

FINAL REPORT

1886-BI-MAR-311/07

Biodegradability in Seawater EC9500A

In accordance with OECD-Guidelines for Testing of Chemicals. Method 306 "Biodegradability in Seawater"

Test substance: EC9500A

Number of test substance: 1886

Test item batch: BR3J0193B

Sponsor: Nalco Brasil Ltda

Rodovia Ìndio Tibiriçá, 3201 08675-000 - Suzano - SP - Brasil

Test Facility: Bioensaios Análises e Consultoria Ambiental Ltda.

Rua Palermo, 257 - Santa Isabel 94480-775 - Viamão - RS - Brasil

Test Facility Manager: Alexandre Brandelli

Study Director: Tiago Juliano Tasso de Souza

Quality Assurance Manager: Ellen Martha Pritsch

Study: 1886-BI-MAR-311-07

Title: Biodegradability in Seawater - EC9500A

Page 1 of 8 Version: English

Biodegradability in Seawater - EC9500A

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STATEMENT

Study: Biodegradability in Seawater - EC9500A

Study Number: 1886-BI-MAR-311-07

I declare that the study was been completed and the objectives established in the plan of study were successfully achieved, that the data generated are valid, and that the final report reflects the procedures used and the raw data.

I declare that the study was conducted in accordance with the principles of Good Laboratory Practice - INMETRO-NIT-DICLA-028 (Sep. 2003), based on OECD-Principles on Good Laboratory Practice (1997).

I declare that GLP principles were fully met.

Viamão, __10__ / _Feb._ / _2020 .

Everton Melo dos Santos Study Director current

Rua Palermo, 257 - Viamão - RS - Brazil

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QUALITY ASSURANCE STATEMENT

Study: Biodegradability in Seawater - EC9500A

Study Number: 1886-BI-MAR-311-07

I declare that the Final Report has been revised and reflects the Raw Data.

I declare that the Study Director signed the statement that the study was conducted under GLP principles on 01 July 2008.

I declare that audits were performed, as specified in the table below, with no deviations or non-conformities that could affect the quality of the results obtained.

Audit object	Audit date	Date reported to Study Director	Date reported to Test Facility Manager	
Study plan	19 Sep. 2007	19 Sep. 2007	19 Sep. 2007	
SOP	22 Nov. 2007	22 Nov. 2007	22 Nov. 2007	
Study phases				
Weighing the test oil	22 Nov. 2007	22 Nov. 2007	22 Nov. 2007	
Test: Temperature, preparation and agitation of the test flasks, phase separation	22 Nov. 2007	22 Nov. 2007	22 Nov. 2007	
Spectrophotometric readings of test and reference solutions	22 Nov. 2007	22 Nov. 2007	22 Nov. 2007	
Raw data	26 June 2008	01 July 2008	01 July 2008	
Final report	26 June 2008	01 July 2008	01 July 2008	

SOP: Standard Operating Procedures.

Viamão, 10 1 FEB. 1 2030 .

Rodrigo Garcia dos Santos Quality Assurance Sector current Rua Palermo, 257 - Viamão - RS - Brazil

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RESUMO

Um estudo de biodegradabilidade com 28 dias de duração foi conduzido em laboratório para determinar a biodegradação do composto EC9500A sob condições aeróbicas e em água do mar.

O estudo foi conduzido de acordo com o OECD - Guidelines for Testing of Chemicals Method 306 "Biodegradability in Seawater". Uma suspensão da substância-teste em água do mar foi inoculada e incubada no escuro durante 28 dias. Um padrão biodegradável (hidrogênio ftalato de potássio) e um controle da atividade endógena do inóculo (branco do inóculo) foram executados em paralelo para verificar os procedimentos de operação. Os efeitos inibitórios da substância-teste sobre o inóculo foram testados através de uma solução contendo substância-teste e substância de referência (controle de toxicidade). A degradação foi acompanhada pela determinação do oxigênio dissolvido em diversos intervalos de tempo.

Os ensaios com o padrão biodegradável e o controle de toxicidade resultaram em respectivamente 80,5% e 200,5% da demanda teórica após 14 dias. Estes resultados comprovam a atividade do inóculo utilizado e também demonstram que a substânciateste não apresentou ação inibitória sobre os microorganismos. A demanda de oxigênio a partir da substância-teste foi de 96,8% do total teórico após 14 dias. Baseado neste resultado o EC9500A pode ser classificado como um composto prontamente biodegradável.

