

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-201AF. (1-Octanesulfonic acid) (CAS # 2795-39-3).

Remarks: The 3M production lot number was not noted. The test sample is FC-201AF. Current information indicates it is a mixture of 3% PFOS, 18.59% diethylene glycol butyl ether, 37.81% water, 14% ethylene glycol, 8.25% Acrylic foamer, 9% sodium octyl sulfate, 0.3% sodium lauryl sulfate, 3% triethanolamine, 6% sulfonated alkyl ether salts, and 0.05% tolyltriazole.

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 Guideline 209

Test type: Static acute

GLP: No

Year Completed: 1995

Analytical monitoring: Dissolved oxygen, temperature, and pH.

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

Test organism source: Activated sludge mixed liquor collected from the Metro Wastewater 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 solution was measured to be 7.3. Test substance was added directly to the test vessels.

Test vessels: Not given.

Number of concentrations: one plus blank control, in duplicate and 1 reference substance

Temperature: 19.3 – 20.7°C.

Total suspended solids and pH on day of testing: 4.2 g/L (TSS in final: 1.7 g/L), initial pH 6.0 and final pH 6.5.

Element Basis: Respiration inhibition as determined by oxygen consumption.

Remarks: Uptake rates corrected to 20°C for calculations.

RESULTS

Nominal concentrations: Blank controls and 1000 mg/L in duplicate plus one reference substance at 10 mg/L.

Element values: 30-minute EC₅₀ = >1000 mg/L
3-hour EC₅₀ = >1000 mg/L

Element values based on nominal concentrations.

Remarks: 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 EC₅₀ 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. This study meets the 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 N2964, 1995.

OTHER

Last changed: 6/26/00

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ACTIVATED SLUDGE MIXED LIQUOR

LR N2964
Date 9/14/95
Analyst GWJ
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Type Test OECD Activated Sludge Respiration Inhibition Test #209

ACTIVATED SLUDGE MIXED LIQUOR

Date Obtained 9/14/95

Date Used 9/14/95

Mixed Liquor Suspended Solids (MLSS) 4.2 g/L Adjusted to g/L

MLSS in final test solution 1.7 g/L

Initial pH 6.0 (1st test soln. prepared)

Final pH 6.5 (last soln. prepared)

Comments After letting sludge settle out, decanted ~2L off the top layer
then mixed remaining sludge.

TEST SUBSTANCE

Sample Description FC-201AF

Initial pH of highest conc. tested (1000 mg/L) = 7.1

Adjusted to pH

Comments

REFERENCE SUBSTANCE

Sample Description 3,5-Dichlorophenol

Initial pH of stock solution (10 mg/L) = 7.3

Adjusted to pH

Comments

EXPOSURE CONDITIONS

Test Duration 3 hrs

Test Temperature Range 19.3-20.7 °C

Comments

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LR N2964

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OECD ACTIVATED SLUDGE RESPIRATION INHIBITION TEST #209 TEST SOLUTIONS PREPARATION

Test Substance: FC-201 AF

Sample Description: Amber brown liquid

Analyst: GWS

Date: 9/14/95

Test Conc. mg/L	mg added per 500 mL	mL MQ(1) water per 500 mL	mL syn. sewage feed per 500 mL	mL microbial inoculum per 500 mL	Total Volume mL
Blank Control	—	284	16	200	500
1000	500	284	16	200	500

Initial pH of 1000 mg/L, pH = 7.1

1) Millipore Milli-Q high purity water

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LR N2964

OECD ACTIVATED SLUDGE RESPIRATION INHIBITION TEST #209

RAW DATA

Test Substance: FC-201AF

Source of Activated Sludge Mixed Liquor: Metro Waste Water Treatment Plant St. Paul, MN

Date Received: 9/14/95

Date Used: 9/14/95

Analyst: GWJ

30-MINUTE EXPOSURE DISSOLVED OXYGEN CONCENTRATIONS, mg/L

Test Substance Concentration (mg/L)	0.5	1.0	1.5	2.0	TIME - MINUTES			4.0	4.5	5.0	5.5	Temp °C
					2.5	3.0	3.5					
BK CONTROL I	7.0	6.6	6.3	6.0	5.6	5.3	5.0	4.7	4.4	4.1	3.7	19.9
1000	4.2	3.4	2.8	2.2	1.5	0.9						20.7
DUP	4.0	3.3	2.8	2.2	1.5	1.0						20.6
BK CONTROL II	8.0	7.6	7.4	7.2	6.9	6.6	6.3	6.1	5.8	5.4	5.0	19.8

