SYN545192

SYN545192 EC (A15457H) -
Acute Oral Toxicity Study in the Rat (Up and Down Procedure)

Final Report

DATA REQUIREMENT(S): OECD Test Guideline 425 (2008)
EPA OPPTS 870.1100 (2002)

AUTHOR(S): Istvánné Kiss, M.Sc.

STUDY COMPLETION DATE: 05 October 2012

PERFORMING LABORATORY: CiToxLAB Hungary Ltd.
H-8200 Veszprém, Szabadságpuszta, Hungary

LABORATORY PROJECT ID: Report Number: 12/063-001PR
Study Number: 12/063-001PR
Task Number: TK0065141

SPONSOR(S): Syngenta Limited
Jealott's Hill International Research Centre
Bracknell, Berkshire, RG42 6EY, United Kingdom

STATEMENT OF DATA CONFIDENTIALITY CLAIMS

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GOOD LABORATORY PRACTICE COMPLIANCE STATEMENT

This study has been performed in accordance with the Principles of Good Laboratory Practice (Hungarian GLP Regulations: 9/2001. (III. 30.) EüM-FVM joint decree of the Minister of Health and the Minister of Agriculture and Regional Development which corresponds to the OECD GLP, ENV/MC/CHEM (98) 17.).

This study was conducted in accordance with a written Study Plan, authorized by the Sponsor and CiToxLAB Hungary Ltd. management, and followed applicable Standard Operating Procedures.

I, the undersigned, declare that this report constitutes a true record of the actions undertaken and the results obtained in the course of this study.

Signature: K. Istvánné Date: 05 October 2012

Istvánné Kiss, M.Sc.
Study Director

Performing Laboratory: CiToxLAB Hungary Ltd.
H-8200 Veszprém, Szabadságpuszta,
Hungary

FLAGGING STATEMENT

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QUALITY ASSURANCE STATEMENT

Study Number: 12/063-001PR

Study Title: SYN545192 EC (A15457H) – Acute Oral Toxicity Study in the Rat (Up and Down Procedure)

Test Item: SYN545192 EC (A15457H)

This study has been inspected, and this report audited by the Quality Assurance Unit in compliance with the Principles of Good Laboratory Practice. As far as it can be reasonably established the methods described and the results incorporated in this report accurately reflect the raw data produced during this study.

All inspections, data reviews and the report audit were reported in written form to the study director and to management. The dates of such inspections and of the report audit are given below:

Date of Inspection	Phase(s) Inspected/Audited	Date of report to	
		Management	Study Director
12 April 2012	Study Plan	12 April 2012	12 April 2012
17 April 2012	Tretment	17 April 2012	17 April 2012
02 August 2012	Draft Report	02 August 2012	02 August 2012
05 October 2012	Final Report	05 October 2012	05 October 2012

Signature: _____ Date: 05 Oct 2012

Ágnes Rédl, M.Sc.
QA Inspector

MANAGEMENT STATEMENT

According to the conditions of the research and development agreement between Syngenta Limited (as Sponsor) and CiToxLAB Hungary Ltd. (as Test Facility) the study titled "SYN545192 EC (A15457H)- Acute Oral Toxicity Study in the Rat (Up and Down Procedure)" has been performed in compliance with the Principles of Good Laboratory Practice.

Signature: _____ Date: 05 October 2012

David J. Esdaile, M.Sc.
Scientific Director

GENERAL INFORMATION

Contributors

The following contributed to this report in the capacities indicated:

Name	Function
Istvánné Kiss, M.Sc.	Study Director
Viktória Zelenák, M.Sc.	Assistant Scientist
István Pásztor, DVM	Veterinary Control
Peter Maslej, DVM, PhD	Head of Pathology Unit
Tamás Mészáros, PhD	Head of Central Dispensary
Claire Elliott	Syngenta Study Manager

Study dates

Experimental Starting Date	17 April 2012
Experimental Completion Date	09 July 2012
Receipt of Animals	12 April and 05 June 2012
Acclimatisation	At least 5 days

