

ACTIVATED SLUDGE RESPIRATION INHIBITION TEST

TEST SUBSTANCE

Identity: A mixture containing perfluorooctanesulfonate, which may also be referred to as PFOS, FC-95, or as a component of FC-203CE. (1-Octanesulfonic acid) (CAS # 2795-39-3).

Remarks: The 3M production lot number was 519. The test sample is FC-203CE (2/21/89). Current information indicates it is a mixture of 1.84% PFOS, 30% diethylene glycol butyl ether, 60.23% water, 2.56% Sultone foamer, 3.1% sodium octyl sulfate, 2.1% polyoxyethylene monooctylphenyl ether, 0.12% sodium lauryl sulfate, and 0.05% tolyltriazole. Antifoamer SWS 214 was added at a nominal concentration of 200 mg/L.

The following summary applies to a mixture with incompletely characterized concentrations of impurities. Data may not accurately reflect the toxicity of the fluorochemical component of the test sample.

METHOD

Method: OECD 209

GLP: No

Year Completed: 1990

Analytical monitoring: Dissolved oxygen concentrations, pH and temperature.

Statistical methods: Results were determined by calculation of the % inhibition in respiration rate.

Test organism source: Activated sludge mixed liquor collected from the Metro Waste Treatment Plant, St. Paul, MN

Test condition:

Dilution water: Millipore Milli-Q™ water

Synthetic sewage: per OECD method

Reference and test solution preparation: A stock solution of the reference substance, 3,5-dichlorophenol, was prepared by dissolving 500 mg in 10mL 1N NaOH, diluted to 30mL with Millipore Milli-Q™ water, then brought to the point of incipient precipitation with 1N H₂SO₄ and diluted to 1L with Milli-Q™ water. The pH of the reference substance was measured to be 7.4. Test substance was added directly to the test vessels. Antifoamer SWS 214 was added to the test vessels at 200 mg/L from a stock solution prepared by diluting 2000 mg of SWS 214 in 500 mL Milli-Q™ water. The test substance and antifoamer appeared miscible in water.

Test vessels: Not given

Number of concentrations: three plus 1 reference substance and 2 blank controls

Temperature: 20 – 22 °C

Total suspended solids and pH on day of testing: 5.033 g/L and initial pH 6.6 and final pH 6.9, Initial TSS of 1000 mg/L concentration was 2.027 g/L, pH 6.6 while final TSS of 1000 mg/L concentration was 2.213 g/L, pH 6.8.

Element Basis: Respiration inhibition as determined by oxygen consumption.

Remarks: Values corrected to 20 °C for calculations.

RESULTS

Nominal concentrations: 2 Blank controls, reference substance, 320, 560 and 1000 mg/L

Element values: 30-minute EC_{50} = >1000 mg/L
3-hour EC_{50} = >1000 mg/L

Element values based on nominal concentrations.

Remarks: There was a slight reduction in the amount of foam formed in the 1000 mg/L exposure concentration when compared to FC-203CE without antifoamer.

Testing was conducted on a mixture as described in the Test Substance Remarks field. The values reported apply to that mixture and not the fluorochemical proportion alone.

CONCLUSIONS

The test substance 3-hour EC_{50} for activated sludge respiration inhibition was determined to be >1000 mg/L.

Submitter: 3M Company, Environmental Laboratory, P.O. Box 33331, St. Paul, Minnesota, 55133

DATA QUALITY

Reliability: Klimisch ranking 2. Testing meets all criteria for quality testing. However, sample purity was not properly characterized and the study lacks analytical confirmation of the amount of fluorochemical proportion in the solution.

REFERENCES

This study was conducted by the 3M Company, Environmental Laboratory, St Paul, MN, Lab Request number H1480-3, 1990.

OTHER

Last changed: 6/26/00

LR H1480-3

Date 3/19/90

Analyst SJ

ENVIRONMENTAL LABORATORY
ACTIVATED SLUDGE MIXED LIQUOR

Type Test Activated Sludge Respiration Inhibition Test OECD #209

Source Metrom Waste Treatment Plant, St. Paul, Mn.