3-HOUR EXPOSURE DISSOLVED OXYGEN CONCENTRATIONS, mg/L

Test Substance Concentration (mg/L)	0.5	1.0	1.5	2.0	TIME - MINUTES			4.0	4.5	5.0	5.5	Temp °C
					2.5	3.0	3.5					
BK CONTROL I		7.4	7.2	6.9	6.7	6.4	6.1	5.8	5.5	5.3	4.9	19.4
1000	5.1	4.5	4.0	3.6	3.0	2.6	2.0	1.6	1.0	0.8		20.3
DUP	6.3	5.8	5.4	4.9	4.4	3.9	3.4	2.8	2.5	2.0	1.5	19.9
BK CONTROL II		7.8	7.6	7.2	7.0	6.7	6.4	6.1	5.8	5.5	5.1	19.2

OE of activated sludge mixed liquor
OE of highest concentration tested
test temperature

Initial DO
Initial DO

Final DO
Final DO

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ENVIRONMENTAL LABORATORY
OECD ACTIVATED SLUDGE RESPIRATION INHIBITION TEST #209
CALCULATION OF RESULTS

Lr: N2864
Date: 9/14/95

Test Substance: EC-201AF

Analyst: GML

30-Minute Exposure		Dissolved Oxygen Concentrations, mg/L										Temp. °C	R (1)	Ra (2)	% Inh. (3)
mg/L		0.5	1.0	1.5	2.0	2.5	3.0	3.5	4.0	4.5	5.0	5.5			
BK1	7.0	6.6	6.3	6.0	5.6	5.3	5.0	4.7	4.4	4.1	3.7	19.9	0.6364	0.6408	97
1000.0	4.2	3.4	2.8	2.2	1.5	0.9						20.7	1.2600	1.2001	86
Dup.	4.0	3.3	2.8	2.2	1.5	1.0						20.6	1.1800	1.1318	
BK2	8.0	7.6	7.4	7.2	6.9	6.6	6.3	6.1	5.8	5.4	5.0	19.8	0.5709	0.5789	
% diff in controls (4):		10.2		EC50 (95% CI): > 1000 mg/L										RaCm (5): 0.6099	

3-Hour Exposure		Dissolved Oxygen Concentrations, mg/L										Temp. °C	R (1)	Ra (2)	% Inh. (3)
mg/L		0.5	1.0	1.5	2.0	2.5	3.0	3.5	4.0	4.5	5.0	5.5			
BK1	7.7	7.4	7.2	6.9	6.7	6.4	6.1	5.8	5.5	5.3	4.9	19.4	0.5552	0.5788	68
1000.0	5.1	4.5	4.0	3.6	3.0	2.6	2.0	1.6	1.0	0.5		20.3	1.0000	0.9794	66
Dup.	6.3	5.8	5.4	4.9	4.4	3.9	3.4	3.0	2.5	2.0	1.5	19.9	0.9600	0.9667	
BK2	8.3	8.0	7.8	7.5	7.2	7.0	6.7	6.4	6.1	5.8	5.5	19.3	0.5600	0.5879	
% diff in controls (4):		1.6		EC50 (95% CI): > 1000 mg/L										RaCm (5): 0.5834	

- (1) R= Oxygen uptake rate (mg O₂/L·min)
- (2) Ra= R adjusted to 20°C= [R/(E-1)]
- (3) % Inhibition= [(RaCm)-RaS]/(RaCm)]100
- (4) % difference in controls: [(RaBk1-RaBk2)/(RaCm)]100 (Two controls must be within 15% of each other.)
- (5) Mean adjusted R, controls= [RaBk1+RaBk2]/2

α= 1.072
RaCm= mean adjusted R, controls
Bk= blank control, S= sample
Tobs= sample temp
T= Tobs-20°C

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LR 112965/112964

ENVIRONMENTAL LABORATORY

OECD ACTIVATED SLUDGE RESPIRATION INHIBITION TEST # 209
TEST SOLUTIONS PREPARATION

Test Substance: 3,5-Dichlorophenol Reference Substance
Sample Description: Aldrich Chemical Co. Red Label Lot # 00410KZ
Analyst: GWJ
Date: 9/14/95

Stock Solution

Preparation: 0.5g/L; 1 mL = 0.5mg
Weighted out 0.5g 3,5-DCP. Then added to 10 mL of 1N NaOH and diluted up to
~30 mL with Mill. Q H₂O, while stirring, until dissolved. Then added ~8 mL of
1N H₂SO₄ while stirring, until the point of incipient precipitation. Then finally
diluted to the mark at 1L with Mill. Q H₂O.

