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[redacted]	[redacted]
[redacted]	[redacted]

Table A7.4.3.5.1.b/01-5: Test conditions

Criteria	Details
Test temperature	24.2 to 25.3°C
Dissolved oxygen	4.6 to 7.4 mg/L (64 to 103 % saturation)
pH	overlying water = 7.02 to 8.31
Adjustment of pH	Not described
Salinity	20.2 to 20.7 ‰
Total hardness	Not described
Ammonia	overlying water = 0.0088 to 9.0 µg/L pore water = 0.0081 to 48 µg/L
Aeration of dilution water	Yes. Aeration was provided at an initial rate of 60-100 bubbles per minute to each test chamber through a glass pipet. The pipet was inserted such that its tip was 2-3 cm from the sediment surface.
Quality/Intensity of irradiation	fluorescent
Photoperiod	16 h daylight, 8 h dark with 30 minute transition periods

Table A7.4.3.5.1.b/01-6: Analytical measured concentrations: HPLC measurements

Mean Measured Sediment Concentrations (mg 14[C] equivalents per kg dry sediment)	Measured Concentrations as mg RH-287 Technical/kg Dry Sediment (Percent of Nominal) Based on HPLC Analysis				
	Day 0	Day 2	Day 7	Day 28	Mean
0.0 Negative Control	< MQL	--	--	< MQL	< MQL
0.0 Acetone Control	< MQL	< MQL	< MQL	< MQL	< MQL
4.6	< MQL	< MQL	< MQL	< MQL	< MQL
10	0.146 (1)	--	--	< MQL	< MQL
19	0.289 (1)	--	--	< MQL	0.170 (<1)
38	0.853 (2)	--	--	< MQL	0.452 (<1)
78	2.20 (2)	0.762 (1)	0.642 (1)	0.359 (<1)	0.991 (1)

Table A7.4.3.5.1.b/01-7: Analytical measured concentrations: LSC measurements

Mean Measured Sediment Concentrations (mg 14[C] equivalents per kg dry sediment)	Measured RH-287 Technical Concentration as mg 14[C] eqivalents/kg Dry Sediment (Percent of Nominal) Based on LSC Analysis					
	Day 0	Day 7	Day 14	Day 21	Day 28	Mean
0.0 Negative Control	< MQL	< MQL	< MQL	< MQL	< MQL	< MQL
0.0 Acetone Control	< MQL	< MQL	< MQL	< MQL	< MQL	< MQL
4.6	5.9 (98)	4.7 (78)	3.8 (63)	4.4 (73)	4.3 (72)	4.6 (77)
10	11 (92)	10 (83)	8.5 (71)	10 (83)	11 (92)	10 (83)
19	22 (92)	20 (83)	19 (79)	18 (75)	18 (75)	19 (79)
38	38 (79)	40 (83)	39 (81)	37 (77)	35 (73)	38 (79)
78	84 (88)	77 (80)	76 (79)	71 (74)	81 (84)	78 (81)

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Table A7.4.3.5.1.b/01-8: Effect and Mortality data

Test-Substance Concentration (nominal) [1] [mg DCOIT/kg dry sediment]	28 Day Mean Survival	28 Day Mean Juveniles per Adult	28 Day Mean dry weight (mg/animal)	28 Day Mean Growth Rate (mg/day)	Day 28 Treatment Percent Survival
control	17.4	9.8	1.86	0.060	87
acetone control	18.2	7.8	1.73	0.056	90
6.0	17.8	9.6	1.69	0.054	89
12	15.2	10.0	1.81	0.058	76
24	12.4	9.0	1.69	0.054	62 *
48	13	5.6	1.53	0.049	65 *
96	9	7.0	1.46♣	0.046♣	45 *

* Statistically significant (p <0.05) reduction in treatment survival as compared to the pooled control value.

♣ Statistically significant (p <0.05) reduction in treatment biomass as compared to the pooled control value (only for comparisons based on pore water concentration of ^{14}C-DCOIT equivalents)

Note: Pooled control survival = 88%.

Table A7.4.3.5.1.b/01-9: Effect data

Biological Parameter	LC_{50} or EC_{50} [a] (95% CI [b])	Statistical Endpoints NOEC [c]	LOEC	MATC
	Expressed as mg DCOIT per kg Dry Sediment			
28-day Survival	≥0.991 (N/C[d])	0.0988 [e]	0.170	0.130
Reproductive Output	>0.991 (N/C)	0.991	>0.991	N/C
Dry Weight	>0.991 (N/C)	0.991	>0.991	N/C
Growth Rate	>0.991 (N/C)	0.991	>0.991	N/C
	Expressed as mg ^{14}C-DCOIT Equivalents per kg Dry Sediment			
28-day Survival	≥78 (N/C)	10	19	14
Reproductive Output	>78 (N/C)	78	>78	N/C
Dry Weight	>78 (N/C)	78	>78	N/C
Growth Rate	>78 (N/C)	78	>78	N/C
	Expressed as µg DCOIT per Liter of Pore Water			
28-day Survival	N/C [f]	N/C	N/C	N/C
Reproductive Output	N/C	N/C	N/C	N/C
Dry Weight	N/C	N/C	N/C	N/C
Growth Rate	N/C	N/C	N/C	N/C
	Expressed as µg ^{14}C-DCOIT Equivalents per Liter of Pore Water			
28-day Survival	>539 (N/C)	32	124	63.0
Reproductive Output	>539 (N/C)	539	>539	N/C
Dry Weight	>539 (N/C)	124	539	259
Growth Rate	>539 (N/C)	124	539	259

[a] Median effect or lethal concentration was determined by trimmed Spearman-Karber method.
[b] CI = confidence interval.
[c] NOEC was determined by Dunnet's test and is presented based on mean measured concentrations
[d] N/C – Could not be calculated.
[e] Estimated value. Actual value is less than MQL (0.103 mg/kg).
[f] There were no measurable concentrations of DCOIT within these samples.

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Table A7.4.3.5.1.b/01-10: Validity criteria

	fulfilled	Not fullfilled
Neonate *L. plumulosus*, size-selected (retained between 0.25-mm and 0.6-mm screens) or age selected	**yes**	
Average survival of amphipods in the negative control sediment must be greater than or equal to 80% at the end of the test, with no single replicate having 60% survival or less.	**yes**	
Measurable growth and reproduction should be observed in all replicates of the negative control treatment.	**yes**	
The time-weighted average of daily temperature readings must be within +2°C of the desired temperature. The instantaneous temperature must always be within +3°C of desired temperature.	**yes**	
The time-weighted average of daily salinity readings must be 5‰ ±2‰ or 20‰ ± 2‰. The instantaneous salinity readings must always be 5‰ ±3‰ or 20‰ ± 3‰.	**yes**	

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Section A7.4.3.5.1 b/02 Chronic toxicity to sediment dwelling organisms-Marine water, *Neanthes arenaceodentata*

Annex Point IIIA XIII.3.4

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		1 REFERENCE
1.1	**Reference**	Reference Type: test repoprt Year: 2003 Report date: 2 October 2003 [redacted]
1.2	**Data protection**	Yes
1.2.1	Data owner	Rohm and Haas Company
1.2.2		
1.2.3	Criteria for data protection	[redacted]
		2 GUIDELINES AND QUALITY ASSURANCE
2.1	**Guideline study**	Yes, American Society for Testing & Methods (ASTM) E1611 and Biocidal Products Directive (98/8/EC) Technical Guidance Document.
2.2	**GLP**	Yes
2.3	**Deviations**	No
		3 MATERIALS AND METHODS
3.1	**Test material**	DCOIT (RH-287 Technical), ^{14}C-DCOIT
3.1.1	Lot/Batch number	[redacted]
3.1.2	Specification	The test substance was radiolabelled. Unlabelled DCOIT specification was as given in section 2
3.1.3	Purity	DCOIT : 99.3%; ^{14}C-DCOIT specific activity = 24.50 mCi/g, radiopurity = 96.80%.
3.1.4	Composition of Product	[redacted]
3.1.5	Further relevant properties	[redacted]
3.1.6	Method of analysis	[redacted]

Section A7.4.3.5.1 b/02 **Chronic toxicity to sediment dwelling organisms-Marine water, *Neanthes arenaceodentata***

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3.2 Preparation of TS solution for poorly soluble or volatile test substances	[redacted]	
3.3 Reference substance	[redacted]	
3.4 Testing procedure		
3.4.1 Dilution water	[redacted]	
3.4.2 Test organisms	[redacted]	
3.4.3 Test system	[redacted]	
3.4.4 Test conditions	see table A7.4.3.5.1.b/02-5	
3.4.5 Duration of the test	28 days	
3.4.6 Test parameter	[redacted]	
3.4.7 Sampling	[redacted]	
3.4.8 Monitoring of TS concentration	[redacted]	
3.4.9 Statistics	[redacted]	
4 RESULTS		
4.1 Limit Test	Not performed	
4.2 Results test substance		
4.2.1 Initial concentrations of test substance	0 (control), 0 (6.3 mL/kg acetone control), 5.0, 10, 20, 40, and 80 mg DCOIT/kg dry sediment	x
4.2.2 Actual concentrations of test substance	Mean Measured Sediment Concentrations (HPLC): <MQL (control), <MQL (6.3 mL/kg acetone control), <MQL, 0.108, 0.223, 0.212, and 1.20 mg DCOIT/kg dry sediment	x