SUMMARY

A 28 days laboratory biodegradability study was conducted to determine the ready biodegradation of EC9500A, under aerobic condition in seawater.

This study was conducted according the OECD-Guidelines for Testing of Chemicals Method 306 "Biodegradability in Seawater".

A suspension of the test substance in sea water was inoculated and incubated in the dark for 28 days. A reference compound (potassium hydrogen phtalate) and a control of the endogenous activity of the inoculum (Blank inoculum) was run parallel to check the operation of the procedures. Also, the possible inhibitory effect of the test substance was checked by adding both test substance and reference substance (toxicity control). The degradation was followed by the determination of dissolved oxygen at frequent time intervals.

The results from reference compound and toxicity control were respectively 80.5% and 200.5% of the maximum theoretical demand in 14 days. These result prove the activity of the inoculum and that the test substance do not had inhibitory effects on microorganisms. For the test-substance, the results shown a 96.8% of the maximum theoretical demand in 14 days. EC9500A can be classified as a compound liable to ready biodegradation.

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1. INTRODUCTION

Biodegradability in Seawater expresses the ability of an organic substance to be metabolized by natural microorganisms in the marine environment. This study provides very limited conditions for biodegradability and acclimatization to occur, so it can be assumed that a chemical that shows a positive result will biodegrade rapidly in the marine environment.

This report presents the result of the Biodegradability in Seawater study of test substance EC9500A.

1.1. Study Dates

Study plan : 11 Oct. 2007 Experiment start : 08 Feb. 2008 Experiment end : 07 Mar. 2008 Final report : 01 July 2008

2. MATERIAL AND METHODS

2.1. Test substance

Name of the test substance : EC9500A

Chemical name of a.i. : Fatty esters of ethoxylated sorbitan10.0 – 30.0 %

Common name of the active

ingredient

: Mixture of ester surfactants, solvents and dipropylene-glycol-monobutyl-ether

Test substance batch : BR3J0193B

Number of test substance : 1886

Expiration of Test substance : 21 Dec. 2008 Sample opening date : 17 Oct. 2007

Stability : Stable for approximately 32 months at room temperature

Homogeneity : Visually homogeneous

2.2. Reference item

Chemical name : Hydrogen potassium phthalate Brand : Cromato Produtos químicos

Batch : 1866.09/06 Purity : 99.8%

Expiration date : 30 Sep. 2009

2.3. Seawater

Collection date : 24 Jan. 2008
Collection depth : Surface
Appearance : Clear
Collection temperature : 20 °C
Salinity : 31 g / L
Bacterial count : 360 CFU / mL

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2.4. Methodology

OECD - Method 306 (1992).

The summary of the test conditions is shown below.

Inoculum	Bacteria from sea water
Test chamber	300 mL BOD bottles
Aeration system	Closed bottle
Luminosity	Absence of light
Temperature	
Exposure Duration	28 days
Answer	Dissolved oxygen consumption
O ₂ measurement system	Oximeter

2.4.1. Half-test

The mineral medium used for dilution is prepared by adding 1 mL of each of the nutrient solutions described below to each 1 L of sea water and aerated for 20 minutes before the test starts:

- pH 7.2 phosphate nutrient solution:

KH₂PO₄ 8.50 g / L K₂HPO₄ 21.75 g / L Na₂HPO₄•2H₂O 33.40 g / L NH₄CI 0.50 g / L

- Magnesium sulfate nutrient solution:

 $MgSO_4$ •7 H_2O 22.50 g / L - Ferric chloride nutrient solution: $FeCl_3$ •6 H_2O 0.25 g / L - Calcium chloride nutrient solution:

 $CaCl_2 \cdot 2H_2O$ 36.30 g / L

2.4.2. Test organisms

The source of the test organisms is sea water, which already has the necessary inoculum to perform the test.

2.4.3. Preparation of Test Solutions

The test is carried out with the following test concentrations:

Treatment A - Inoculum white: half-test

Treatment B - Control procedure: potassium hydrogen phthalate

Concentration: 7.0 mg / L **Theoretical oxygen demand**: 7.9 mg O_2 / L

Treatment C - Test suspension: Test substance

Concentration: 3.4 mg / L EC9500A / L

Theoretical oxygen demand: 6.8 mg O₂ / L

Treatment D - Toxicity control: hydrogen potassium phthalate and test substance.

