB): FINAL REPORT

TITLE

Abamectin EC (A8612AB): Acute Oral Toxicity Study in Rats

DATA REQUIREMENT

EPA Guideline Number OPPTS 870.1100

AUTHOR

Janice O. Kuhn, Ph.D., D.A.B.T.

COMPLETION DATE

February 17, 2004

PERFORMING LABORATORY

STILLMEADOW, Inc.
12852 Park One Drive
Sugar Land, TX 77478

LABORATORY STUDY IDENTIFICATION

STILLMEADOW Number 7973-03
Syngenta Number 3337-03

SUBMITTER/SPONSOR

Syngenta Crop Protection, Inc.
410 Swing Road
Post Office Box 18300
Greensboro, NC 27419

VOLUME 1 OF 1 OF STUDY

STATEMENTS OF DATA CONFIDENTIALITY CLAIMS

1) *The following statement applies to submissions to regulatory agencies in the United States of America.*

STATEMENT OF NO DATA CONFIDENTIALITY CLAIM

No claim of confidentiality is made for any information contained in this study on the basis of its falling within the scope of FIFRA Section 10 (d) (1) (A), (B), or (C).

Company: Syngenta Crop Protection, Inc.

Company Representative: Carolyn Brinkley

Title: Senior Regulatory Product Manager

Signature: Carolyn Brinkley Date: 4/14/04

These data are the property of Syngenta Crop Protection, Inc. and, as such, are considered to be confidential for all purposes other than compliance with the regulations implementing FIFRA Section 10.

Submission of these data in compliance with FIFRA does not constitute a waiver of any right to confidentiality, which may exist under any other provision of common law or statute or in any other country.

2) *The following statement applies to submissions to regulatory agencies other than in the United States of America.*

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It should not be disclosed in any form to an outside party, nor should information contained herein be used by a registration authority to support registration of this product or any other product without the written permission of Syngenta Limited.

GOOD LABORATORY PRACTICE COMPLIANCE STATEMENT

This study was designed and performed in STILLMEADOW, Inc.'s laboratory and was conducted in compliance with:

- United States Environmental Protection Agency FIFRA: Good Laboratory Practice Standards, 40 CFR 160
- United States Environmental Protection Agency TSCA 40 CFR 792
- Organization for Economic Cooperation and Development's Principles of Good Laboratory Practice, Annex 2, C(98)17
- Japan Ministry of Agriculture, Forestry and Fisheries, Notification 11-Nousan-6283, Director-General of Agricultural Production Bureau

Janice O. Kuhn, Ph.D., DABT
Study Director, STILLMEADOW, Inc.

17 Feb 04
Date

Merrill Tisdel
Representative of Submitter/Sponsor

MARCH 22 2004
Date

Submitter/Sponsor: Syngenta Crop Protection, Inc.
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QUALITY ASSURANCE STATEMENT

Test Substance: Abamectin EC
Study Title: Acute Oral Toxicity Study in Rats

The study report and data have been audited in accordance with STILLMEADOW, Inc. Standard Operating Procedures (SOPs). The final report accurately reflects the study data. The findings from the inspection and audit were reported to the Study Director and Management as follows:

Study Phase Inspected	Inspection Type	Date Inspected	Reported to Study Director	Reported to Management
Protocol Review	Study-based	9 Dec 03	9 Dec 03	9 Dec 03
Body weights/necropsy	Study-based	16 Dec 03 24 Dec 03	17 Dec 03 24 Dec 03	17 Dec 03 24 Dec 03
Report/Data Audit	Study-based	2 Feb 04	2 Feb 04	2 Feb 04

B. Lynn Murphy, B.S., RQAP
Quality Assurance Director, STILLMEADOW, Inc.

17 Feb 04
Date

SUMMARY

The test substance, Abamectin EC (A8612AB, FL-031782), was evaluated for its acute oral toxicity potential in albino rats when administered as a gavage dose at a level of 5000 mg/kg. Since the test substance failed the limit test, the main test was conducted following the up-and-down procedure (UDP) at 175, 550, 1750 and 5000 mg/kg. The study was terminated following the stopping rules of this procedure. No mortality occurred at the 175 mg/kg level. The only clinical sign in survivors was activity decrease, which was no longer evident by Day 3. There was no effect on body weight gain in survivors. Abnormal necropsy findings occurred only in the animals dying on test, and pertained to fur, lungs, liver and contents of the gastrointestinal tract. The acute oral LD_{50} was estimated to be 891 mg/kg.