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	Mean Measured Sediment Concentrations (LSC): <MQL (control), <MQL (6.3 mL/kg acetone control), 4.9, 9.9, 20, 28, and 69 mg ^{14}C-DCOIT equivalents/kg dry sediment
	Analytical results can be found in tables A7.4.3.5.1.b/02-6 and A7.4.3.5.1.b/02-7.
	Mean Measured overlying water Concentrations (HPLC): <MQL (control), <MQL (6.3 mL/kg acetone control), <MQL,, 0.982, 5.48, 13.2 and 20.48 µg DCOIT/L
	Mean Measured overlying water Concentrations (LSC): <MQL (control), <MQL (6.3 mL/kg acetone control), 40.3 (4.9 mg ^{14}C-DCOIT equivalents/kg dry sediment), 122 (20 mg ^{14}C-DCOIT equivalents/kg dry sediment) and 458 (69 mg ^{14}C-DCOIT equivalents/kg dry sediment) µg ^{14}C-DCOIT equivalents/L
	Mean Measured pore water Concentrations (HPLC): <MQL (control), <MQL (6.3 mL/kg acetone control), <MQL,, <MQL, <MQL, <MQL and <MQL
	Mean Measured pore water Concentrations (LSC): <MQL (control), <MQL (6.3 mL/kg acetone control), 72 (4.9 mg ^{14}C-DCOIT equivalents/kg dry sediment), 341 (20 mg ^{14}C-DCOIT equivalents/kg dry sediment) and 1220 (69 mg ^{14}C-DCOIT equivalents/kg dry sediment) µg ^{14}C-DCOIT equivalents/L
4.2.3 Effect data	see table A7.4.3.5.1.b/02-8 and see table A7.4.3.5.1.b/02-9
4.2.4 Concentration / response curve	Not described in report
4.2.5 Other effects	Not applicable
4.3 Results of controls	see table A7.4.3.5.1.b/02-8
4.4 Test with reference substance	Not performed

5 APPLICANT'S SUMMARY AND CONCLUSION

5.1 Materials and methods	Yes, American Society for Testing & Methods (ASTM) E1611 and Biocidal Products Directive (98/8/EC) Technical Guidance Document, Chronic *Neanthes arenaceodentata* toxicity study in a sediment-water system with analytical confirmation of TS concentrations.
5.2 Results and discussion	The DCOIT recoveries from the natural sediment matrix used in the the survival and reproduction study with *Neanthes arenaceodentata* (Rohm and Haas Report N° 02RC-0052) and *Leptocheirus plumulosus* (Rohm and Haas Report N° 02RC-0050) were much lower than the recoveries from the formulated sediment used in the survival and emergence study with *Chironomus riparius* (Rohm and Haas Report N° 02RC-0051). The differences in the particle size distribution as well as the particle

Section A7.4.3.5.1 b/02 **Chronic toxicity to sediment dwelling organisms-Marine water, *Neanthes arenaceodentata***

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		size and types of organic carbon within these different matrices most likely attributed to the differing amounts of recoverable DCOIT in the sediment extracts.
5.2.1	LOEC	see table A7.4.3.5.1.b/02-9
5.2.2	NOEC	see table A7.4.3.5.1.b/02-9
5.2.3	LC_{50}	see table A7.4.3.5.1.b/02-9
5.2.4	MATC	see table A7.4.3.5.1.b/02-9
5.3	**Conclusion**	
5.3.1	Reliability	(1), reliable without restriction
5.3.2	Deficiencies	No

Evaluation by Competent Authorities	
	Evaluation by Rapporteur Member State
Date	18 January 2008
Materials and Methods	Agree with applicant's version
Results and discussion	**Comment (4.2.1):** The concentrations given here are nominal concentrations of DCOIT.
	Comment (4.2.2): Test concentrations in sediment have been measured with HPLC and LSC. From the HPLC measurements it becomes clear that parent DCOIT rapidly disappears from the test system. Measurements already at day 0 show that DCOIT concentrations have declined considerably: MQL (control), <MQL (acetone control), <MQL, 0.164, 0.394, 0.373, and 2.80 mg DCOIT/kg dry sediment. Therefore, results have to be calculated based on mean measured concentrations.
Conclusion	**Comment (5.3):** No LOEC can be established from this test as no effects have been seen at the highest concentration tested.
Reliability	1, reliable without restrictions
Acceptability	Acceptable
Remarks	-

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Section A7.4.3.5.1 b/02 Annex Point IIIA XIII.3.4	Chronic toxicity to sediment dwelling organisms-Marine water, *Neanthes arenaceodentata* – **TABLES AND FIGURES**

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Table A7.4.3.5.1.b/02-5: Test conditions

Criteria	Details
Test temperature	overlying water: 17.8 to 21.4°C
Dissolved oxygen	overlying water: 3.6 to 8.8 mg/L (49 to 117 % saturation)
pH	overlying water: 7.42 to 8.29
Adjustment of pH	Not described
Salinity	overlying water: 30.7 to 31.5‰
Total hardness	Not described
Ammonia	overlying water: 0.71 to 66 µg/L pore water: 0.0053 to 280 µg/L
Aeration of dilution water	Yes. Aeration was provided at an initial rate of 60-100 bubbles per minute to each test chamber through a glass pipet. The pipet was inserted such that its tip was 2-3 cm from the sediment surface.
Quality/Intensity of irradiation	fluorescent
Photoperiod	16 h daylight, 8 h dark with 30 minute transition periods

Table A7.4.3.5.1.b/02-6: Analytical measurements: result of HPLC measurements

	Measured Concentrations as mg RH-287 Technical/kg Dry Sediment (Percent of Nominal) Based on HPLC Analysis				
Mean Measured Sediment Concentrations (mg 14[C] equivalents per kg dry sediment)	Day 0	Day 2	Day 7	Day 28	Mean
0.0 Negative Control	< MQL	--	--	< MQL	< MQL
0.0 Acetone Control	< MQL	< MQL	0.603*	< MQL	< MQL
4.9	< MQL	< MQL	< MQL	< MQL	< MQL
9.9	0.164 (2)	--	--	< MQL	0.108 (1)
20	0.394 (2)	--	--	< MQL	0.223 (1)
28	0.373 (1)	--	--	< MQL	0.212 (<1)
69	2.80 (4)	1.53 (2)	0.407 (1)	< MQL	1.20 (2)

*Due to contamination and not included in statistical analysis.

Table A7.4.3.5.1.b/02-7: Analytical measurements: result of LSC measurements

Mean Measured Sediment Concentrations (mg 14[C] equivalents per kg dry sediment)	Measured RH-287 Technical Concentration as mg 14[C] eqivalents/kg Dry Sediment (Percent of Nominal) Based on LSC Analysis					
	Day 0	Day 7	Day 14	Day 21	Day 28	Mean
0.0 Negative Control	< MQL	< MQL	< MQL	< MQL	< MQL	< MQL
0.0 Acetone Control	< MQL	< MQL	< MQL	< MQL	< MQL	< MQL
4.9	5.1 (102)	5.0 (100)	4.8 (96)	4.7 (94)	4.9 (90)	4.9 (98)
9.9	11 (110)	10 (100)	9.4 (94)	9.8 (98)	9.5 (95)	9.9 (99)
20	20 (100)	21 (105)	20 (100)	19 (95)	19 (95)	20 (100)
28	28 (70)	28 (70)	28 (70)	28 (70)	27 (68)	28 (70)
69	70 (88)	72 (90)	68 (85)	68 (85)	66 (83)	69 (86)

Table A7.4.3.5.1.b/02-8: Effect and Mortality data

Test-Substance Concentration (nominal) [1] [mg DCOIT/kg dry sediment]	Day 28 Mean growth rate (mg per animal per day)	Day 28 Mean dry weight (mg per animal)	Day 28 Percent mortality
control	0.24	7.29	4
acetone control	0.26	7.87	4
5	0.24	7.14	4
10	0.23	7.09	0
20	0.26	7.88	8
40	0.24	7.19	4
80	0.24	7.19	4

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Table A7.4.3.5.1.b/02-9: Effect data

Biological Parameter	Statistical Endpoints			
	LC_{50} or EC_{50} (95% CI [a])	NOEC [b]	LOEC	MATC
	Expressed as mg DCOIT per kg Dry Sediment			
28-day Survival	>1.20 (N/C[c])	1.20	>1.20	N/C
Dry Weight	>1.20 (N/C)	1.20	>1.20	N/C
Growth Rate	>1.20 (N/C)	1.20	>1.20	N/C
	Expressed as mg ^{14}C-DCOIT equivalents per kg Dry Sediment			
28-day Survival	>69 (N/C)	69	>69	N/C
Dry Weight	>69 (N/C)	69	>69	N/C
Growth Rate	>69 (N/C)	69	>69	N/C
	Expressed as µg DCOIT per Liter of Pore Water			
28-day Survival	N/C[d]	N/C	N/C	N/C
Dry Weight	N/C	N/C	N/C	N/C
Growth Rate	N/C	N/C	N/C	N/C
	Expressed as µg ^{14}C-DCOIT equivalents per Liter of Pore Water			
28-day Survival	>1,220 (N/C)	1,220	>1,220	N/C
Dry Weight	>1,220 (N/C)	1,220	>1,220	N/C
Growth Rate	>1,220 (N/C)	1,220	>1,220	N/C

[a] CI = confidence interval.
[b] NOEC was determined by the Dunnett's test and is presented based on mean measured concentrations
[c] N/C – Could not be calculated.
[d] There were no measurable concentrations of DCOIT within these samples.

