

Test No.: 943512
Test substance : CGA 48988 T

**Report on the test for
Activated Sludge Respiration Inhibition
of CGA 48988 techn.**

Study Director : 5.12.e.Woo

Testing Facility : CIBA-GEIGY Ltd.
Product Safety
Ecotoxicology
CH-4002 Basel / Switzerland

Test Guideline : OECD Guideline No. 209 (April 1984)

Study completed : 24/03/94

Sponsor : CIBA-GEIGY Ltd
PP- Division
CH-4002 Basel / Switzerland

represented by : Dr. 5.12.e.Woo

Project No. of Sponsor : 943512

This report contains 13 pages.

European Registration Dossier Dossier File N°: 8.7/1 Ciba File N°: 48988/3528

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Certification of GLP and Verification of the Report

(Certification of Good Laboratory Practice and Verification of a Complete and unaltered Copy of the Report by the sponsor)

The Statement of Compliance with Good Laboratory Practice found on this page of this report, and signed by the Study Director is truthful and accurate. This report as provided by the testing facility is complete and unaltered.

For the Sponsor : Dr.

Signature :

date :

Statement of Compliance with Good Laboratory Practices

This study has been performed in compliance with Good Laboratory Practice (GLP) in Switzerland, Procedures and Principles, March 1986 (Verfahren und Grundsätze der Guten Laborpraxis (GLP) in der Schweiz), issued by the Swiss Federal Department of the Interior and the Intercantonal Office for the Control of Medicaments. These procedures are in essence consistent with:

- OECD Principles of Good Laboratory Practice (Council Decision 81/30, adopted on May 12, 1981, and the OECD Recommendation 83/95 concerning the 'Mutual Recognition of Compliance with Good Laboratory Practice', adopted on July 26, 1983).
- United States Environmental Protection Agency, Title 40 Code of Federal Regulations Part 160 (FIFRA); Federal Register, August 17, 1989.
- United States Environmental Protection Agency, Title 40 Code of Federal Regulations Part 792 (TSCA); Federal Register, August 17, 1989.
- Japan Ministry of Agriculture, Forestry and Fisheries, NohSan, Notification No. 3850, Agricultural Production Bureau, August 10, 1984.

Study Director :

Signature :

Date :

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Reserved page for flagging statements

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Quality Assurance Statement

Ciba-Geigy Ltd., Toxicology Services, Quality Assurance (GLP), 4002 Basle

Project 943512

Test Substance CGA 48988 tech.

Study Title Test for Activated Sludge Respiration Inhibition of
CGA 48 988 techn.

Study Director 5.12.e Woo

QA-Inspector

I hereby certify that the following Quality Assurance activities were performed:

Activity	Performed	Reported
Facility Inspection	October 06, 1993	November 03, 1993
Protocol Audit	February 11, 1994	February 11, 1994
Final Report Audit	March 22, 1994	March 23, 1994

hard 28, 1994

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Date

5.12.e Woo

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1. Summary and results

Study : Determination of the inhibitory concentration of a chemical substance on the respiration of aerobic waste water bacteria

Test system : Activated sludge from a sewage treatment plant.

Duration : 3 hours

Guideline : OECD Guideline No. 209 (April 1984)

Deviations : Instead of a centrifuged sludge a settled sludge was used.
The test substance was applied directly to the test medium and not as a stock solution (see paragraph 3.5)

Results : Calculations based on nominal concentrations:

EC50(3h) : >100 mg/l

EC20(3h) : >100 mg/l

EC80(3h) : >100 mg/l

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2. Introduction

2.1. Purpose : To determine the inhibitory concentration of the test substance on aerobic waste-water bacteria.

Results from pretests for this study or studies not fulfilling the validity criteria are not reported but documented in the raw data.

2.2. Guideline : OECD Guideline No. 209 (April 1984)

2.3. Deviations :

- Guideline :

Instead of a centrifuged sludge a settled sludge was used.

The test substance was applied directly to the test medium and not as a stock solution (see paragraph 3.5)

2.4. Testing Facility : CIBA-GEIGY Ltd.
Product Safety
Ecotoxicology
R-1066.P.
CH-4002 Basel / Switzerland

2.5. Archives : CIBA-GEIGY Ltd.
R-1066.K.247
CH-4002 Basel

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2.6. Personnel

- Study director :

5.1.2.e Wop

Date :

24/03/94

- Test Facility Management :

Dr.