Date Obtained 3/19/90 Date Used 3/19/90

Mixed Liquor Suspended Solids (MLSS) 5.033 g/L Adjustment g/L

pH initial 6.6, final 6.9

Comments _____

Test Substance

Sample Description MC203 CE, Lot 519 (2/21/89) with Antifoamer SWS 214 (200 mg/L)
highest concentration tested, 1000 mg/L

Sample pH initial 6.6, final 6.8 Adjustment _____

Comments MLSS of 1000 mg/L test soln; initial 2.027 g/L, final (3 Hr) 2.213 g/L

Reference Substance

Sample Description 3,5-Dichlorophenol
highest concentration tested, 10 mg/L

Sample pH initial 6.7, final 6.8 Adjustment _____

Comments Stock Soln. pH 7.4

Exposure Conditions

Test Duration 3 Hours

Test Temperature 20-22 °C

Comments _____

Lab Request No. H1480-3

Environmental Lab — Work Sheet

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Sample Description

Date: 3/19/90OECD ACTIVATED SLUDGE
RESPIRATION INHIBITION TEST #209Analyst: [Signature]

Test Solutions Preparation

FC203 CE, Lot. 519 (2/21/90) with Antifoamer SWS 214 (200 mg/L). Individual weights used.
 SWS 214 Antifoamer at 200 mg/L dosage. SWS 214 Stock solution: 2000 mg/
 500 mL Milli-Q water. Each 1 mL = 4 mg. (SWS 214 very miscible) Stock pH 6.7.
 Stock solution

For 200 mg/L = 100 mg/500 mL. Used 25 mL/500 mL.

Test Conc. mg/L	mg added per 500 ml	ml water per 500 ml	ml syn. sewage feed per 500 ml	ml microbial inoculum per 500 ml	Total volume ml
320	160	259	25	16	200
560	280	259	25	16	200
1000*	500	259	25	16	200
BK Control II	0	284	0	16	200

Shared BK Control I with LR H1480-1.

* Slight reduction in amount of foam formed when compared
 to FC203 CE without antifoamer.

This test solution was duplicated. Used in measuring TSS.

ENVIRONMENTAL LABORATORY

OECD ACTIVATED SLUDGE RESPIRATION INHIBITION TEST #209

RAW DATA

Test Substance: FC-203CE Lot 519 (2/21/89) with Antifoamer SWS 214 (200mg/L)
 Source of Activated Sludge Mixed Liquor: Metro Waste Treatment Plant, St Paul, MN
 Date Received: 3/19/90 Date Used: 3/19/90
 Analyst: Rochelle Robideau

30 - MINUTE EXPOSURE

DISSOLVED OXYGEN CONCENTRATIONS mg/L												
Test Substance Concentration (mg/L)	TIME-MINUTES											TEMP °C
	0.5	1.0	1.5	2.0	2.5	3.0	3.5	4.0	4.5	5.0	5.5	
CONTROL I	7.1	6.3	5.9	5.4	5.0	4.6	4.1	3.7	3.3	2.9	2.5	21
320	3.5	2.5	1.8	1.2	0.7	2:41 0.5						20
5100	3.1	2.2	1.6	1.0	2:25 0.5							20
1000	3.4	2.7	2.0	1.3	0.5							20
CONTROL II	5.8	5.3	4.9	4.4	4.1	3.6	3.2	2.8	2.4	2.0	1.6	21

3 - HOUR EXPOSURE

ROCK EXPOSURE		DISSOLVED OXYGEN CONCENTRATIONS mg/L										
Test Substance Concentration (mg/L)	TIME-MINUTES											TEMP °C
	0.5	1.0	1.5	2.0	2.5	3.0	3.5	4.0	4.5	5.0	5.5	
CONTROL I	5.9	5.6	5.2	4.8	4.5	4.1	3.7	3.2	2.8	2.4	2.0	21
320	5.0	4.5	4.0	3.6	3.1	2.8	2.3	1.9	1.5	1.2	0.9	21
5100	5.2	4.7	4.3	3.9	3.5	3.1	2.7	2.3	1.9	1.5	1.1	21
1000	6.3	5.9	5.6	5.3	5.0	4.6	4.3	4.0	3.6	3.3	2.9	21
CONTROL II	5.9	5.5	5.1	4.6	4.2	3.8	3.3	2.9	2.5	2.1	1.6	22

pH of Activated Sludge Mixed Liquor: Initial 6.6 Final 6.9
 pH of highest concentration tested: Initial 6.6 Final 6.8
 Test Temperature: 20-22 °C