Concentration: 3.5 mg / L of hydrogen potassium phthalate

3.2 mg / L EC9500A

Theoretical oxygen demand: 6.5 mg O₂ / L (referring to potassium

phthalate hydrogen)

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2.4.3. Procedure

The test solutions are siphoned into 300 mL BOD bottles and kept at 20 \pm 1° C in the absence of light. Dissolved oxygen readings are taken every 7 days in two vials for treatments A, B and C and 1 vial for treatment D.

3. RESULTS

The percentage of biodegradation for each time period is calculated using the expression:

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 $D_t = \frac{B_t - C_t}{DO_t} x 100$

Where:

% Dt - Percentage of degradation at time t (days)

 B_t - Dissolved oxygen concentration (mg O_2/L) of the blank test at time t (days)

 C_t - Dissolved oxygen concentration (mg O_2 / L) of the test with standard substance or test at time t (days)

DO_t - Theoretical oxygen demand (mg O₂ / L) of the substance tested.

To be considered biodegradable, the test substance and the reference substance must achieve 60% degradation of the theoretical oxygen demand (DOt) in a short time and not exceeding a total of 28 days.

The toxicity control shows the occurrence or not of inhibitory effects.

The test results are presented in Annex I.

4. CONCLUSION

According to the results obtained, test substance EC9500A can be classified as **readily biodegradable**, as it reached a degree of degradation of 96.8% in 14 days.

5. ARCHIVES

The Study Plan, Raw Data and Final Report were archived for, at least, the next five years and the test item for, at least, 60 days after the conclusion of all studies at Bioensaios Análises e Consultoria Ambiental Ltda.

6. REFERENCES

OECD Guideline for Testing of Chemicals (1992). Method 306 "Biodegradability in Seawater".

Viamão, 10 / Feb. / 2020 .

Everton Melo dos Santos Study Director current

Rua Palermo, 257 - Viamão - RS - Brazil

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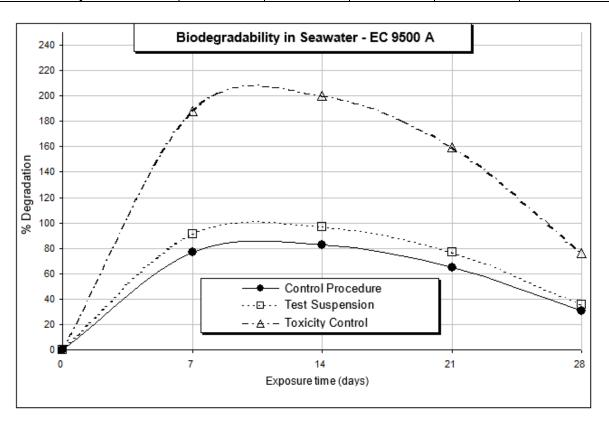
ANNEX I

Test Results Determinations of Dissolved Oxygen (mg O₂ / L)

		Exposure time						
Treatment		Day 0	Day 7	Day 14	Day 20	Day 28		
		08 Feb.2008	15 Feb.2008	22 Feb.2008	29 Feb.2008	07 March 2008		
	A1	7.08	6.44	6.89	5.68	3.25		
Inoculum White	A2	7.07	6.38	6.83	5.32	1.78		
	Mean	7.08	6.41	6.86	5.50	2.52		
Control	B1	7.00	0.29	0.30	0.34	0.09		
Control Procedure	B2	7.12	0.25	0.26	0.32	0.08		
Procedure	Mean	7.08	0.27	0.28	0.33	0.09		
	C1	7.04	0.26	0.28	0.28	0.10		
Test Suspension	C2	7.03	0.20	0.28	0.29	0.06		
	Mean	7.08	0.23	0.28	0.29	0.08		
Toxicity Control	D	7.18	0.26	0.30	0.29	0.09		

Percentage of Degradation (%)

: oroomago or 2 ogradadion (70)							
	Exposure time						
Treatment	Day 0	Day 7	Day 14	Day 20	Day 28		
	08 Feb.2008	15 Feb.2008	22 Feb.2008	29 Feb.2008	07 March 2008		
Control Procedure	0.0	76.9	82.5	64.8	30.5		
Test Suspension	0.0	90.9	96.8	76.7	35.8		
Toxicity Control	0.0	188.1	200.5	159.9	76.1		





Amendment No. 001 to the Final Report 1886-BIMAR-311-07 "Biodegradability in Seawater - EC9500A" Study: 1886-BIMAR-311-07 of 01 July 2008