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Table A7.4.3.5.1.b/02-10: Validity criteria

	fulfilled	Not fullfilled
The emergence in the controls must be at least 70% at the end of the test (1)(6);	**yes**	
>/= 90% mean survival for the control animals with >/= 80% control survival for individual replicates	**yes**	
All test chambers must be identical	**yes**	
Treatments must be randomly assigned	**yes**	
Test organisms must be impartially or randomly assigned	**yes**	
A negative, reference sediment, positive, or solvent controls must be included in the testing	**yes**	
Test animals must be the same species and from the same population or culture	**yes**	
Neanthes must be less than two to three weeks post-emergence at test initiation	**yes**	
DO must be measured in at least one test chamber in each concentration at the beginning and end of the test	**yes**	
Temperature should be measured in a test chamber from each concentration daily during the test	**yes**	
Aeration must not be off for an extended time period such that the DO drops below 60%	**yes**	
The solvent concentration did not adversely affect survival or growth	**yes**	
The analytical method must be validated prior to initiation of the test	**yes**	

Section A7.4.3.5.2 Aquatic plant toxicity - Growth inhibition test, *Lemna gibba*

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1 REFERENCE

1.1 Reference

Reference Type: test repoprt

Year: 2002

Report date: 2 July 2002

[redacted]

1.2 Data protection Yes

1.2.1 Data owner: Rohm and Haas Company

1.2.2

1.2.3 Criteria for data protection: [redacted]

2 GUIDELINES AND QUALITY ASSURANCE

2.2 Guideline study Yes, US EPA OPPTS 850.4400, OECD 221, US EPA TSCA 797.1160, US EPA FIFRA 122-2 and 123-2, EC Council Directive 67/548/EEC

2.3 GLP Yes

2.4 Deviations No

3 MATERIALS AND METHODS

3.2 Test material DCOIT (RH-287 Technical)

3.2.1 Lot/Batch number: [redacted]

3.2.2 Specification: As given in section 2

3.2.3 Purity: 98.42%

3.2.4 Composition of Product: [redacted]

3.2.5 Further relevant properties: [redacted]

3.2.6 Method of analysis: [redacted]

3.3 Preparation of TS solution for poorly soluble or volatile test substances [redacted]

3.4 Reference substance [redacted]

3.5 Testing procedure

Section A7.4.3.5.2 Aquatic plant toxicity - Growth inhibition test, *Lemna gibba*

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3.5.1	Culture medium	[redacted]	
3.5.2	Test organisms	[redacted]	
3.5.3	Test system	[redacted]	x
3.5.4	Test conditions	see table A7.4.3.5.2/01-4	
3.5.5	Duration of the test	7 days	
3.5.6	Test parameter	[redacted]	
3.5.7	Sampling	[redacted]	
3.5.8	Monitoring of TS concentration	[redacted]	
3.5.9	Statistics	[redacted]	

4 RESULTS

4.1 Limit Test — Not performed

4.2 Results test substance

4.2.1	Initial concentrations of test substance	Nominal (mg DCOIT/L) 0.0078, 0.016, 0.031, 0.063, 0.13, 0.25, 0.5, 1.0, 2.0	
4.2.2	Actual concentrations of test substance	measured day 0 / measured day 7 (mg DCOIT/L): 0.00454 / <0.00241 0.0118 / <0.00241 0.0218 / <0.00241 0.0467 / <0.00241 0.104 / <0.00241 0.196 / <0.00241 0.444 / 0.00932 0.632 / 0.0634 1.37 / 0.169	x
4.2.3	Growth curves	see Figure A7.4.3.5.2/01-1 and -2	x
4.2.4	Concentration /	Not described	

Section A7.4.3.5.2 **Aquatic plant toxicity - Growth inhibition test, *Lemna gibba***

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	response curve		
4.2.5	Frond count data	see table A7.4.3.5.2/01-5	
4.2.6	Effect data (growth inhibition)	see table A7.4.3.5.2/01-6	
4.2.7	Other observed effects	Not applicable	
4.3 Results of controls		Doubling time for the control was 1.8 days indicating acceptable growth in the control. No significant differences between the control and acetone control were detected and they were pooled for comparison to the DCOIT treatments.	
4.4 Test with reference substance		Not performed	
		5 APPLICANT'S SUMMARY AND CONCLUSION	
5.1 Materials and methods		US EPA OPPTS 850.4400, OECD 221, US EPA TSCA 797.1160, US EPA FIFRA 122-2 and 123-2, EC Council Directive 67/548/EEC, Acute static toxicity test to duckweed with analytical confirmation of TS concentrations.	
5.2 Results and discussion		see table A7.4.3.5.2/01-6	x
5.2.1	EC_5	see table A7.4.3.5.2/01-6	
5.2.2	EC_{50}	see table A7.4.3.5.2/01-6	
5.2.3	EC_{90}	see table A7.4.3.5.2/01-6	
5.3 Conclusion		the test compound was not stable in the test media following a 7-day exposure	x
5.3.1	Reliability	(1), valid with restrictions	x
5.3.2	Deficiencies	No	x

Evaluation by Competent Authorities	
	Evaluation by Rapporteur Member State
Date	8 January 2008
Materials and Methods	**Comment (3.5.5):** A semi-static or flow-through test design should have been chosen as it is known that DCOIT is not stable under aquatic ecotoxicity testing.
Results and discussion	**Comment (4.2.3):** The effect of exposure period on the endpoints frond number and area under growth curve is better visualized by growth curves plotted on an arithmetic rather than a logarithmic scale as shown in figure -2. This is because the endpoints "frond number" and "area under growth curve" do not account for the fact that the growth is exponential. For this reason the relative difference in frond number that are observed after 3 days almost the same as after 7 days, and calculations of these endpoints after 3 days are not essential. **Comment (4.2.4):** Test substance concentrations should have been monitored more closely as it is known that DCOIT rapidly disappears from aquatic test systems. **Comment (5.2):** Figure -1 shows that the control cultures grow exponentially during the entire test period, while the cultures exposed to DCOIT are inhibited mainly during the first three days of the test. The cultures with the highest concentration (nominal 1.37 mg/l) grew at the same rate as the controls between day 3 and 7 (when plotted against on a logarithmic scale straight curves indicate exponential growth and the growth rate is proportional to the slope of the lines). This shows clearly the effect of the disappearance of the test material from the solutions. Since the growth inhibiting effect is declining during the exposure period, the calculations of the endpoints are based on the initial phase of the test, in this case day 0-3. Although the analysis of the data after 7 days did not show a significant difference between the control and solvent control, the increase in frond numbers after 3 days was significantly different between the control and the solvent control. Therefore the data for day 3 should be compared to the solvent control. The calculation of the effect values for the end points frond number and frond weight are not strongly depending on the time period for which they have been calculated. However, the effect can be seen as a result of the exposure during the first three days. The NOEC for frond weight was higher than for frond number. This depends, however, on the definition of the term NOEC. The estimation of the NOEC by hypothesis testing is somewhat in conflict with a basic role of the scientific method, because there is an attempt to "prove" a null hypothesis of no effect. More correctly, the LOEC is estimated as the lowest concentration showing a significant effect and the NOEC is then defined as the test concentration below the LOEC. With this approach, the NOEC for reduction of frond weight in the Lemna test becomes 4.54 µg/l. This is because the nominal concentration 11.8 µg/l showed a significant, 15 % reduction of frond weight, while at the next higher concentration, 21.8 µg/l the reduction was only 13 %, and not significant according to the statistical calculation done in this test. According to Dunnett's test, the difference from the control was not statistically significant at 21.8 µg/l. However, with William's test all concentrations apart from the lowest are significantly different compared to the control; It seems therefore justified to take 4.5 µg/l as the NOEC for the end point frond weight even if the biological relevance of this NOEC value is somewhat uncertain because of the irregular response pattern.