INTRODUCTION

The objective of this study was to assess the acute oral toxicity potential of the test substance when administered by gavage to rats in accordance with US EPA OPPTS 870.1100, which is intended to meet testing requirements of FIFRA 7 USC 136, *et seq*, and TSCA 15 USC 2601. This study was conducted for Syngenta Crop Protection, Inc., according to the approved protocol and STILLMEADOW, Inc. SOPs. There were no deviations from the protocol which affected the quality or outcome of the study. All procedures used in this study are in compliance with Animal Welfare Act Regulations. In the opinion of the sponsor, the study did not unnecessarily duplicate any previous work. The protocol, raw data, this report and a sample of test substance are archived at STILLMEADOW, Inc. The study was initiated on 10 Dec 03, the pre-dose experimental portion began on 9 Dec 03, and the animals were treated as follows:

Dose (mg/kg)	Treatment		Animal Number	In-life Termination Date
	Date	Time		
5000	10 Dec 03	1207	103	11 Dec 03
5000	10 Dec 03	1210	102	11 Dec 03
5000	10 Dec 03	1211	101	11 Dec 03
175	12 Dec 03	0855	104	26 Dec 03
550	13 Dec 03	1005	105	27 Dec 03
1750	14 Dec 03	0825	106	28 Dec 03
1750	15 Dec 03	1014	107	16 Dec 03
550	16 Dec 03	1015	108	21 Dec 03
1750	17 Dec 03	1042	109	18 Dec 03
550	19 Dec 03	1153	110	2 Jan 04
1750	23 Dec 03	0920	111	24 Dec 03

TEST SUBSTANCE

Identification:	Abamectin EC FL-031782 A8612AB
Date & Quantity Received:	31 Oct 03; 0.12 L
Physical Description:	Pale yellow liquid
Storage:	Room temperature
Density:	0.9782 g/mL
Purity:	2.00% active ingredient
Stability:	Reassay: Oct 04

Records pertaining to stability, characterization, identity, synthesis methods and location of documentation are the responsibility of the sponsor. A copy of the sponsor's Analytical Report is retained in the study file.

TEST SYSTEM

Experimental Animals

Species & Strain:	Albino rat; Sprague-Dawley
Justification of Species:	The rat is a representative rodent species preferred by various regulatory agencies for use in an acute oral study.
Source:	Texas Animal Specialties, Humble, TX
Date Received:	20 Nov & 11 Dec 03
Quarantine Period:	5 days
Quantity & Sex:	11 females (nulliparous and non-pregnant) were selected for testing
Group/Animal ID:	Cage cards/Ear punch
Fasted Wt on Dosing Day:	150-234 g
Date of Birth:	29 Sep & 20 Oct 03

Animal Husbandry

Cage Type:	Suspended, wire bottom, stainless steel
Housing:	1 per cage
Environmental Controls Set to Maintain:	·Temperature Range 22°C±3° ·Humidity Range 30-70% ·12-hour light/dark cycle ·10-12 air changes/hour
Food:	PMI Feeds Inc.™ Formulab #5008; available *ad libitum* except for approximately 16 hours before dosing
Water:	Municipal water supply analyzed by TCEQ Water Utilities Division; available *ad libitum* from automatic water system.

Animal husbandry and housing at STILLMEADOW, Inc. comply with Animal Welfare Act Regulations. No contaminants were expected to have been present in the feed or water which would have interfered with or affected the results of the study.

PROCEDURES

Test Substance Administration

The test substance was administered as received and was not diluted. An individual dose was calculated for each animal based on its fasted body weight and administered by gavage at a volume ranging from 0.18 mL/kg at the 175 mg/kg level to 5.11 mL/kg at the 5000 mg/kg level. Each dose was administered using an appropriately sized syringe and stainless steel ball-tipped intubation needle. The animals were returned to their cages immediately after dosing.

In-life Observations

Observations for mortality and clinical/behavioral signs of toxicity were made at least three times on the day of dosing (Day 0) and at least once daily thereafter for 14 days. Individual body weights were recorded just prior to dosing and on Days 7 and 14, or at the time of discovery after death.