Initial pH = 7.3

Test Conc. mg/L	mL added per 500 mL	mL MQ(1) water per 500 mL	mL syn. sewage feed per 500 mL	mL microbial inoculum per 500 mL	Total Volume mL
Blank Control	—	284	16	200	500
10	10	274	16	200	500

(4) Millipore Milli-Q[®] high purity water

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ENVIRONMENTAL LABORATORY

OECD ACTIVATED SLUDGE RESPIRATION INHIBITION TEST #209

RAW DATA

Test Substance: 3,5-Dichlorophenol Reference Substance
 Source of Activated Sludge Mixed Liquor: Metro Waste Water Treatment Plant St. Paul, MN
 Date Received: 9/14/95 Date Used: 9/14/95
 Analyst: G.W.J.

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	
BK CONTROL I	7.0	6.6	6.3	6.0	5.6	5.3	5.0	4.7	4.4	4.1	3.7	19.9
1.0	8.0	7.8	7.7	7.5	7.2	7.0	6.8	6.6	6.4	6.1	5.9	20.3
BK CONTROL II	8.0	7.6	7.4	7.2	6.9	6.6	6.3	6.1	5.8	5.4	5.0	19.8

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	
BK CONTROL I	7.7	7.4	7.2	6.9	6.7	6.4	6.1	5.8	5.5	5.3	4.9	19.4
1.0	8.3	8.2	8.1	8.0	7.9	7.7	7.6	7.4	7.3	7.1	7.0	20.4
BK CONTROL II	8.3	8.2	8.1	7.9	7.7	7.6	7.4	7.3	7.1	7.0	6.5	19.5

OE of activated sludge mixed liquor
 OE of highest concentration tested
 Test temperature

Initial DO
 Initial DO

Final DO
 Final DO

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ENVIRONMENTAL LABORATORY OECD ACTIVATED SLUDGE RESPIRATION INHIBITION TEST #209 CALCULATION OF RESULTS

Test Substance: 3,5-DICHLOROPHENOL

Analyst: GML

30-Minute Exposure												Dissolved Oxygen Concentrations, mg/L												Temp. °C			R (1)		Ra (2)		% inh. (3)				
mg/L	0.5	1.0	1.5	2.0	2.5	3.0	3.5	4.0	4.5	5.0	5.5																								
BK1	7.0	6.6	6.3	6.0	5.6	5.3	5.0	4.7	4.4	4.1	3.7																								
10	8.0	7.8	7.7	7.5	7.2	7.0	6.8	6.6	6.4	6.1	5.9																								
BK2	8.0	7.6	7.4	7.2	6.9	6.6	6.3	6.1	5.8	5.4	5.0																								
% diff in controls (4): 10.2												RaCm (5): 0.6099												19.8		0.5709		0.5789							

3-Hour Exposure												Dissolved Oxygen Concentrations, mg/L												Temp. °C			R (1)			Ra (2)			% inh. (3)		
mg/L	0.5	1.0	1.5	2.0	2.5	3.0	3.5	4.0	4.5	5.0	5.5																								
BK1	7.7	7.4	7.2	6.9	6.7	6.4	6.1	5.8	5.5	5.3	4.9																								
10	8.3	8.2	8.1	8.0	7.9	7.7	7.6	7.4	7.3	7.1	7.0																								
BK2	8.3	8.0	7.8	7.5	7.2	7.0	6.7	6.4	6.1	5.8	5.5																								
% diff in controls (4): 1.6												RaCm (5): 0.5834												19.3			0.5600			0.5879					

- (1) R= Oxygen uptake rate (mg O₂/L·min)
- (2) Ra= R adjusted to 20°C= [R/E^{Ex}]
- (3) % Inhibition= [(RaCm)-RaS/(RaCm)]100
- (4) % difference in controls: [(RaBk1-RaBk2)/(RaCm)]100 (Two controls must be within 15% of each other.)
- (5) Mean adjusted R, controls= (RaBk1+RaBk2)/2

a= 1.072
 RaCm= mean adjusted R, controls
 Bk= blank control, S= sample
 Tobs= sample temp
 x= Tobs-20°C

L/R N2965/N2964
 Date: 9/4/95

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