CHANGE 01: Item "2. MATERIAL AND METHODS"

Previous to the change: Theoretical Oxygen Demands (DTO) of the test substance and

reference substance were not provided

After the change:

2.1. Test substance

Name of the test substance : EC9500A

Chemical name of a.i. : Fatty esters of ethoxylated sorbitan10.0 – 30.0 %

Common name of the active: Mixture of ester surfactants, solvents and

ingredient dipropylene-glycol-monobutyl-ether

Test substance batch : BR3J0193B

Number of test substance : 1886

Expiration of Test substance : 21 Dec. 2008 Sample opening date : 17 Oct. 2007

Stability : Stable for approximately 32 months at room temperature

Homogeneity : Visually homogeneous

Theoretical oxygen demand : 1.99 mg O₂ / mg (obtained from the sample COD)

2.2. Reference item

Chemical name : Hydrogen potassium phthalate Brand : Cromato Produtos químicos

Batch : 1866.09/06 Purity : 99,8%

Expiration date : 30 Sep. 2009 Theoretical oxygen demand : 1.136 mg O₂ / mg

Reason for change: Inform a fundamental parameter for calculating the results and

concluding the study.



CHANGE 02: Item "2.4.3. Preparation of Test Solutions" Previous to the change:

Treatment D - Toxicity control:

hydrogen potassium phthalate and test substance.

Concentration:

3.5 mg/L of hydrogen potassium phthalate

3.2 mg/L EC9500A

Theoretical oxygen demand: 6.5 mg O₂/L (referring to potassium

phthalate hydrogen)

After the change:

Treatment D - Toxicity control:

hydrogen potassium phthalate and test substance.

Concentration:

3.5 mg/L of hydrogen potassium phthalate

3.2 mg/L EC9500A

Theoretical oxygen demand: 4.0 mg O₂ / L (referring to potassium

phthalate hydrogen)

Reason for change:

Correct the potassium hydrogen phthalate concentration value and

the theoretical oxygen demand value.

CHANGE 03: Item "ANNEX I - Test Results".

Previous to the change: Annex I presented two tables with the results of dissolved oxygen and

the results in percentage of degradation.

After the change:

A third table has been added with the results of specific oxygen

demand (see below).

Reason for change:

Provide partial data of the biodegradability calculation step

GLP COMPLIANCE STATEMENT

QUALITY ASSURANCE STATEMENT

I declare that the Amendment to the Final Report reflects the Raw Data obtained. I declare that the Study for the elaboration of the Amendment was conducted in accordance with the principles of Good Laboratory Practice - INMETRO-NIT-DICLA-028 (Sep. 2003), based on OECD-Principles on Good Laboratory Practice (1997).

I declare that the Amendment to the Final Report has been revised and reflects the Raw Data and no deviations or nonconformities were observed that could affect the quality of the results.

Everton Melo dos Santos Study Director current

Rodrigo Garcia dos Santos Quality Assurance Sector current



ANNEX I Test Results

Determinations of Dissolved Oxygen (mg O₂/L)

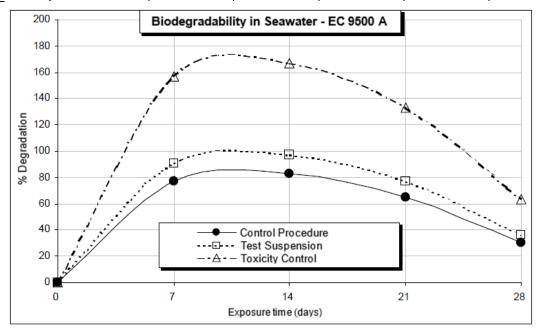
		Exposure time					
Treatment	Treatment		Day 7	Day 14	Day 20	Day 28	
		08 Feb.2008	15 Feb.2008	22 Feb.2008	29 Feb.2008	07 March 2008	
	A1	7.08	6.44	6.89	5.68	3.25	
Inoculum White	A2	7.07	6.38	6.83	5.32	1.78	
	Mean	7.08	6.41	6.86	5.50	2.52	
Control	B1	7.00	0.29	0.30	0.34	0.09	
Control Procedure	B2	7.12	0.25	0.26	0.32	0.08	
Procedure	Mean	7.06	0.27	0.28	0.33	0.09	
	C1	7.04	0.26	0.28	0.28	0.10	
Test Suspension	C2	7.03	0.20	0.28	0.29	0.06	
·	Mean	7.04	0.23	0.28	0.29	0.08	
Toxicity Control	D	7.18	0.26	0.30	0.29	0.09	