Document III-A / Section A7.4.3

Conclusion	**Comment (5.3):** The scientifically preferred endpoint growth rate, calculated for the exposure period 0-3 days, is considered to be the most relevant endpoint from this study. This results in an EC50 of 0.206 mg/l and a NOEC of 0.00454 mg/L based on initial measured concentrations.
Reliability	**Comment (5.3.1 and 5.3.2):** Due to the restrictions described, the reliability is changed from 1 to 2, reliable with restrictions.
Acceptability	Acceptable with the restrictions noted above.
Remarks	As in the algae tests most of the observed effecst occur within the initial phase of the test, and the differences in frond numbers or weight observed after 7 days are mainly due to growth inhibition in the initial phase of the test. The endpoints are all estimated based on intial measured concentrations. An alternative presentation, suggested by OECD in case test concentrations are declining during the test, is to use the geometric mean concentrations during the exposure period. However, it would not be correct to express the NOECs as geometric mean concentrations over 7 days since the differences seen between the treatments are mainly due to effects of the initial exposure.

Document III-A / Section A7.4.3

Section A7.4.3.5.2	Aquatic plant toxicity-Growth inhibition test *Lemna gibba* – TABLES AND FIGURES

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Table A7.4.3.5.2/01-4: Test conditions

Criteria	Details
Test temperature	23.4 to 24.1 °C
pH	4.92 to 6.03
Aeration of dilution water	Not described
Light intensity	9435 ± 149 lux
Photoperiod	continuous "warm-white" fluorescent light

Table A7.4.3.5.2/01-5: Frond count data

Day 0 measured concentrations (mg DCOIT/L)	Day 3 normal fronds	Day 5 normal fronds	Day 7 normal fronds	Day 7 Treatment mean of normal fronds	Percent Difference (compared to pooled control)
control	160	387	824	275	---
acetone control	181	430	902	301	---
pooled control	---	---	---	288	---
0.00454	164	396	840	280	-3
0.0118	148	362	764	255	-11
0.0218	140	358	694	231	-20
0.0467	146	306	581	194	-33
0.104	140	272	504	168	-42
0.196	126	233	396	132	-54
0.444	90	195	357	119	-59
0.632	69	169	354	118	-59
1.37	60	148	297	99	-66

Table A7.4.3.5.2/01-6: Effect data

RESULTS FOR NUMBER OF NORMAL FRONDS AS COMPARED TO VEHICLE CONTROL (1-TAILED DUNNETT'S TEST)

Day 3 Growth Parameter	EC Type	EC Value (mg a.i./L)	95% Confidence Limits (mg a.i./L)	NOEC (mg a.i./L)
Normal Frond Number*	EC_5	0.00612	0.000539-0.0117	0.00450
	EC_{50}	0.438	0.354-0.523	
	EC_{90}	>1.37	---	
Area Under the Growth Curve*	EC_5	0.0210	0.000671-0.0353	0.00454
	EC_{50}	0.206	0.164-0.249	
	EC_{90}	1.13	0.723-1.54	
Growth Rate*	EC_5	0.0454	0.0180-0.0728	0.00454
	EC_{50}	0.336	0.280-0.393	
	EC_{90}	>1.37	---	

* Significant difference between control & vehicle control

"---" Indicates value could not be estimated.

Day 5 Growth Parameter	EC Type	EC Value (mg a.i./L)	95% Confidence Limits (mg a.i./L)	NOEC (mg a.i./L)
Normal Frond Number	EC_5	<0.00454	---	
	EC_{50}	0.310	0.244-0.376	0.00454
	EC_{90}	>1.37	---	
Area Under the Growth Curve*	EC_5	<0.00454	---	
	EC_{50}	0.172	0.134-0.211	0.00454
	EC_{90}	>1.37	---	
Growth Rate	EC_5	<0.00454	---	
	EC_{50}	1.15	0.786-1.51	0.0218
	EC_{90}	>1.37	---	

* Significant difference between control & vehicle control

"---" Indicates value could not be estimated.

Day 7 Growth Parameter	EC Type	EC Value (mg a.i./L)	95% Confidence Limits (mg a.i./L)	NOEC (mg a.i./L)
Normal Frond Number	EC_5	<0.00454	---	0.0118
	EC_{50}	0.203	0.152-0.255	
	EC_{90}	>1.37	---	
Area Under the Growth Curve*	EC_5	<0.00454	---	0.00454
	EC_{50}	0.162	0.129-0.194	
	EC_{90}	>1.37	---	
Growth Rate	EC_5	<0.00454	---	0.0118

* Significant difference between control & vehicle control

"---" Indicates value could not be estimated.

Figure A7.4.3.5.2/01-1: Growth curves for Duckweed, Lemna gibba, during a 7-day exposure to DCOIT

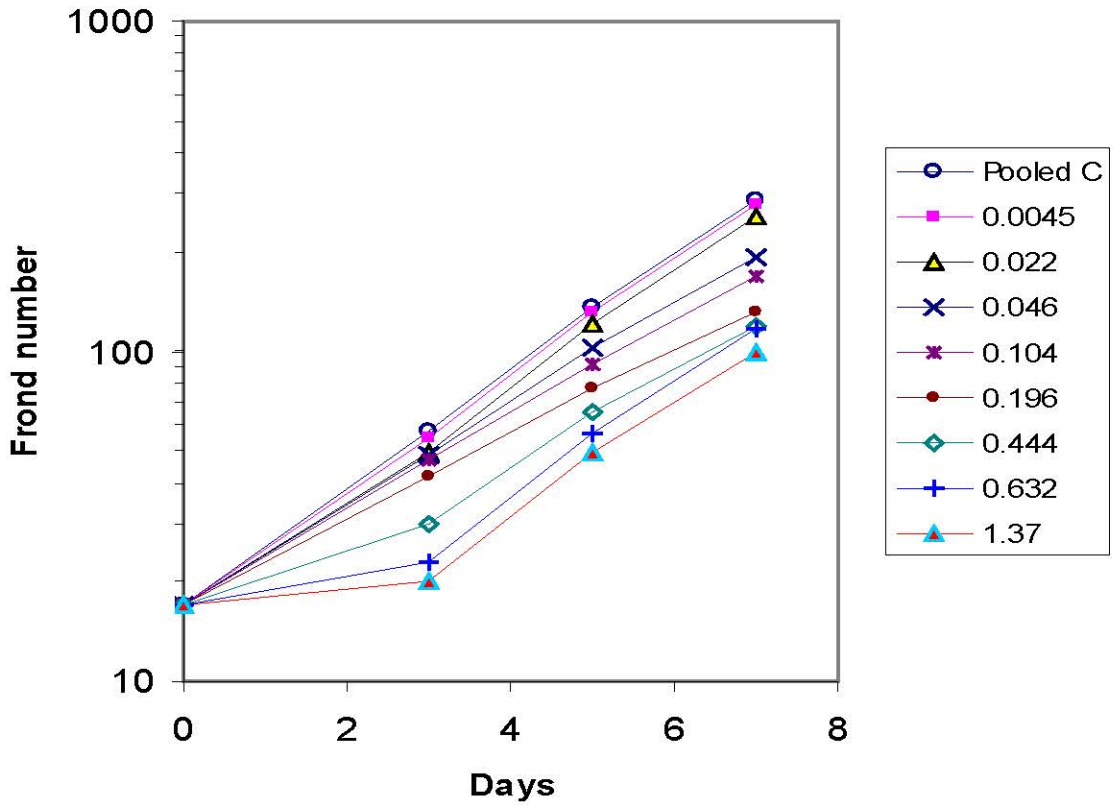
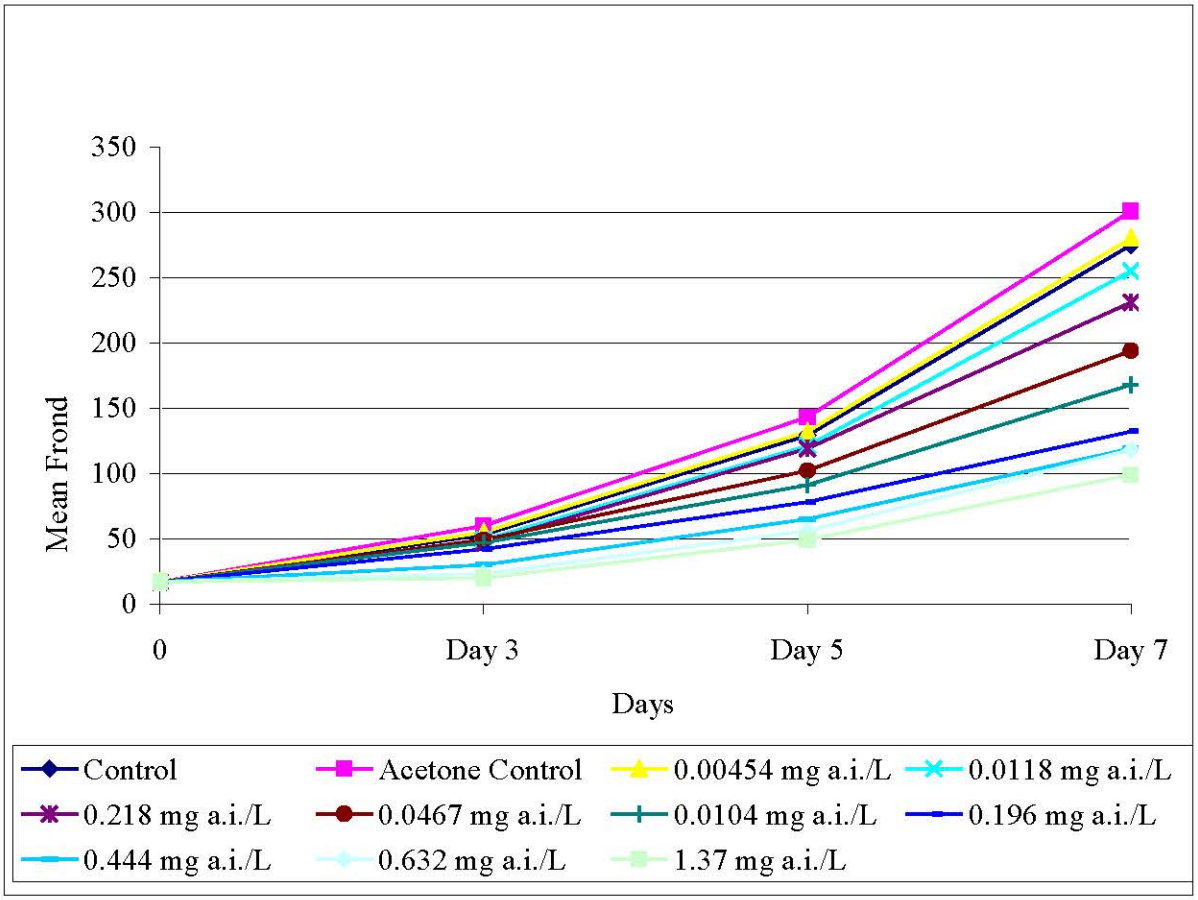