Treatment	17 April 2012 (female no. 2453) 19 April 2012 (female no. 2454) 25 April 2012 (female no. 2456) 27 April 2012 (female no. 2457) 30 April 2012 (female no. 2458) 20 June 2012 (female no. 3829) 22 June 2012 (female no. 3830) 25 June 2012 (female no. 3831) 27 June 2012 (female no. 3832)
Observation	17 April – 01 May 2012 (female no. 2453) 19 April – 03 May 2012 (female no. 2454) 25 April – 09 May 2012 (female no. 2456) 27 April – 11 May 2012 (female no. 2457) 30 April 2012 (female no. 2458) 20 June – 04 July 2012 (female no. 3829) 22 June 2012 (female no. 3830) 25 June – 09 July 2012 (female no. 3831) 27 June 2012 (female no. 3832)

Deviations from the Study Plan

The reception dates of the animals were 12 April and 05 June 2012 instead of 12 April 2012 as it was indicated in the study plan.

The in-life phase was ended later than it was indicated in the study plan.

The draft report is issued later than it was indicated in the study plan.

Animal no. 2456 was treated with a dose volume of 10 mL/kg/bw instead of 0.5 mL/kg/bw as it was indicated in the study plan.

These deviations considered to have no impact on the outcome of the study and interpretation of the results.

Deviations from the Guideline

A single mortality (animal no. 2455) was observed at 550 mg/kg bw which was considered to be due to gastric reflux and not due to toxicity of the test item based on the macroscopic observations, so this animal is excluded from the calculations. Brown liquid in the digestive content of the stomach and oesophagus (cervical and thoracic regions of oesophagus) with obstruction of the oesophagus by firm brown material at the level of thyroids, and non-collapsed lungs were recorded macroscopically in this found dead rat.

Performing laboratory test substance reference number

12003A

Other

The study documents:
- study plan,
- all raw data,
- sample of the test item,
- original study report and any amendments,
- correspondence

will be archived according to the Hungarian GLP regulations and to applicable SOP's in the archives of CiToxLAB Hungary Ltd. 8200 Veszprém, Szabadságpuszta, Hungary. This is for a period of 15 years.

After the retention time of 15 years has elapsed all the archived materials listed above will be returned to the Sponsor or retained for a further period if agreed by a contract. Otherwise the materials will be discarded.

TABLE OF CONTENTS

1.0 EXECUTIVE SUMMARY

1.1 Study Design

An acute oral toxicity (up and down procedure) study was conducted with 9 animals (female CRL:WI rat). The starting dose was 550 mg/kg bw. Animals were treated with a single oral (gavage) dose of SYN545192 EC (A15457H) at a dose level of 175, 550 or 2000 mg/kg bw followed by a 14 day observation period. The animals were fasted overnight prior to treatment and food was returned approximately 3 hours after dosing.

Surviving animals were observed individually after dosing at 30 minutes, 1, 2, 3, 4 and 6 hours post treatment and once each day for 14 days thereafter. Body weight was measured on Day -1 (prior to removal of food), Day 0 (prior to administration) and weekly thereafter. All surviving animals were examined macroscopically at the end of the study. Moreover, the animals found dead were examined macroscopically and body weight was recorded at necropsy.

1.2 Results

2000 mg/kg body weight (four animals)

Mortality was observed in all animals at 2000 mg/kg bw (4/4).

Treatment with SYN545192 EC (A15457H) at the dose level of 2000 mg/kg bw caused decreased activity, hunched back and incoordination in three animals. In addition, clonic convulsion, piloerection, lethargy, prone position, increased respiratory rate and dyspnoea were noted in one or two animals.

Dark/red discoloration and/or non-collapsing lungs was found in 4/4 found dead females at necropsy. Dark/red discoloration was also seen in the thymus and stomach glandular mucosa.