Postmortem Observations

On Day 14 after dosing, each surviving animal was euthanized by an overdose of CO_2. All study animals were subjected to gross necropsy and all abnormalities were recorded.

Statistical Analysis

The LD_{50} value with 95% confidence interval was calculated using the AOT425 Stat Program supplied by the EPA.

RESULTS AND DISCUSSION

Mortality/Estimated Lethality Values

Individual mortality data, including time of death, are presented in Table 1. A summary of the mortality/survival incidence is presented below.

Main Test Sequence	Animal Number	Dose (mg/kg)	Results	Main Test Sequence	Animal Number	Dose (mg/kg)	Results
1	104	175	O	6	108	550	X
2	105	550	O	7	109	1750	X
3	106	1750	O	8	110	550	O
4	102	5000	X	9	111	1750	X
5	107	1750	X				

X = died; O = survived; Note: Animals 101 and 103 dosed for limit test (5000 mg/kg) died.

The acute oral LD_{50} for female rats was estimated to be 891 mg/kg, with 95% confidence interval of 0 - 9320 mg/kg.

RESULTS AND DISCUSSION (cont.)

Body Weights

Individual body weights are presented in Table 1. Body weight gain in surviving animals was unaffected by the administration of the test substance.

Clinical Signs

Clinical signs are presented in Table 2. The only clinical sign in surviving animals was activity decrease, which was no longer evident by Day 3. Body tremors, diarrhea, piloerection, slow/labored breathing and stained muzzle were observed only in animals that died on test.

Necropsy Findings

Individual necropsy findings are presented in Table 1. The gross necropsy on animals that died on test revealed stained fur; discolored lungs, liver and contents of the stomach/large intestine; gas in the stomach and empty gastrointestinal tract. The gross necropsy on animals surviving to termination of the study revealed no observable abnormalities.

CONCLUSION

The test substance, Abamectin EC (A8612AB), was evaluated for its acute oral toxicity potential when administered to albino rats. The acute oral LD_{50}, as indicated by the data, is estimated to be 891 mg/kg in females.

Study Director: Janice O Kuhn 17 Feb 04

Janice O. Kuhn, Ph.D., DABT — Date
Sr. Toxicologist, STILLMEADOW, Inc.

STUDY PERSONNEL

Technical Staff: Carol Morris, B.A.
Hector Fuentes
Theresa Balch, B.S.
Paul Siemens, B.A.
Robert Preston

Data Services: Connie Pavatte, Report Preparation

TABLE 1
ACUTE ORAL TOXICITY STUDY: UP & DOWN PROCEDURE (UDP) IN RATS
Body Weights, Time of Death, and Gross Necropsy
Test Substance: Abamectin EC

Dose Level: 175 mg/kg (0.18 mL/kg)

Animal Number	Date of Dosing	Body Weights (g)			Time of Death*	Gross Necropsy Findings
		Day 0	Day 7	Final		
104-F	12 Dec 03	166	190	207	Day 14	NOA

Dose Level: 550 mg/kg (0.57 mL/kg)

Animal Number	Date of Dosing	Body Weights (g)			Time of Death*	Gross Necropsy Findings
		Day 0	Day 7	Final		
105-F	13 Dec 03	196	236	253	Day 14	NOA
108-F	16 Dec 03	234	---	203	Day 5	Face & body stained red; liver black; stomach & intestines empty.
110-F	19 Dec 03	150	192	213	Day 14	NOA

Dose Level: 1750 mg/kg (1.77 mL/kg)

Animal Number	Date of Dosing	Body Weights (g)			Time of Death*	Gross Necropsy Findings
		Day 0	Day 7	Final		
106-F	14 Dec 03	168	183	204	Day 14	NOA
107-F	15 Dec 03	151	---	146	Day 1	Lungs red; stomach full of gas & yellow liquid; intestines empty.
109-F	17 Dec 03	214	---	208	Day 1	Urogenital area yellow; lungs bright red; stomach full of yellow liquid; sm intestine empty; lg intestine full of green paste.
111-F	23 Dec 03	173	---	169	Day 1	Lungs red; liver mottled; stomach full of gas & yellow liquid; intestines empty.