5.1.2.e Wop

Date :

25/03/94

Technical personnel : 5.1.2.e Wop (assistant)

The job descriptions and the summaries of training and professional experience for all personnel participating in this study are archived in the test facility.

2.7. Dates :

Start of study (study plan) : 08/02/94

Experimental start : 23/02/94

Experimental end : 23/02/94

Study completed : see page 1

2.8. Distribution :

Sponsor

Quality assurance unit

Archives

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3. Materials and Methods

3.1. Test substance

Identification Code : CGA 48988 T
Generic/Trade name : Metalaxyl
Batch No.: EN 603107
Appearance : beige solid
Purity : 96.1 %
Solubility (in water) : 0.71 % at 20°C
Received : 28/01/94
Storage : room temperature
Stability : 06/94

3.2. Reference substance

3,5-Dichlorophenol

3.3. Test system / inoculum

Activated sludge collected from the sewage treatment plant of CH-4153 Reinach on 22/02/94. The pH after collection was 6.8. The preparation was carried out according to the method described in the guideline. The sludge was separated from the aqueous layer by settling instead of centrifugation. The pH of the sludge before use was 7.8.

3.4. Design and procedure

Vessels : 250 ml BOD flasks with gas inlet
Water : Dechlorinated drinking water
Temperature : $20 \pm 2^\circ\text{C}$
Duration : 3 hours
Nutrient solution : 16.0 g Peptone
11.0 g Meat extract
3.0 g Urea
0.7 g NaCl
0.4 g $\text{CaCl}_2 \cdot 2 \text{H}_2\text{O}$
0.2 g $\text{MgSO}_4 \cdot 7 \text{H}_2\text{O}$
2.8 g K_2HPO_4

were dissolved and the volume was made up to one liter with dechlorinated drinking water.

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3.5. Application

The test substance amount of each concentration (see ahead) was firstly added to water, each mixture was homogenized and directly added to the test medium (using a total volume of 114 ml water).

3.6. Test concentrations

Test substance : 106, 61.0, 37.5, 23.0 and 13.0 mg/l
 21.2, 12.2, 7.5, 4.6 and 2.6 mg of the test substance were weighed and added to the test medium. The volume was adjusted to 200 ml with water and aerated for 3 hours.

Reference substance : 32.1, 10.0 and 3.2 mg/l

The sludge concentration in the test bottles was 1.68 g/l (dry weight).

3.7. Measurements

Oxygen consumption per hour in mg/liter with a ORION - Electrode Type 97-08, ORION Microprocessor Ionalyzer 901 and plotted on a recorder.

3.8. Calculations/Statistical Analysis

The inhibitions were calculated on the basis of a measured time dependent oxygen consumption of the Blank and the test solution. The EC values were determined after calculating the linear regression of the results.

The following formulas were used:

c = concentration
 i = inhibition
 n = number of values

$$A = \frac{\sum(\ln(c) * i) - (\sum(\ln(c)) * \sum(i) / (n - 1))}{\sum(\ln(c))^2 - (\sum(\ln(c)) * \sum(\ln(c)) / (n - 1))}$$

$$B = \sum(i) / (n-1) - A * \sum(\ln(c)) / (n - 1)$$

$$EC_{xx} = \text{Exp}((xx - B) / A)$$

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4. Results

4.1. Values

Calculations based on nominal concentrations lead to the following results

For the test substance:

EC50(3h): >100 mg/l

EC20(3h) : >100 mg/l

EC80(3h): >100 mg/l

For the reference substance::

EC50(3h) = 16.4 mg/l

5. Tables and Figures

Sample	Conc. [mg/l]	Consumption rate [mg/l/h]	Inhibition [%]	pH
Blank 1	0.0	45.6	3	8.2
Blank 2	0.0	48.0	-3	8.2
Reference 1	32.1	12.6	73	8.2
Reference 2	10.0	34.1	27	8.2
Reference 3	3.2	42.3	10	8.2
Test conc. 1	106.0	48.5	-4	8.3
Test conc. 2	61.0	48.0	-3	8.2
Test conc. 3	37.5	48.0	-3	8.2
Test conc. 4	23.0	50.8	-9	8.2
Test conc. 5	13.0	48.0	-3	8.2

¹The pH was measured after 3h aeration.

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Inhibitory concentration of CGA 48988 techn.