Brown liquid was observed in the stomach of animal 3830 which was found dead. This was considered to be associated with the administrated test item.

550 mg/kg body weight (four animals)

Clinical signs of toxicity were observed in two of the four animals. These included decreased activity, hunched back, piloerection, prone position and incoordination.

There were no macroscopic findings noted in animals dosed at 550 mg/kg bw.

175 mg/kg body weight (one animal)

No adverse clinical signs were observed following treatment at 175 mg/kg bw.

There were no macroscopic findings noted in animal dosed at 175 mg/kg bw.

Body weight measurements of the surviving animals during the study showed no indication of a treatment-related effect.

1.3 Conclusion

Under the conditions of this study, the acute oral median lethal dose (LD_{50}) of SYN545192 EC (A15457H) was calculated to be 1086 mg/kg bw in female CRL:(WI) rats.

Approximate 95% confidence interval is 550 to 2000 mg/kg body weight.

2.0 INTRODUCTION

2.1 Purpose

The purpose of the study was to assess the oral toxicity of the test item SYN545192 EC (A15457H) when administered as a single oral gavage dose to female rats at one or more defined dose levels.

This study was performed with vertebrate animals as no in vitro alternative is available. The study has been designed such that the minimum number of animals were used.

2.2 Guidelines

The study was performed according to the following guidelines:

- OECD Guideline Reference 425 (2008): Acute Oral Toxicity - Up-and-Down Procedure.
- United States Environmental Protection Agency, Health Effects Test Guidelines, OPPTS 870.1100 Acute Oral Toxicity EPA 712-C-02-190, December 2002.

3.0 MATERIALS AND METHODS

3.1 Test Substance

Data as supplied by the Sponsor.

Name:	SYN545192 EC (A15457H)
Batch number:	SMU1LP001
Product code:	A15457H
Purity:	Content of benzovindiflupyr (SYN545192) – 10.1 % w/w corresponding to 98.6 g/l Content of water – 0.74 % w/w
Density:	976 kg/m^3
Appearance:	brown liquid
Recertification date:	End of July 2014
Storage conditions:	Room temperature (<30 ^{o}C)
Safety Precautions:	Routine safety precautions (lab coat, gloves, goggles, face mask) for unknown materials will be applied to assure personnel health and safety.

The Certificate of Analysis is presented in Appendix 2.

3.1.1 Identification, receipt

The test item of a suitable chemical purity together with all precautions required in the handling and disposal of the test item were supplied by the Sponsor. The identification of the test item was made in the Central Dispensary Unit of CiToxLAB Hungary Ltd. on the basis of the information provided by Sponsor.

3.1.2 Formulation

The test item was administered undiluted or in the case of one animal at the dose level 175 mg/kg bw, the test item was diluted in 0.5 % CMC (Exp. date: 21 May 2012) with the concentration of 17.5 mg/ml.

3.2 Experimental Animals

Species and strain:	CRL:(WI) rats
Source:	Charles River Laboratories, Research Models and Services, Germany GmbH, Sandhofer Weg 7, D-97633 Sulzfeld
Hygienic level at arrival:	SPF
Hygienic level during the study:	Standard housing conditions
Justification of strain:	Recognized by international guidelines as a recommended test system.
Number of animals:	9
Sex:	female
Age when treated:	Young adult rats, 8 -11 weeks old.
Body weight (at dosing):	203-230 g
Randomization:	Selected by hand at time of delivery.
Acclimatization time:	at least 5 days

3.2.1 Husbandry

Animal health:	Only healthy animals were used for the test. The health status was certified by the veterinarian.
Housing:	Individual caging
Cage type:	Type II. polypropylene/polycarbonate
Bedding:	Lignocel Bedding for Laboratory Animals was available to animals during the study.
Light:	12 hours daily, from 6.00 a.m. to 6.00 p.m.
Temperature:	22 ± 3 °C
Relative humidity:	30 - 70%
Ventilation:	15-20 air exchanges/hour
Enrichment:	Rodents were housed with deep wood sawdust bedding to allow digging and other normal rodent activities.