Biological oxygen demand (mg O₂/L)

	Exposure time					
Treatment	Day 0	Day 7	Day 14	Day 20	Day 28	
	08 Feb.2008	15 Feb.2008	22 Feb.2008	29 Feb.2008	07 March 2008	
Control Procedure	0.00	6.14	6.58	5.17	2.43	
Test Suspension	0.00	6.18	6.58	5.22	2.44	
Toxicity Control	-0.03	6.26	6.67	5.32	2.53	

Percentage of Degradation (%)

· or contage or a cylindrical (70)							
	Exposure time						
Treatment	Day 0	Day 7	Day 14	Day 20	Day 28		
	08 Feb.2008	15 Feb.2008	22 Feb.2008	29 Feb.2008	07 March 2008		
Control Procedure	0.0	76.9	82.5	64.8	30.5		
Test Suspension	0.0	90.9	96.8	76.7	35.8		
Toxicity Control	0.0	156.8	167.1	133.2	63.4		





Amendment No. 002 to the Final Report 1886-BI-MAR-311-07 "Biodegradability in Seawater - EC9500A" Study: 1886-BI-MAR-311-07 of 01 July 2008

CHANGE 01: Item "STATEMENT"

Previous to the change:

Study: Biodegradability in Seawater - EC9500A

Study Number: 1886-BI-MAR-311-07

I declare that the study was been completed and the objectives established in the plan of study were successfully achieved, that the data generated are valid, and that the final report reflects the procedures used and the raw data.

I declare that the study was conducted in accordance with the principles of Good Laboratory Practice - INMETRO-NIT-DICLA-028 (Sep. 2003), based on OECD-Principles on Good Laboratory Practice (1997).

I declare that GLP principles were fully met.

After the change:

Study: Biodegradability in Seawater - EC9500A

Study Number: 1886-BI-MAR-311-07

I declare that the study was been completed and the objectives established in the plan of study were successfully achieved, that the data generated are valid, and that the final report reflects the procedures used and the raw data.

I declare that the study was conducted in accordance with the principles of Good Laboratory Practice - INMETRO-NIT-DICLA-028 (Sep. 2003), based on OECD-Principles on Good Laboratory Practice (1997).

I declare that the GLP principles were partially complied with, as a test substance analysis certificate was not made available, not complying with item 6.2.2 of NIT-DICLA-028.

Reason for change: Correctly inform the full compliance with the GLP principles in the Study.

GLP COMPLIANCE STATEMENT

I declare that the Amendment to the Final Report reflects the Raw Data obtained. I declare that the Study for the elaboration of the Amendment was conducted in accordance with the principles of Good Laboratory Practice - INMETRO-NIT-DICLA-034 (Sep. 2003) and INMETRO-NIT-DICLA-035 to 041 (Dec. 2007), based on OECD-Principles on Good Laboratory Practice (1997).

QUALITY ASSURANCE STATEMENT

I declare that the Amendment to the Final Report has been revised and reflects the Raw Data and no deviations or nonconformities were observed that could affect the quality of the results.

Everton Melo dos Santos Study Director current

Date

Rodrigo Garcia dos Santos Quality Assurance Sector current

Date



Amendment No. 003 to the Final Report 1886-BIMAR-311-07 "Biodegradability in Seawater - EC9500A" Study: 1886-BIMAR-311-07 of 01 July 2008

CHANGE 01: This amendment renders without effect the amendment No. 001 to the Final Report 1866-BIMAR-311-07

CHANGE 02: Item "2. MATERIAL AND METHODS"

Previous to the change: Theoretical Oxygen Demands (DTO) of the test substance and

reference substance were not provided

After the change:

2.1. Test substance

Name of the test substance : EC9500A

Chemical name of a.i. : Fatty esters of ethoxylated sorbitan10.0 – 30.0 %

Common name of the active: Mixture of ester surfactants, solvents and

ingredient dipropylene-glycol-monobutyl-ether

Test substance batch : BR3J0193B

Number of test substance : 1886

Expiration of Test substance : 21 Dec. 2008 Sample opening date : 17 Oct. 2007

Stability : Stable for approximately 32 months at room temperature

Homogeneity : Visually homogeneous

Theoretical oxygen demand : 1.99 mg O₂ / mg (obtained from the sample DQO)

2.2. Reference item

Chemical name : Hydrogen potassium phthalate Brand : Cromato Produtos químicos

 Batch
 : 1866.09/06

 Purity
 : 99.8%

 Expiration date
 : 30 Sep. 2009

 Theoretical oxygen demand
 : 1.136 mg O₂ / mg

Reason for change: Inform a fundamental parameter for calculating the results and

concluding the study.