Figure A7.4.3.5.2/01-2: Effect of exposure period on the endpoints frond number and area under growth curve



Directive 98/8/EC on the placing of biocidal products on the market.

Dossier for the inclusion of an active substance in the Annex 1

4,5-Dichloro-2-octyl-2H-isothiazol-3-one (DCOIT)

Product type 21: Antifouling products

Document III-A (A7)
Study summaries – Active substance
Ecotoxicological profile including environmental fate and behaviour

Part VI

Fate and behaviour in the environment

Section A7.5: Effects on terrestrial organisms

TABLE OF CONTENTS

Document III-A / Section A7.5

Section A7.5.1.1 Inhibition to microbial activity (terrestrial)

Annex Point IIA7.4

			Official use only
		1 REFERENCE	
1.1	**Reference**	Reference Type: test repoprt Year: 2002 Report date: 15 January 2002 [redacted]	
1.2	**Data protection**	Yes	
1.2.1	Data owner	Rohm and Haas Company	
1.2.2			
1.2.1	Criteria for data protection	[redacted]	
		2 GUIDELINES AND QUALITY ASSURANCE	
2.1	**Guideline study**	Yes, OECD 216 and OECD 217	
2.2	**GLP**	Yes	
2.3	**Deviations**	No	
		3 MATERIALS AND METHODS	
3.1	**Test material**	DCOIT (RH-287 Technical)	
3.1.1	Lot/Batch number	[redacted]	
3.1.2	Specification	As given in section 2	
3.1.3	Purity	99.3%	
3.1.4	Composition of Product	[redacted]	x
3.1.5	Further relevant properties	[redacted]	
3.1.6	Method of analysis	[redacted]	
3.2	**Reference substance**	[redacted]	
3.2.1	Method of analysis for reference substance	[redacted]	
3.3	**Testing procedure**		
3.3.1	Soil sample /	[redacted]	

Section A7.5.1.1 Inhibition to microbial activity (terrestrial)

Annex Point IIA7.4

	inoculum / test organism		
3.3.2	Test system	[redacted]	
3.3.3	Application of TS	[redacted]	
3.3.4	Test conditions	see table A7.5.1.1/01-5	
3.3.5	Test parameter	carbon mineralization and nitrogen mineralization and transformation by soil microflora	
3.3.6	Analytical parameter	CO_2, nitrite, nitrate and ammonium measurements	
3.3.7	Duration of the test	28 days	
3.3.8	Sampling	days 0, 7 and 28 for respiration and nitrification	
3.3.9	Monitoring of TS concentration	[redacted]	
3.3.10	Controls	[redacted]	
3.3.11	Statistics	[redacted]	
		4 RESULTS	
4.1	**Range finding test**	Performed	
4.1.1	Concentration	0 (control), 1, 10, 100, 500, 1000 mg DCOIT/kg dry soil	
4.1.2	Effect data	nitrification data deviated from control: -19, 50, 66, 11, -91% respiration data deviated from control: -11, -33, -67, -78, -78%	x
4.2	**Results test substance**		
4.2.1	Initial concentrations of test substance	nitrification: 0 (control), 100, 200, 400, 600, 800, 1000 mg DCOIT/kg dry soil respiration: 0 (control), 2.1, 6.2, 19, 56, 167, 500 mg DCOIT/kg dry soil	
4.2.2	Actual concentrations of test substance	Not applicable	x
4.2.3	Growth curves	Not applicable	
4.2.4	Cell concentration data	Not applicable	
4.2.5	Concentration/ response curve	see Figure A7.5.1.1/01-1/5	
4.2.6	Effect data	The initial hourly CO_2 production rates on day 0 and day 28 during the first hours were 0.329 and 0.292 ml CO_2/100 g dry soil, which equals 13.6 and 12.2 mg carbon per 100 g dry soil, respectively. The microbial biomass was between 1.5 and 1.3% of the total soil organic carbon content (0.92% C_{org}) during the study.	x

		see tables A7.5.1.1/01-6 and A7.5.1.1/01-7	
4.2.7	Other observed effects	see tables A7.5.1.1/01-6 and A7.5.1.1/01-7	
4.3	**Results of controls**	see tables A7.5.1.1/01-6 and A7.5.1.1/01-7	
4.4	**Test with reference substance**	Performed: Dinoseb acetate	
4.4.1	Concentrations	33.3 mg ai/kg dry soil	
4.4.2	Results	Effects larger than 25% were found during the 28 day of the respiration part and the nitrification part of the study.	
		5 APPLICANT'S SUMMARY AND CONCLUSION	
5.1	**Materials and methods**	OECD 216 and OECD 217, Effects on soil microflora respiration transformation and nitrification transformation.	
5.2	**Results and discussion**		x
5.2.1	EC_{10}	respiration = 15.3 mg DCOIT/kg dry soil (C.I. 1.20-139) nitrification = 42.9 mg DCOIT/kg dry soil (C.I. 6.94-115)	
5.2.2	EC_{25}	respiration = 67.0 mg DCOIT/kg dry soil (C.I. 7.33-847) nitrification = 77.0 mg DCOIT/kg dry soil (C.I. 16.9-189)	
5.2.3	EC_{50}	respiration = 393* mg DCOIT/kg dry soil (C.I. 46.9-10070), nitrification = 155 mg DCOIT/kg dry soil (C.I. 46.5-363). * value calculated, however effect at 500 mg ai/kg was below 50% inhibition, therefore very broad confidence interval.	
5.3	**Conclusion**	Results of controls and reference substance are in acceptable range. The variations between the replicate control samples in this test were less than $\pm$ 15% for both the respiration rate and nitrate concentration at all sampling intervals, showing the validity of the study. In addition, the results of the reference study, where effects of dinoseb acetate of larger than 25% were found, showed that the methods used were appropriate.	x
5.3.1	Reliability	(1), reliable without restriction	x
5.3.2	Deficiencies	No	x

Evaluation by Competent Authorities	
	Evaluation by Rapporteur Member State
Date	13 September 2007
Materials and Methods	**Comment (3.1.4):** Carbon and nitrogen content of test substance is 47% and 5%, respectively. However, results indicate that DCOIT contribution to respiration or nitrate accumulation is negligible.
Results and discussion	**Comment (4.1.2):** Range finding results were partly misleading with regard to respiration (see comment 4.2.6). **Comment (4.2.2):** Due to the fact that DCOIT is rapidly degraded in soil it can be assumed that test substance concentrations declined during the test (see also comment 5.2). **Comment (4.2.6 and 5.2.3):** For respiration, the effect of the highest concentration was less than 50% in the definitive test, and was determined by extrapolation giving a very broad confidence interval. The upper confidence limit is certainly far beyond the "true" endpoint. Thus, the EC50 for respiration is not valid. However, the reported value of 393 mg/kg could be seen as a conservative estimate. It would be more correct to state that the EC50 (respiration) is > 500 mg/kg. The EC50 for nitrification can be considered valid.
Conclusion	**Comment (5.2):** Nitrification was completely inhibited at all concentrations at 7 days (Table A7.5.1.1/01-7). After 28 days activity was partly resumed in the lower concentration range. This is probably due to the fact that tetst substance concentrations were declining over the course of the test due to biodegradation of the test substance. **Comment (5.3):** Agree with applicant's version. However, the EC50 for soil respiration cannot be used, but the results of the study support the results gained from the nitrification inhibition study.
Reliability	Due to the restrictions noted above the reliability is changed from 1 to 2
Acceptability	Acceptable with the restrictions noted above
Remarks	The great discrepancy between the range finding (4.1) and definite study (Tables A7.5.1.1/01-6&7) may be explained according to the following consideration. Records of the range finding tests are not reported. The definitive study was conducted about 4 weeks after soil sampling. Thus, it must be assumed that the preliminary tests were performed on fresh soil samples. During the first week after soil sampling, microbial activity and community structure may change significantly. Toxicity testing should not be performed until basal respiration is stable, which often awaits one ore two weeks. This condition is probably not met by the range finding test. Immediately after soil disturbance, microbes are less protected and more exposed to chemicals compared to udisturbed soils. This may explain the more toxic response of the respiration range finding test. The results of this inhibition test is partly supported by the degradation studies (Document III-A/Section A7.2.1 Aerobic degradation in soil including extent and nature of bound residues) which stated that 5 ppm DCOIT appeared to partly inhibit microbial activity and degradation (pargraph 4.1 Preliminary studies)