The temperature and relative humidity were recorded twice daily during the acclimatisation period and throughout the study.

3.2.2 Food and water supply

Animals received ssniff® SM R/M-Z+H "Autoclavable complete diet for rats and mice – breeding and maintenance" produced by ssniff Spezialdiäten GmbH, D-59494 Soest Germany *ad libitum* (Lot number: 719 6627 and 601 7197). Tap water from the municipal supply was supplied from 500 mL bottles *ad libitum*. The food was considered not to contain any contaminants that could reasonably be expected to affect the purpose or integrity of the study and the water was considered to be fit for human consumption. Details of the diet are archived with the raw data at CiToxLAB Hungary Ltd.

Water quality control analysis is performed once every three months and microbiological assessment is performed monthly by Veszprém County Institute of State Public Health and Medical Officer Service (ÁNTSZ, H-8201 Veszprém, József A. street 36., Hungary). The quality control results are retained in the archives at CiToxLAB Hungary Ltd.

3.2.3 Identification

Animals were individually identified by numbers written on the tail with a permanent marker pen. The numbers were given on the basis of CiToxLAB Hungary Ltd.'s master file, for each animal allocated to the study. The boxes were identified by cards holding information on the study code, the sex of animals, the dose, the cage number and the individual animal number.

3.3 Administration of the Test Item

3.3.1 Dosages

Justification of the doses:

An acute oral toxicity (up and down procedure) study was conducted with 9 animals. The starting dose was 550 mg/kg bw. Animals were treated with a single oral (gavage) dose of SYN545192 EC (A15457H) at a dose level of 175, 550 or 2000 mg/kg bw. The dose volume used at these concentrations is shown below.

Animal Number	Dosage [mg/kg body weight]	Volume Dosed [mL]	Viability/Mortality
2453	550	0.12	Survived
2454	2000	0.42	Died
2455	550	0.13	Died*
2456	175	2.3**	Survived
2457	550	0.12	Survived
2458	2000	0.46	Died
3829	550	0.12	Survived
3830	2000	0.44	Died
3831	550	0.12	Survived
3832	2000	0.42	Died

*This found dead animal was excluded from the study as the death was due to gastric reflux as a consequence of an intubation error.

** This animal was treated with a dose volume of 10 mL/kg/bw instead of 0.5 mL/kg/bw.

Oral administration was considered to be an appropriate dose route as it is a possible route of human exposure.

Oral administration was considered to be an appropriate dose route as it is a possible route of human exposure.

3.3.2 Procedure

A single oral (gavage) dose was followed by a 14 day observation period. The night before treatment the animals were fasted. The food, but not water, was withheld overnight. Animals were weighed before dosing and the food was returned 3 hours after the treatment.

Single animals were dosed sequentially following an interval of at least approximately 48 hours. The time intervals between dosing were determined by the onset, duration and severity of toxic signs.

3.4 Observations

3.4.1 Clinical observations

Surviving animals were observed individually after dosing at 30 minutes, then at approximately 1, 2, 3, 4, and 6 hours after dosing and once each day for 14 days thereafter depending on the time of death. Individual observations were performed on the skin and fur, eyes and mucous membranes and also respiratory, circulatory, autonomic and central nervous system, somatomotor activity and behaviour pattern were assessed.

Particular attention was directed to observation of tremors, convulsions, salivation, diarrhoea, lethargy, sleep and coma.

3.4.2 Body weight measurement

The body weights of the surviving animals were recorded on Day -1 and Days 0 (beginning of the experiment) 7 and 14. The body weight of animals found dead were recorded at necropsy.

3.5 Necropsy

All animals were euthanised at the end of the observation period by exsanguination under pentobarbital anaesthesia (Euthasol® 40%, Lot No.: 11H15 8, Expiry Date: July 2014, Produced by: Produlab Pharma B.V.)