Dose Level: 5000 mg/kg (5.11 mL/kg)

Animal Number	Date of Dosing	Body Weights (g)			Time of Death*	Gross Necropsy Findings
		Day 0	Day 7	Final		
101-F	10 Dec 03	186	---	182	Day 1	Lungs dark red; brown liquid in stomach.
102-F	10 Dec 03	203	---	201	Day 1	Lungs mottled pink; stomach full of yellow liquid.
103-F	10 Dec 03	180	---	175	Day 1	Lungs dark red; liver pale; brown liquid in stomach.

* - Indicates time of discovery after death (Day of dosing is Day 0; Hr is after Day 0 dosing; Day 14 is terminal sacrifice). If discovery was between scheduled observations, the time of death was recorded under the next scheduled observation.
F - Female; NOA - No Observable Abnormalities

TABLE 2
ACUTE ORAL TOXICITY STUDY: UP & DOWN PROCEDURE (UDP) IN RATS
Pharmacologic and/or Toxicologic Signs
Test Substance: Abamectin EC

		Time After Treatment																
		DAY 0			DAYS													
Animal No.	Reaction and Severity	1st	2nd	3rd	1	2	3	4	5	6	7	8	9	10	11	12	13	14
Dose Level: 175 mg/kg (0.18 mL/kg)																		
104-F	Activity decrease	-	-	m	m	m	-	-	-	-	-	-	-	-	-	-	-	-
Dose Level: 550 mg/kg (0.57 mL/kg)																		
105-F	Animal appeared normal for the duration of the study.																	
108-F	Activity decrease	-	-	-	-	m	m	e										
	Body tremors	-	-	-	-	p	s	v										
	Diarrhea	-	-	-	-	m	-	-										
	Muzzle stained red	-	-	-	-	p	p	-										
	Piloerection	-	-	-	-	-	-	s										
	Death	-	-	-	-	-	-	-	D									
110-F	Activity decrease	m	m	m	e	-	-	-	-	-	-	-	-	-	-	-	-	-
Dose Level: 1750 mg/kg (1.77 mL/kg)																		
106-F	Animal appeared normal for the duration of the study.																	
107-F	Activity decrease	m	e	e														
	Piloerection	-	s	s														
	Death	-	-	-	D													
109-F	Activity decrease	-	-	e														
	Death	-	-	-	D													
111-F	Death	-	-	-	D													
Dose Level: 5000 mg/kg (5.11 mL/kg)																		
101-F	Activity decrease	e	e	e														
	Labored breathing	p	-	-														
	Piloerection	-	m	m														
	Slow breathing	-	e	-														
	Death	-	-	-	D													
102-F	Activity decrease	e	e	e														
	Piloerection	-	m	m														
	Death	-	-	-	D													
103-F	Activity decrease	e	e	e														
	Piloerection	-	m	m														
	Slow breathing	-	e	-														
	Death	-	-	-	D													

F = Female; v = very slight; s = slight; m = moderate; e = extreme; p = present; - = observation not present; D = death
Note: Digits indicate number of animals exhibiting reaction. Time of death indicates time of discovery after death. If discovery was between scheduled observations, death is presented under next observation time.

TABLE 3
LD_{50} Analysis*
ACUTE ORAL TOXICITY STUDY: UP & DOWN PROCEDURE (UDP) IN RATS
Test Substance: Abamectin EC

Test Type: Main Test
Limit Dose: 5000 mg/kg
Assumed LD_{50}: Default
Assumed sigma: 0.5 mg/kg

Recommended dose progression (mg/kg): 5000, 1750, 550 and 175

Test Sequence	Animal Number	Dose (mg/kg)	Short Term Results**	Long Term Results
1	104	175	O	O
2	105	550	O	O
3	106	1750	O	O
4	102	5000	X	X
5	107	1750	X	X
6	108	550	O	X
7	109	1750	X	X
8	110	550	O	O
9	111	1750	X	X

X = died; O = survived

Dose Recommendation: Main Test Complete

Stopping Criteria Met: LR criterion

Summary of Results

Dose	O	X	Total
175	1	0	1
550	2	1	3
1750	1	3	4
5000	0	1	1
All Doses:	4	5	9

Estimated LD_{50} = 891 mg/kg with 95% confidence interval of 0 – 9320 mg/kg.

* - AOT425statpgm (Version 1.0) Test Results and Recommendations
Acute Oral Toxicity (OECD Test Guideline 425) Statistical Program
** - At ~48 hrs after dosing that animal or when next dose is selected.