ENVIRONMENTAL LABORATORY

ACTIVATED SLUDGE RESPIRATION INHIBITION TEST #209
CALCULATION OF RESULTSLR 41480-3
Date 3/19/90
Analyst RRR
page 8 of 8Test Substance EC203CE Lot 519 (2/21/90) with Antifoamer SWS214 (200 mg/L)

30 - MINUTE EXPOSURE

Test Substance Concentration (mg/L)	Oxygen Uptake Rate R	R Corrected ⁽¹⁾ to 20°C	% Inhibition in Respiration Rate ⁽²⁾
CONTROL I	0.8497	0.8116	
320	1.1747	1.1747	— -47
560	1.1976	1.1976	— -50
1000	1.4600	1.4600	— -83
CONTROL II	0.8230	0.7861	

Percent Difference in Controls⁽³⁾ 3.2%EC₅₀ (95% Confidence Limits) mg/L _____

3 - HOUR EXPOSURE

Test Substance Concentration (mg/L)	Oxygen Uptake Rate R	R Corrected ⁽¹⁾ to 20°C	% Inhibition in Respiration Rate ⁽²⁾
CONTROL I	0.8036	0.7675	
320	0.8073	0.7711	0.7
560	0.8000	0.7641	1.6
1000	0.6655	0.6356	18
CONTROL II	0.81606	0.7852	

Percent Difference in Controls⁽³⁾ 2.3%EC₅₀ (95% Confidence Limits) mg/L _____

$$(1) \text{ Temp. Correction} = \frac{R_{T, \text{obs}}}{\theta^x} \quad (\text{mg O}_2/\text{L-min.})$$

$$(2) \% \text{ Inhibition} = \frac{\left(\frac{RC_1 + RC_2}{2} \right) - R_S}{\left(\frac{RC_1 + RC_2}{2} \right)} \times 100$$

Where:

R = Oxygen uptake rate (mg O₂/L-min.)T_{obs} = Sample temp., °Cθ = 1.047, x = T_{obs} - 20°C

C = Control

$$(3) \% \text{ Difference} = \frac{|RC_1 - RC_2|}{\left(\frac{RC_1 + RC_2}{2} \right)} \times 100$$

The two control respiration rates should be within 15% of each other

LR H1480-5

Date 3/23/90

Analyst JS

ENVIRONMENTAL LABORATORY
ACTIVATED SLUDGE MIXED LIQUOR

Type Test Activated Sludge Respiration Inhibition Test (OECD) #209

Source Metro Waste Treatment Plant, St. Paul, Mn

Date Obtained 3/23/90 Date Used 3/23/90

Mixed Liquor Suspended Solids (MLSS) 7.853 g/L Adjustment g/L

pH initial 6.7, final 6.9

Comments _____

Test Substance

Sample Description SW5214 Antifoamer (4/7/86) Silicones Corporation, Adrian, Mi.
highest concentration tested, 1000 mg/L

Sample pH initial 6.7, final 6.8 Adjustment _____

Comments Stock solution pH 6.7.

Reference Substance

Sample Description 3,5-Dichlorophenol
highest concentration tested, 10 mg/L

Sample pH initial 6.8, final 7.0 Adjustment _____

Comments Stock solution pH 7.2.

Exposure Conditions

Test Duration 3 hours

Test Temperature 18-19 °C

Comments _____

Lab Request No. H1480-5

Environmental Lab — Work Sheet

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Sample Description

OECD ACTIVATED SLUDGE
RESPIRATION INHIBITION TEST #209Date: 3/23/90Analyst: SPJ

Stock and Test Solutions Preparation

SWS 214 Antifoamer (4/7/86) Silicones Corporation, Adrian, Mi.

STOCK SOLUTION: 2000 mg/500 ml Each 1 ml = 4 mg

- 2000 mg SWS 214 / 500 ml Milli-Q water.

(SWS 214 very miscible). Stock soln. pH 6.7.