CHANGE 03: Item "2.4.3. Preparation of Test Solutions"

Previous to the change:

Treatment B - Control procedure: potassium hydrogen phthalate

Concentration: 7.0 mg / L Theoretical oxygen demand: 7.9 mg O_2 / L

Treatment C - Test suspension: Test substance

Concentration: 3.4 mg / L EC9500A / L

Theoretical oxygen demand: $6.8 \text{ mg O}_2 / \text{L}$

Treatment D - <u>Toxicity control:</u> hydrogen potassium phthalate and test substance.

Concentration: 3.5 mg / L of hydrogen potassium phthalate

3.2 mg / L EC9500A

Theoretical oxygen demand: 6.5 mg O₂ / L (referring to potassium

phthalate hydrogen)

After the change:

Treatment B - Control procedure: potassium hydrogen phthalate

Treatment C - Test suspension: Test substance

Concentration: 3.4 mg / L EC9500A / L

Theoretical oxygen demand: $6.8 \text{ mg O}_2 / \text{L}$

Treatment D - <u>Toxicity control:</u> hydrogen potassium phthalate and test substance.

Concentration: 2.1 mg / L of hydrogen potassium phthalate

2.0 mg / L EC9500A

Theoretical oxygen demand: 6.5 mg O₂ / L (referring to potassium

phthalate hydrogen)

Reason for change: Correct the value of the theoretical oxygen demand for potassium

hydrogen phthalate in the control procedure (rounding error) and the value of the concentrations of potassium phthalate hydrogen and test

substance in the toxicity control (typing error).

CHANGE 04: Item "ANNEX I - Test Results".

Previous to the change: Annex I presented two tables with the results of dissolved oxygen and

the results in percentage of degradation.

After the change: Tables were added with the results of biochemical oxygen demands,

specific oxygen demands, and expected oxygen demands for the control of toxicity according to the biochemical oxygen demand of the

test substance and the reference substance (see Annex I).

Reason for change: Provide partial data of the biodegradability calculation step

Amendment No. 003 to the Final Report 1886-BIMAR-311-07 Biodegradability in Seawater - EC9500A Page 3 of 6



GLP COMPLIANCE STATEMENT

I declare that the Amendment to the Final Report reflects the Raw Data obtained. I declare that the Study for the elaboration of the Amendment was conducted in accordance with the principles of Good Laboratory Practice - INMETRO-NIT-DICLA-028 (Sep. 2003), based on OECD-Principles on Good Laboratory Practice (1997).

QUALITY ASSURANCE STATEMENT

I declare that the Amendment to the Final Report has been revised and reflects the Raw Data and no deviations or nonconformities were observed that could affect the quality of the results.

Everton Melo dos Santos Study Director current

Date

Rodrigo Garcia dos Santos Quality Assurance Sector current

Date



ANNEX I Test Results

Determinations of Dissolved Oxygen (mg O₂ / L)

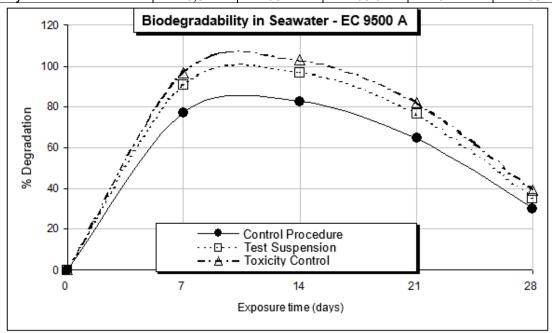
				, , , , , , , , , , , , , , , , , , , 				
		Exposure time						
Treatment		Day 0	Day 7	Day 14	Day 20	Day 28		
		08 Feb.2008	15 Feb.2008	22 Feb.2008	29 Feb.2008	07 March 2008		
	A1	7.08	6.44	6.89	5.68	3.25		
Inoculum White	A2	7.07	6.38	6.83	5.32	1.78		
	Mean	7.08	6.41	6.86	5.50	2.52		
Control	B1	7.00	0.29	0.30	0.34	0.09		
Control	B2	7.12	0.25	0.26	0.32	0.08		
Procedure	Mean	7.06	0.27	0.28	0.33	0.09		
	C1	7.04	0.26	0.28	0.28	0.10		
Test Suspension	C2	7.03	0.20	0.28	0.29	0.06		
	Mean	7.04	0.23	0.28	0.29	0.08		
Toxicity Control	D	7.18	0.26	0.30	0.29	0.09		