A macroscopic examination was performed including external appearance. The cranial, thoracic and abdominal cavities were opened and the appearance of the tissues and organs were observed. All gross pathological changes were recorded for each animal on the post mortem record sheets and the animals were discarded.

3.6 Data Evaluation

Type, severity and duration of clinical observations are described. Body weight and body weight changes are summarised in tabular form. Necropsy findings are described and summarised in tabular form.

4.0 RESULTS AND DISCUSSION

Individual clinical observations and mortality results are presented in Table 1. Individual body weights and necropsy results are presented in Tables 2 and 3, respectively. The Pathology report is presented in Appendix 1.

4.1 Mortality

Mortality was observed in all animals receiving a single dose of SYN545192 EC (A15457H) at 2000 mg/kg bw (4/4).

One animal died following dosing at 550 mg/kg bw. However, the cause of death was attributed to gastric reflux as a consequence of an intubation error and not due to the toxicity of the test item. Brown liquid in the digestive content of the stomach and oesophagus (cervical and thoracic regions of oesophagus) with obstruction of the oesophagus by firm brown material at the level of thyroids, and non-collapsed lungs were recorded macroscopically in this found dead rat.Therefore this animal was excluded from the study and was not included in calculation or the acute oral median lethal dose and is not included in the tables of observations. No mortality was observed in a further 4 animals that were dosed at 550 mg/kg bw (0/4) nor in the single animal dosed at 175 mg/kg.

4.2 Clinical Signs

Treatment with SYN545192 EC (A15457H) at the dose level of 2000 mg/kg bw caused decreased activity, hunched back and incoordination in three animals. In addition, clonic convulsion, piloerection, lethargy, prone position, increased respiratory rate and dyspnoea were noted in one or two animals.

Clinical signs of toxicity were observed in two animals of the four animals treated with 550 mg/kg bw. These included decreased activity, hunched back, piloerection, prone position and incoordination.

No adverse clinical signs were observed following treatment at 175 mg/kg bw.

4.3 Body Weights

Body weight and body weight changes of the surviving animals during the study showed no indication of a treatment-related effect.

4.4 Macroscopic Findings

A single oral gavage of SYN545192 EC (A15457H) to the CRL: (WI) female rat led to the death of four animals at a dose level of 2000 mg/kg bw. A specific cause of death was not determined for these animals. Dark/red discoloration and/or non-collapsing lungs was found in 4/4 found dead females at necropsy. Dark/red discoloration was also seen in the thymus and stomach glandular mucosa.

In one female, brown liquid was observed in the stomach which was considered to be associated with the administrated test item.

No gross observations were seen in any animals dosed at 175 or 550 mg/kg bw of SYN545192 EC (A15457H), subjected to the necropsy on Day 14.

5.0 CONCLUSIONS

Under the conditions of this study, the acute oral median lethal dose (LD_{50}) of SYN545192 EC (A15457H) was calculated to be 1086 mg/kg bw in female CRL:(WI) rats.

Approximate 95% confidence interval is 550 to 2000 mg/kg body weight.

TABLES SECTION

TABLE 1 Individual Findings – Clinical Signs

DOSE LEVEL: 175 mg/kg bw, Treatment on Day 0 **SEX: FEMALE**

Cage No.	Animal Number	Observations	Observation days									Frequency
			0						1	2	3-14	
			30'	1h	2h	3h	4h	6h				
4	2456	Symptom Free	+	+	+	+	+	+	+	+	+	20/20