Test Conc. mg/L	ml stock soln per 500 ml	ml MQ water per 500 ml	ml syn. sewage feed per 500 ml	ml microbial inoculum per 500 ml	Total volume ml				
1000	125	+	159	+	16	+	200	=	500

Shared BK Controls with LK H1480-244.

ENVIRONMENTAL LABORATORY

OECD ACTIVATED SLUDGE RESPIRATION INHIBITION TEST #209

RAW DATA

Test Substance: SW5214 Antifoamer (4/7/86) Silicones Corporation, Adrian, Mi.
Source of Activated Sludge Mixed Liquor: Metro Waste Treatment Plant, St. Paul, Mn
Date Received: 3/23/90 Date Used: 3/23/90
Analyst: Rechele Robideau

30 - MINUTE EXPOSURE

DISSOLVED OXYGEN CONCENTRATIONS mg/L												
Test Substance Concentration (mg/L)	TIME-MINUTES											TEMP °C
	0.5	1.0	1.5	2.0	2.5	3.0	3.5	4.0	4.5	5.0	5.5	
CONTROL I	7.4	6.5	5.8	5.3	4.7	4.2	3.7	3.1	2.6	2.1	1.6	18
1000	3.0	1.9	1.3	0.8	2:17 0.5							18
CONTROL II	5.8	5.2	4.5	4.0	3.5	2.9	2.5	1.9	1.4	0.9	5:27 0.5	18

3 - HOUR EXPOSURE

		DISSOLVED OXYGEN CONCENTRATIONS mg/L											
Test Substance Concentration (mg/L)	TIME-MINUTES											TEMP °C	
	0.5	1.0	1.5	2.0	2.5	3.0	3.5	4.0	4.5	5.0	5.5		
CONTROL I	6.5	6.1	5.6	4.9	4.4	3.8	3.2	2.4	2.0	1.5	1.0	19	
1000	3.0	2.2	1.5	1.0	2.2 0.5							19	
CONTROL II	5.8	4.9	4.4	3.8	3.3	2.7	2.4	1.7	1.2	0.5		18	

pH of Activated Sludge Mixed Liquor: Initial 6.7
pH of highest concentration tested: Initial 6.7
Test Temperature: 18-19°C

Final 6.9
Final 6.8

ENVIRONMENTAL LABORATORY

ACTIVATED SLUDGE RESPIRATION INHIBITION TEST #209
CALCULATION OF RESULTSLR H1480-5
Date 3/23/90
Analyst SJS
Page 3 of 3Test Substance SWS 214 Antifoamer (4/1/86) Silicones Corporation, Adrian, Mi.

30 - MINUTE EXPOSURE

Test Substance Concentration (mg/L)	Oxygen Uptake Rate R	R Corrected ⁽¹⁾ to 20°C	% Inhibition in Respiration Rate ⁽²⁾
CONTROL I	1.0764	1.1803	
1000	1.0849	1.1896	1.6
CONTROL II	1.1297	1.2387	

Percent Difference in Controls⁽³⁾ 4.8%EC₅₀ (95% Confidence Limits) mg/L _____

3 - HOUR EXPOSURE

Test Substance Concentration (mg/L)	Oxygen Uptake Rate R	R Corrected ⁽¹⁾ to 20°C	% Inhibition in Respiration Rate ⁽²⁾
CONTROL I	1.1939	1.2502	
1000	1.1556	1.2101	—
CONTROL II	1.0767	1.1806	

Percent Difference in Controls⁽³⁾ 5.7%EC₅₀ (95% Confidence Limits) mg/L _____

$$(1) \text{ Temp. Correction} = \frac{R_{T, \text{obs}}}{\theta^x} \quad (\text{mg O}_2/\text{L-min.})$$

$$(2) \% \text{ Inhibition} = \frac{\left(\frac{RC_1 + RC_2}{2} \right) - R_S}{\left(\frac{RC_1 + RC_2}{2} \right)} \times 100$$

Where:

R = Oxygen uptake rate (mg O₂/L-min.)T_{obs} = Sample temp., °Cθ = 1.047, x = T_{obs} - 20°C

C = Control

$$(3) \% \text{ Difference} = \frac{|RC_1 - RC_2|}{\left(\frac{RC_1 + RC_2}{2} \right)} \times 100$$

The two control respiration rates should be within 15% of each other