Biological oxygen demand (mg O₂/L)

Diological exygen demand (mg 02/ 2)								
Treatment		Exposure time						
	Day 0	Day 7	Day 14	Day 20	Day 28			
	08 Feb.2008	15 Feb.2008	22 Feb.2008	29 Feb.2008	07 March 2008			
Control Procedure	0.00	6.13	6.57	5.16	2.42			
Test Suspension	0.00	6.14	6.54	5.18	2.40			
Toxicity Control	0.00	6.26	6.67	5.32	2.53			

Percentage of Degradation (%)

referringe of Degradation (70)								
Treatment		Exposure time						
	Day 0	Day 7	Day 14	Day 20	Day 28			
	08 Feb.2008	15 Feb.2008	22 Feb.2008	29 Feb.2008	07 March 2008			
Control Procedure	0,0	76.9	82.4	64.7	30.3			
Test Suspension	0,0	90.7	96.7	76.5	35.4			
Toxicity Control	0.0	96.7	103.0	82.2	39.1			





Specific Biochemical Oxygen Demand (mg O₂ / mg):

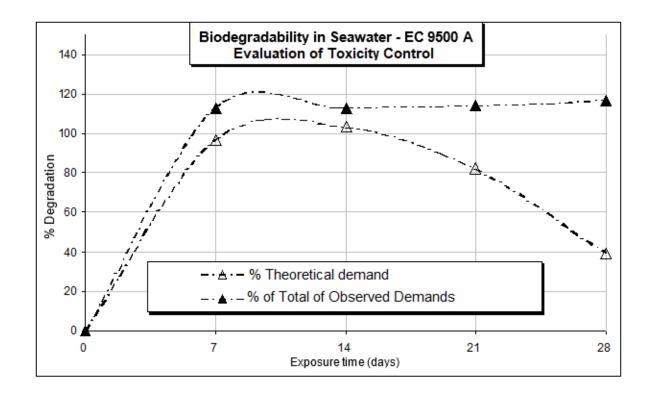
	Exposure time						
Treatment	Day 0	Day 7	Day 14	Day 20	Day 28		
	08 Feb.2008	15 Feb.2008	22 Feb.2008	29 Feb.2008	07 March 2008		
Reference Substance	0.00	0.87	0.94	0.74	0.34		
Test Substance	0.00	1.81	1.92	1.52	0.70		

Observed Biochemical Demand (DBO_o) for Toxicity control depending on the demands observed in the separate solutions (mg O₂ / L):

	Exposure time						
Treatment	Day 0	Day 7	Day 14	Day 20	Day 28		
	08 Feb.2008	15 Feb.2008	22 Feb.2008	29 Feb.2008	07 March 2008		
Reference Substance	0.0	1.9	2.0	1.6	0.7		
Test Substance	0.0	3.7	3.9	3.1	1.4		
Total	0.0	5.5	5.9	4.7	2.2		

Degradation observed in Toxicity Control due to Observed Biochemical Oxygen Demand (DBOo) and Theoretical Oxygen Demand (%)

	Exposure time				
Treatment	Day 0	Day 7	Day 14	Day 20	Day 28
	08 Feb.2008	15 Feb.2008	22 Feb.2008	29 Feb.2008	07 March 2008
DTO Control	6.5	6.5	6.5	6.5	6.5
DBO₀ Control	0.0	5.5	5.9	4.7	2.2
DBO	0.00	6.26	6.67	5.32	2.53
%DTO	0.0	96.7	103.0	82.2	39.1
%DBO。	0.0	113.0	112.8	114.0	116.7



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Thus, both in relation to the sum of the DTO of the test and reference substances present in the toxicity control, and in relation to the sum of the DBO observed for the test and reference substances separately, no toxicity of the test substance was observed, as this reached up to 102.9% degradation in relation to DTO and up to 116.7% degradation in relation to the sum of DBO.