DOSE LEVEL: 550 mg/kg bw, Treatment on Day 0 **SEX: FEMALE**

Cage No.	Animal Number	Observations	Observation days									Frequency
			0						1	2	3-14	
			30'	1h	2h	3h	4h	6h				
1	2453	Symptom Free	+	+	+	+	+	+	+	+	+	20/20
5	2457	Symptom Free	-	-	-	-	-	+	+	+	+	15/20
		Activity decreased	1	1	1	1	-	-	-	-	-	5/20
		Hunched back	+	+	+	+	+	-	-	-	-	5/20
		Piloerection	-	+	+	+	+	-	-	-	-	4/20
7	3829	Symptom Free	+	+	+	+	+	+	+	+	+	20/20
9	3831	Symptom Free	-	-	-	-	-	-	+	+	+	14/20
		Activity decreased	2	1	1	1	1	1	-	-	-	6/20
		Hunched back	+	+	+	+	+	+	-	-	-	6/20
		Prone position	-	-	-	-	+	-	-	-	-	1/20
		Incoordination	1	1	1	1	1	1	-	-	-	6/20

Remarks: + = present - = absent

h = hour (s) ' = minute

Frequency of observation = number of occurence of observation / total number of observations

Severities: 1 = Slight/Small/Few; 2 = Moderate/Medium; 3 = Marked/Large/Many

TABLE 1 Individual Findings – Clinical Signs (Continued)

DOSE LEVEL: 2000 mg/kg bw, Treatment on Day 0 **SEX: FEMALE**

Cage No.	Animal Number	Observations	Observation days									Frequency
			0						1	2	3-14	
			30'	1h	2h	3h	4h	6h				
2	2454#	Activity decreased	-	2	2	2	2	2				5/6
		Hunched back	-	-	-	-	+	+				2/6
		Incoordination	2	2	2	2	2	2				6/6
		Clonic convulsion	2	-	-	-	-	-				1/6
		Piloerection	-	-	+	+	+	+				4/6
		Found Dead	-	-	-	-	-	-	+			-
6	2458#	Found Dead	+									-
8	3830#	Activity decreased	-	2	-							1/3
		Lethargy	-	-	+							1/3
		Hunched back	-	+	-							1/3
		Prone position	-	+	+							2/3
		Incoordination	1	-	-							1/3
		Respiratory rate increased	-	2	-							1/3
		Dyspnoea	-	-	+							1/3
		Found Dead	-	-	-	+						-
10	3832#	Activity decreased	1	1	2	2	2	3				6/6
		Hunched back	-	+	+	+	+	+				5/6
		Prone position	-	-	-	-	-	+				1/6
		Incoordination	2	2	2	2	2	-				5/6
		Clonic convulsion	-	-	-	-	-	3				1/6
		Piloerection	-	-	+	+	+	-				3/6
		Dyspnoea	-	-	-	-	-	+				1/6
		Found Dead	-	-	-	-	-	-	+			-

Remarks: + = present - = absent # = Found Dead

h = hour (s) ' = minute

Frequency of observation = number of occurence of observation / total number of observations

Severities: 1 = Slight/Small/Few; 2 = Moderate/Medium; 3 = Marked/Large/Many

TABLE 2 Body Weight and Body Weight Gain

DOSE LEVEL:175 mg/kg bw, Treatment on Day 0 **SEX: FEMALE**

Cage No.	Animal No.	Body weight (g) Days				Body Weight Gain (g)			
		-1	0	7	14	-1-0	0-7	7- 14	-1 - 14
4	2456	242	230	262	277	-12	32	15	35

DOSE LEVEL:550 mg/kg bw, Treatment on Day 0 **SEX: FEMALE**

Cage No.	Animal No.	Body weight (g) Days				Body Weight Gain (g)			
		-1	0	7	14	-1-0	0-7	7- 14	-1 - 14
1	2453	224	208	253	272	-16	45	19	48
5	2457	234	216	236	242	-18	20	6	8
7	3829	238	221	254	288	-17	33	34	50
9	3831	234	223	237	252	-11	14	15	18
Mean:		232.5	217.0	245.0	263.5	-15.5	28.0	18.5	31.0
Standard deviation:		6.0	6.7	9.8	20.6	3.1	13.8	11.7	21.2

TABLE 2 Body Weight and Body Weight Gain (Continued)