Section A7.5.1.1	Inhibition to microbial activity (terrestrial)– **TABLES AND FIGURES**

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Table A7.5.1.1/01-5: Test conditions

Criteria	Details
Organic substrate	0.4 g Lucerne meal (containing 3.25% nitrogen) was added to each replicate of the nitrification part of the study
Incubation temperature	19.5 to 21.0 °C
Soil moisture	maintained at 45.5% (20.4 g water/100 g dry soil) of maximum water holding capacity
Method of soil incubation	Not described
Aeration	Not described

Table A7.5.1.1/01-6: Respiration rates

Test Substance Concentration (nominal) [mg DCOIT/kg dry soil]	Measured ($mg O_2$/kg dry soil/hour)			% difference to control		
	Day 0	Day 7	Day 28	Day 0	Day 7	Day 28
0 (control)	4.2	5.2	4.9	---	---	---
2.1	4.4	5.3	4.7	4	3	-3
6.2	3.4	5.2	4.7	-19	0	-3
19	3.6	4.9	4.2	-15	-6	-13
56	3.4	3.4	3.4	-19	-34	-30
167	1.9	2.8	2.6	-54	-47	-47
500	2.1	2.8	2.8	-50	-47	-43

Table A7.5.1.1/01-7: Nitrate Transformation Rates

Test Substance Concentration (nominal) [mg DCOIT/kg dry soil]	Measured ($mg NO_3^-$/kg dry soil/day)		% difference to control	
	Day 7	Day 28	Day 7	Day 28
0 (control)	12.1	8.3	---	---
100	-0.40	5.8	-103	-30
200	-2.4	3.2	-120	-62
400	-2.3	1.7	-119	-79
600	-0.35	0.31	-103	-96
800	1.0	0.57	-92	-93
1000	0.88	0.49	-93	-94

Figure A7.5.1.1/01-1: Glucose induced short term respiration

9. FIGURES

Figure 1. **Glucose induced short term respiration: Percent deviation of the RH-287 Technical treated samples from the control represented as dose-response track.**

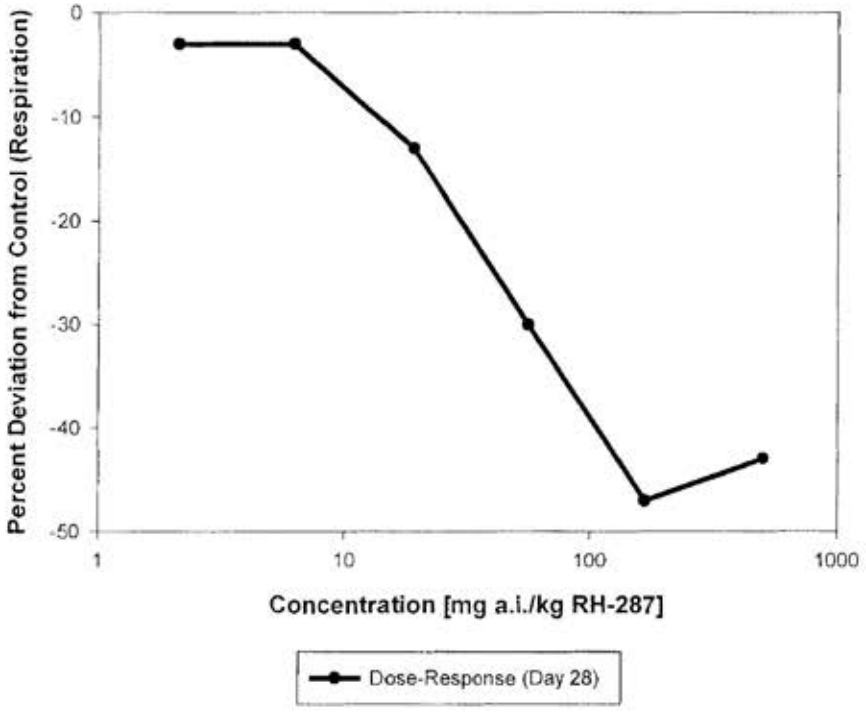

Figure A7.5.1.1/01-2: Representative ion chromatogram of the measurement of nitrate in a day 28 control sample

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Figure 2. Representative Ion Chromatogram of the measurement of nitrate in a day 28 control sample.

1007.079.747

212

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SAMPLE: day 28

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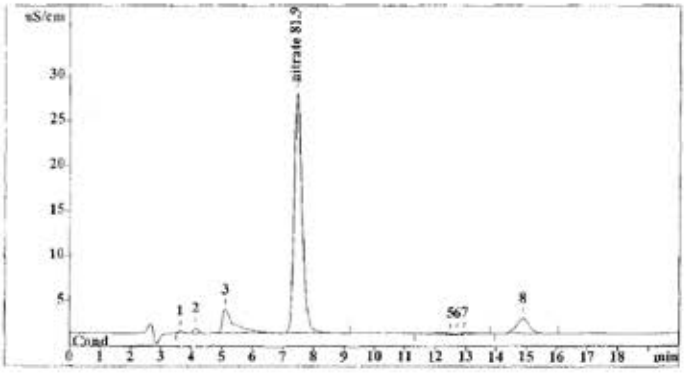
Volume: 20.0 µl
Dilution: 1.00
Amount: 1.0000



Quantitation method: Custom

No	Retention min	Height uS/cm	Area uS/cm*sec	Conc. mg/L	Name
4	7.44	26.73	478.092	81.900	nitrate

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Figure A7.5.1.1/01-3: Representative ion chromatogram of the measurement of nitrate in a day 28 treated sample at 100 mg DCOIT/kg

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Figure 3. **Representative Ion Chromatogram of the measurement of nitrate in a day 28 treated sample at 100 mg a.i. of RH-287 Technical/kg.**

1007.079.747

214

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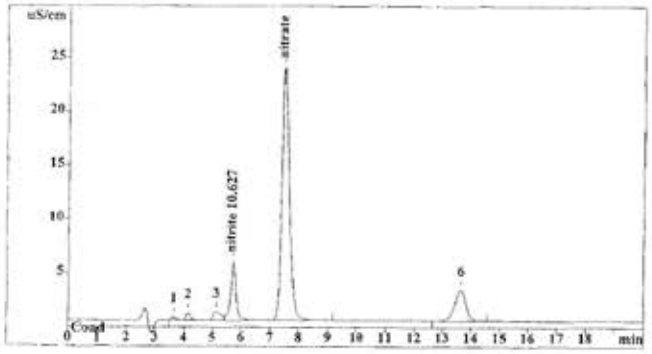
SAMPLE: day 28

Vial number: 18
Volume: 20.0 ul
Dilution: 1.00
Amount: 1.0000



Quantitation method: Custom

No	Retention min	Height uS/cm	Area uS/cm*sec	Conc. mg/L	Name
4	5.72	5.39	71.562	10.627	nitrite
5	7.47	23.47	422.114	74.337	nitrate
2	20.01	28.86	493.675	84.964	

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Figure A7.5.1.1/01-4: Representative ion chromatogram of the measurement of nitrate in a day 28 treated sample at 1000 mg DCOIT/kg

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Figure 4. Representative Ion Chromatogram of the measurement of nitrate in a day 28 treated sample at 1000 mg a.i. of RH-287 Technical/kg.