DOSE LEVEL:2000 mg/kg bw, Treatment on Day 0 **SEX: FEMALE**

Cage No.	Animal No.	Body weight (g) Days				Body Weight Gain (g)			
		-1	0	7	14	-1-0	0-7	7- 14	-1 - 14
2	2454#	225	204	-	-	-21	-	-	-
6	2458#	243	225	-	-	-18	-	-	-
8	3830#	235	216	-	-	-19	-	-	-
10	3832#	214	203	-	-	-11	-	-	-
Mean:		229.3	212.0	-	-	-17.3	-	-	-
Standard deviation:		12.6	10.5	-	-	4.3	-	-	-

= Found Dead

TABLE 3 Macroscopic Findings

DOSE LEVEL: 175 mg/kg bw, Treatment on Day 0 **SEX:FEMALE**

Cage No.	Animal ID	Necropsy Day	External Observations	Internal Observations	Organ/Tissue
4	2456	Day 14	No external observations recorded	No internal observations recorded	Not applicable

DOSE LEVEL: 550 mg/kg bw, Treatment on Day 0 **SEX:FEMALE**

Cage No.	Animal ID	Necropsy Day	External Observations	Internal Observations	Organ/Tissue
1	2453	Day 14	No external observations recorded	No internal observations recorded	Not applicable
5	2457	Day 14	No external observations recorded	No internal observations recorded	Not applicable
7	3829	Day 14	No external observations recorded	No internal observations recorded	Not applicable
9	3831	Day 14	No external observations recorded	No internal observations recorded	Not applicable

TABLE 3 Macroscopic Findings (Continued)

DOSE LEVEL: 2000 mg/kg bw, Treatment on Day 0 **SEX:FEMALE**

Cage No.	Animal ID	Necropsy Day	External Observations	Internal Observations	Organ/Tissue
2	2454#	Day 1	No external observations recorded	Dark discoloration, red, diffuse	Thymus
				Dark discoloration, red, diffuse, all lobes	Lungs
				Non collapsed	
				Dark discoloration, red, multifocal, glandular mucosa	Stomach
6	2458#	Day 0	No external observations recorded	Non collapsed	Lungs
				Dark discoloration, red, diffuse, all lobes	
8	3830#	Day 0	No external observations recorded	Non collapsed	Lungs
				Liquid material, brown	Stomach
10	3832#	Day 1	No external observations recorded	Non collapsed	Lungs
				Dark discoloration, red, diffuse, all lobes	

= Found Dead

APPENDICES SECTION

APPENDIX 1 Pathology Report

PATHOLOGY REPORT

Introduction

The objective of the study was to assess the acute oral toxicity of SYN545192 EC (A15457H) when administered in a single dose to rats at dose levels of 175, 550 and 2000 mg/kg bw. The results of the study allows the calculation of the estimated oral LD_{50} of the test item and permits the test item to be ranked according to most classification systems currently in use.

Results and Discussion

Surviving animals were euthanized upon completion of the observation period on Day 14. Rats were anesthetized with pentobarbital, followed by exsanguination. Gross pathology consisted of an external examination, including identification of all clinically-recorded lesions, as well as a detailed internal examination. Histopathological examination was not performed.

Mortality

Four females dosed at 2000 mg/kg bw were found dead on Day 0 or 1. Necropsy was performed on 4/4 female rats dosed at 2000 mg/kg bw.

FOUND DEAD

Dark/red discoloration and/or non-collapsing lungs was found in 4/4 found dead females at necropsy. Dark/red discoloration was also seen in the thymus and stomach glandular mucosa.

In addition, brown liquid was observed in the stomachof the found dead animal 3830 which was considered to be associated with administrated test item.

TERMINAL (DAY 14)

Macroscopic Findings

There was no evidence of the macroscopic observations at dose levels of 175 mg/kg bw and 550 mg/kg bw.