1007.079.747

229

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Printed by: guest

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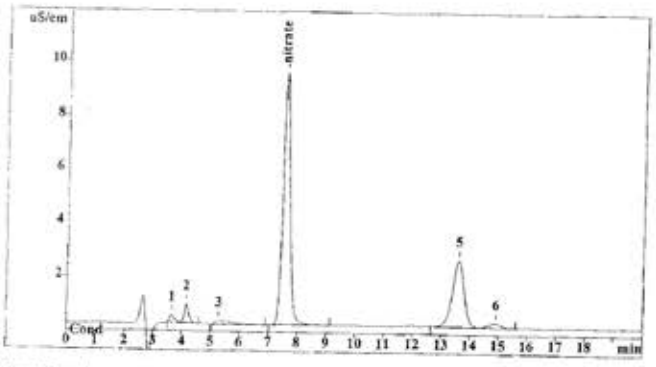
Run operator: guest
Analysis number: 729

SAMPLE: day 28

Vial number: 33
Volume: 20.0 µl
Dilution: 1.00
Amount: 1.0000



Quantitation method: Custom

No	Retention min	Height uS/cm	Area uS/cm*sec	Conc. mg/L	Name
4	7.54	9.37	157.164	30.857	nitrate

This report has been created by 761 Compact IC

Figure A7.5.1.1/01-5: Nitrate transformation

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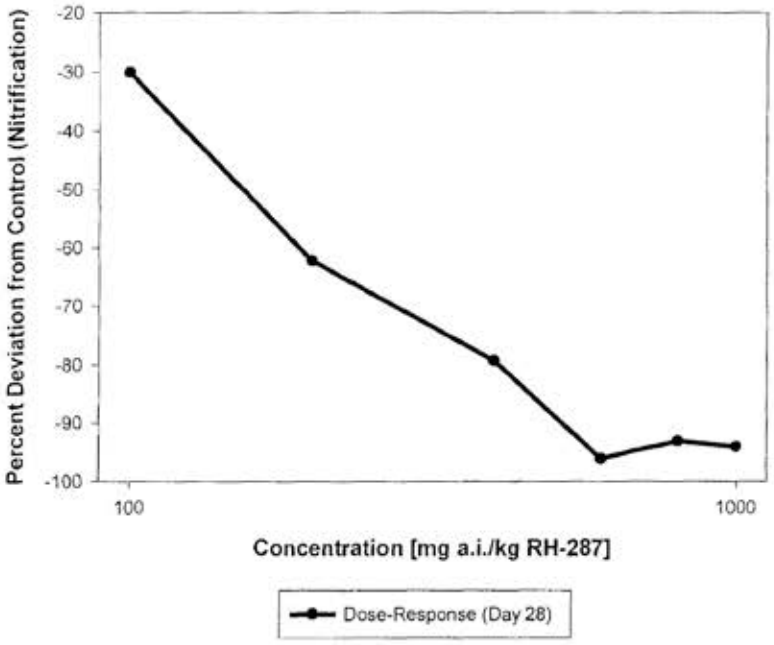
Figure 5. Nitrate transformation: Percent deviation of the RH-287 Technical treated samples from the control represented as dose-response.



Section A7.5.1.2 Earthworm, acute toxicity test
Annex Point IIIA XIII 3.2

Official use only

		1 REFERENCE
1.1	**Reference**	Reference Type: test repoprt Year: 2002 Report date: 28 February 2002 [redacted]
1.2	**Data protection**	Yes
1.2.1	Data owner	Rohm and Haas Company
1.2.2		
1.2.3	Criteria for data protection	[redacted]
		2 GUIDELINES AND QUALITY ASSURANCE
2.1	**Guideline study**	Yes, OECD Method 207
2.2	**GLP**	Yes
2.3	**Deviations**	No
		3 METHOD
3.1	**Test material**	DCOIT (RH-287 Technical)
3.1.1	Lot/Batch number	[redacted]
3.1.2	Specification	As given in section 2
3.1.3	Purity	99.3 %
3.1.4	Composition of Product	[redacted]
3.1.5	Further relevant properties	[redacted]
3.1.6	Method of analysis	[redacted]
3.2	**Reference substance**	[redacted]
3.3	**Testing procedure**	
3.3.1	Preparation of the test substance	[redacted]
3.3.2	Application of the	[redacted]

Section A7.5.1.2 Earthworm, acute toxicity test
Annex Point IIIA XIII 3.2

	test substance	[redacted]	
3.3.3	Test organisms	[redacted]	x
3.3.4	Test system	[redacted]	
3.3.5	Test conditions	see table A7.5.1.2/01-4	x
3.3.6	Test duration	14 days	
3.3.7	Test parameter	[redacted]	
3.3.8	Examination	[redacted]	
3.3.9	Monitoring of test substance concentration	[redacted]	
3.3.10	Statistics	[redacted]	

4 RESULTS

4.1	**Filter paper test**	Not performed
4.2	**Soil test**	
4.2.1	Initial concentrations of test substance	17, 33, 65, 130, 250 and 500 [mg DCOIT/kg wet weight artificial soil]
4.2.2	Effect data (Mortality)	see table A7.5.1.2/01-5
4.2.3	Concentration / effect curve	See Figure A7.5.1.2/01-1
4.2.4	Other effects	Not applicable
4.3	**Results of controls**	
4.3.1	Mortality	90% survival and no sublethal effects in control and acetone control, although one control worm was observed to be slightly lethargic on day 7.
4.3.2	Number/ percentage of earthworms showing adverse effects	see table A7.5.1.2/01-6

Section A7.5.1.2 Earthworm, acute toxicity test
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4.3.3	Nature of adverse effects	lethargy and increased time to burrow
4.4	**Test with reference substance**	Performed
4.4.1	Concentrations	2-chloracetamide
4.4.2	Results	LC_{50} = 17 mg/kg, wet weight
		5 APPLICANT'S SUMMARY AND CONCLUSION
5.1	**Materials and methods**	OECD Method 207, Acute toxicity to the earthworm
5.2	**Results and discussion**	All surviving worms exposed to the control, solvent control and 17, 33 and 65 mg DCOIT/kg burrowed into the soil within 10 minutes on days 0, 7 and 14. Worms exposed to 130, 250 and 500 mg DCOIT/kg required more than 30 minutes to burrow on day 0. Worms exposed to 250 mg DCOIT/kg required more than 30 minutes to burrow on day 7 and worms exposed to 500 mg DCOIT/kg, replicate 4, required more than 30 minutes to burrow on day 14.
5.2.1	NOEC	14 d NOEC = 130 mg DCOIT/kg (soil) based on survival and sublethal (behavioral) effects and 500 mg DCOIT/kg based on weight change data
5.2.2	LC_{50}	14 d LC_{50} = 250 mg DCOIT/kg (soil)
5.2.3	$LC_{0\ or}LC_{100}$	no concentration caused 0% or 100% mortality
5.3	**Conclusion**	see table A7.5.1.2/01-7 and see table A7.5.1.2/01-8
5.3.1	Other Conclusions	
5.3.2	Reliability	(1), reliable without restriction
5.3.3	Deficiencies	No

Evaluation by Competent Authorities	
	Evaluation by Rapporteur Member State
Date	23 August.2006
Materials and Methods	**Comment (3.3.3):** The age of the earthworms are not reported, only weight and sexual status (developed clitella). **Comment (3.3.5):** In Table A7.5.1.2701-4 it is stated that the temperature was 20 ± 2 °C. However, in the study report (XIII. Protocol deviations) it is stated that the temperature was not continuously recorded during the first 7 days of the definite toxicity test. During days 8 to 14 the continuously recorded temperature was not always 20 ± 2 °C (individually recorded daily temperatures in each vessel were always within the specified range). These deviations did not affect the outcome of the study according to the study report. It is not possible to control these deviations in temperature in the report, as all temperatures given in table A.1 are within the given range.
Results and discussion	Agree with applicant's version
Conclusion	Agree with applicant's version
Reliability	1, reliable without restrictions
Acceptability	Acceptable
Remarks	-

Section A7.5.1.2	Earthworm, acute toxicity test – **TABLES AND FIGURES**

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Table A7.5.1.2/01-4: Test conditions

Criteria	Details
Test temperature	20 ± 2 °C
Moisture content	The initial and final moisture content was approximately 26%.
pH	Day 0 = 5.6; Day 14 = 5.5
Adjustment of pH	Yes, pH was adjusted to 6.0 ± 0.5 by the addition of calcium carbonate.
Light intensity / photoperiod	approximately 400 to 800 lux, 24 h light and 0 h dark
Relevant degradation products	Not applicable

Table A7.5.1.2/01-5: Mortality data

Test Substance Concentration (nominal)[1] [mg DCOIT/kg artificial soil]	Mortality			
	Number Dead or Missing		Percentage	
	7 d	14 d	7 d	14 d
0 (control)	3	4	8	10
0 (solvent control)	4	4	10	10
17	5	5	13	13
33	7	8	18	20
65	4	5	10	13
130	6	6	15	15
250	12	20	30	50
500	23	26	58	65
Temperature [°C]	20.8-21.6	20.9-21.9		
pH	5.5-5.6	5.5-5.6		
Moisture content	26 %	26 %		

[1] specify, if TS concentrations were nominal or measured

Table A7.5.1.2/01-6: Number affected data

Test Substance Concentration (nominal)[1] [mg DCOIT/kg artificial soil]	Number Affected			
	Number affected		Percentage	
	7 d	14 d	7 d	14 d
0 (control)	1	0	3	--
0 (solvent control)	0	0	--	--
17	1	0	3	--
33	0	0	--	--
65	0	0	--	--
130	0	0	--	--
250	7	0	25	--
500	8	6	47	43
Temperature [°C]	20.8-21.6	20.9-21.9		
pH	5.5-5.6	5.5-5.6		
Moisture content	26 %	26 %		

[1] specify, if TS concentrations were nominal or measured

Table A7.5.1.2/01-7: Effect data

	14 d [mg/kg soil][1]	95 % c.l.
LC_0	not applicable	
LC_{50}	250 mg (n)	130 to >500 mg DCOIT/kg (n)
LC_{100}	not applicable	

[1] indicate if effect data are based on nominal (n) or measured (m) concentrations

Table A7.5.1.2/01-8: Validity criteria for acute earthworm test according to OECD 207

	fulfilled	Not fulfilled
Mortality of control animals < 10%	**yes**	

Figure A7.5.1.2/01-1: Survival of earthworms, *Eisenia foetida*, exposed to DCOIT for 14 days

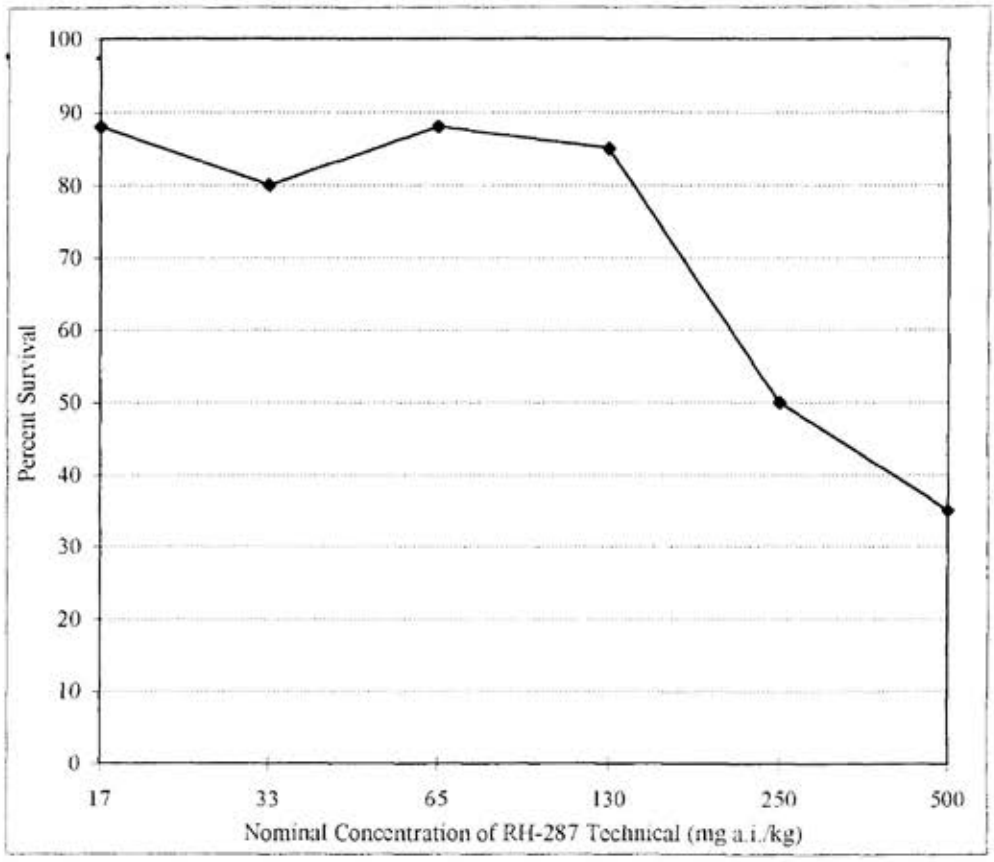

Figure 1. Survival of earthworms, *Eisenia foetida*, exposed to RH-287 Technical for 14 days.

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Section A7.5.1.3/01 Annex Point IIIA XIII 3.4 — Terrestrial plant toxicity – seedling emergence and growth

Official use only

1 REFERENCE

1.1	**Reference**	Reference Type: test repoprt Year: 2002 Report date: 25 November 2002 [redacted]
1.2	**Data protection**	Yes
1.2.1	Data owner	Rohm and Haas Company
1.2.2		
1.2.3	Criteria for data protection	[redacted]

2 GUIDELINES AND QUALITY ASSURANCE

2.1	**Guideline study**	Yes, OECD Draft Guideline 208, Part A and US EPA OPPTS Draft Guidelines 850.4100 and 850.4225
2.2	**GLP**	Yes
2.3	**Deviations**	No

3 METHOD

3.1	**Test material**	DCOIT (RH-287 technical)
3.1.1	Lot/Batch number	[redacted]
3.1.2	Specification	As given in section 2
3.1.3	Purity	99.3%
3.1.4	Composition of Product	[redacted]
3.1.5	Further relevant properties	[redacted]
3.1.6	Method of analysis	[redacted]
3.2	**Preparation of TS solution for poorly soluble or volatile test substances**	[redacted]
3.2.1	TS Concentrations	[redacted]

Section A7.5.1.3/01 Terrestrial plant toxicity – seedling emergence and growth
Annex Point IIIA XIII 3.4

		[redacted]	x
3.3	**Reference substance**	[redacted]	
3.3.1	Method of analysis for reference substance	[redacted]	
3.4	**Testing procedure**		
3.4.1	Dilution water	[redacted]	
3.4.2	Test plants	[redacted]	
3.4.3	Test system	[redacted]	
3.4.4	Test conditions	see table A7.5.1.3/01-5	
3.4.5	Test duration	25 days	
3.4.6	Test parameter	[redacted]	
3.4.7	Sampling	[redacted]	
3.4.8	Method of analysis of the plant material	[redacted]	
3.4.9	Quality control	[redacted]	
3.4.10	Statistics	[redacted]	x

4 RESULTS

4.1 Results test substance	
4.1.1 Applied initial concentration	not applicable

Section A7.5.1.3/01 **Terrestrial plant toxicity – seedling emergence and growth**
Annex Point IIIA XIII 3.4

4.1.2 Phytotoxicity rating	see table A7.5.1.3/01-6
4.1.3 Plant height	see table A7.5.1.3/01-6
4.1.4 Plant dry weights	see table A7.5.1.3/01-6
4.1.5 Root dry weights	Not applicable
4.1.6 Root length	Not applicable
4.1.7 Number of dead plants	see table A7.5.1.3/01-6
4.1.8 Effect data	see table A7.5.1.3/01-6
4.1.9 Concentration / response curve	Graph of the concentration-response curve at test termination not described in report
4.1.10 Percent emergence	see table A7.5.1.3/01-6
4.1.11 Other effects	Canola: necrosis, chlorosis, leaf curl, absence of flowers and dead plants were noted. Red clover: necrosis, chlorosis and dead plants were noted. Rice: necrosis, chlorosis, leaf curl, and dead plants were noted. see table A7.5.1.3/01-7
4.2 Results of controls	
4.2.1 Number/ percentage of plants showing adverse effects	Canola: no effects Rice: no effects Red clover: one dead plant in solvent control
4.2.2 Nature of adverse effects	Red clover: one dead plant in solvent control
4.3 Test with reference substance	Not performed
4.3.1 Concentrations	Not applicable
4.3.2 Results	Not applicable
	5 APPLICANT'S SUMMARY AND CONCLUSION
5.1 Materials and methods	OECD Draft Guideline 208, Part A and OPPTS Draft Guidelines 850.4100 and 850.4225, growth test in terrestrial plants with analytical confirmation of dosing solutions.
5.2 Results and discussion	
5.2.1 NOEC	see table A7.5.1.3/01-8
5.2.2 EC_{25}	see table A7.5.1.3/01-8
5.2.3 EC_{50}	see table A7.5.1.3/01-8
5.3 Conclusion	see table A7.5.1.3/01-8
5.3.1 Reliability	(1), reliable without restriction
5.3.2 Deficiencies	No

Evaluation by Competent Authorities	
	Evaluation by Rapporteur Member State
Date	30 August 2006
Materials and Methods	**Comment (3.2.1):** Only red clover was tested at 2.5 mg/kg dwt soil. In the tests with rice and canola 5 mg/kg dwt soil was the lowest concentration. **Comment (3.4.10):** If a NOEC value was established to be less than the lowest concentration tested, an EC10 could be calculated by linear regression, to provide a concervative NOEC. This is also done, for shoot length in rice. However, in part 2.9 Data Analysis in the study report it is stated that an EC05 should be used as a concervative NOEC in the above mentioned situations. An EC05 will normally have a wider 95 % confidence interval than EC10, so we suggest keeping EC10 as a NOEC. However, extrapolating outside the concentration range in linear regression is certainly not recommended. Moreover, the EC50 is the most relevant endpoint from this acute test.
Results and discussion	Agree with applicant's version
Conclusion	Agree with applicant's version
Reliability	1, valid without restrictions
Acceptability	Acceptable
Remarks	-

Section A7.5.1.3/01	Terrestrial plant toxicity – **TABLES AND FIGURES**

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