APPENDIX 1 Pathology Report (continued)

Conclusion

A single oral gavage of SYN545192 EC (A15457H) to the CRL:(WI) female rat led to the death of four animals at a dose level of 2000 mg/kg bw. A specific cause of death was not determined for these animals. In one female, brown liquid observed in the stomach was considered to be associated with the administrated test item.

No gross observations were seen in animals dosed at 175 or 550 mg/kg bw of SYN545192 EC (A15457H), subjected to the necropsy on Day 14.

05 04 2012

Peter Maslej, D.V.M., Ph.D. Date
Head, Pathology Department

APPENDIX 2 Certificate of Analysis

GLP Testing Facility WMU
Analytical Development &
Product Chemistry GS2131

Syngenta Crop Protection
Münchwilen AG
Breitenloh 5
CH-4333 Münchwilen

Certificate of Analysis

A15457H
benzovindiflupyr EC (100)
SMU1LP001

Batch Identification	**SMU1LP001**
Product Code	**A15457H**
Other Product Code(s)	benzovindiflupyr EC (100) SYN545192 EC (100)

Chemical Analysis (Active Ingredient Content)

– **Identity of the Active Ingredient(s)***	**confirmed**
– **Content of benzovindiflupyr (SYN545192) ***	**10.1 % w/w corresponding to 98.6 g/l**
– **Content of water ***	**0.74 % w/w**

The Active Ingredient(s) content is within the FAO limits.

Methodology used for Characterization	HPLC, Karl Fischer titration, OECD 109 (oscillating density meter)

Physical Analysis

– **Appearance**	**brown liquid**
– **Density ***	**976 kg/m^3**

Stability:

– **Storage Temperature**	**< 30°C**
– **Recertification Date**	**End of July 2014**

If stored under the conditions given above, this test substance can be considered stable until the recertification date is reached.

This Certificate of Analysis summarizes data which originates either from a single study or from several individual studies. Tests marked with an asterisk (*) have been conducted in compliance with GLP. Raw data, documentation, study plans, any amendments to study plans and reports pertaining to this/these study/studies are stored under the study number(s) referenced below within the archives of the GLP Testing Facility WMU at Syngenta Crop Protection Muenchwilen AG.

Study number of batch characterization: 124140

Authorisation:

06 February 2012

E. Ebi
Analytical Development & Product Chemistry

APPENDIX 3 GLP Certificate

ORSZÁGOS GYÓGYSZERÉSZETI INTÉZET
National Institute of Pharmacy

FŐIGAZGATÓ
1051 Budapest, Zrínyi u. 3.
tel: (1) 8869-320
fax: (1) 8869-480
e-mail: szepezdi.zsuzsanna@ogyi.hu

Ref. no: OGYI/8242-11/2010
Admin.: Urbin Magdolna Zita
Date: 16 December, 2010

GOOD LABORATORY PRACTICE (GLP) CERTIFICATE

It is hereby certified that the test facility

LAB Research Kft.

(Base facility: H-8201 Veszprém, Szabadságpuszta, Hungary)

is able to carry out

physico-chemical testing, toxicity studies, mutagenicity studies, environmental toxicity studies on aquatic or terrestrial organisms, studies on behaviour in water, soil and air;bio-accumulation , safety pharmacology testing, reproduction toxicology, inhalation toxicology, analytical chemistry and contract archiving

in compliance with the Principles of GLP (Good Laboratory Practice) and also complies with the corresponding OECD/European Community requirements.

Date of the inspection: **4-8 October, 2010.**

Zsuzsanna Szepezdi, Ph. D.
Director-General

Translation (from Hungarian to English):
Stamp Translation = Országos Gyógyszerészeti Intézet (OGYI) = National Institute of Pharmacy
Főigazgató = Director-General

The facility name was LAB Research Ltd until 1st September 2011. From this date, the registered name is now CiToxLAB Hungary Ltd., this information has been transmitted to the GLP competent authority. The above GLP certificate is valid for this facility (now known as CiToxLAB Hungary Ltd) until the certificate expires (16 December 2012).