

ACTIVATED SLUDGE RESPIRATION INHIBITION TEST

TEST SUBSTANCE

Identity: Perfluorooctanoic acid, ammonium salt; may also be referred to as PFOA ammonium salt, Ammonium perfluorooctanoate, FC-116, FC-126, FC-169, or FC-143. (Octanoic acid, pentadecafluoro-, ammonium salt, CAS # 3825-26-1)

Remarks: 3M production lot number 390. The test sample is FC-126, a white powdery solid. It's purity was not sufficiently characterized, though current information indicates it is a mixture of 78 - 93% test substance and 7 - 22% C₅, C₆, and C₇ perfluoro analogue compounds.

METHOD:

Method: OECD 209

Test type: Static acute

GLP: No

Year completed: 1987

Analytical monitoring: Dissolved oxygen concentrations.

Test organism source: Activated sludge mixed liquor collected from the metro Wastewater Treatment Plant, St Paul, MN. Used the same day as collection.

Test conditions:

Dilution water: Aerated, distilled, deionized water

Synthetic sewage: per OECD 209

Test vessels: Not stated

Number of concentrations: 5 plus one reference substance and 2 blank controls.

Test temperature: 20 - 21 °C

Mixed liquor Suspended Solids: 2.7 g/L

pH of Activated Sludge Mixed Liquor: 7.8 (initial) and 8.0 (final)

pH of 1000 mg/L test concentration: 7.4 (initial) and 8.1 (final)

Exposure period: 30-minutes and 3-hours

Reference and test solution preparation: All test substance solutions were created by adding individual weights to solutions containing 284 mL DI water, 16 mL synthetic sewage feed, and 200 mL inoculum. The pH of these solutions was then adjusted to 7.0 using 1N NaOH. Reference solutions were created using a 500 mg/L stock solution of 3,5-dichlorophenol

Element basis: Respiration inhibition as determined by dissolved oxygen consumption.

RESULTS

Nominal concentrations: Two Blank controls, 100, 180, 320, 560, and 1000 mg/L test material solution.

Percent Inhibition in Respiration Rate

Test Substance Concentrations (mg/L)	30-Minute Exposure	3-Hour Exposure
Control I	0	0
Control II	0	0
100	1.1	4.2
180	0	4.7
320	7.0	14
560	11	17
1000	29	38

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

Control response: Adequate – 2.2% difference in respiration rate of the two controls for the 30-minute exposure and 0.8% difference in respiration rate of the two controls for the 3-hour exposure.

CONCLUSIONS

The test material induced 38% inhibition in respiration rate at 1000 mg/L after 3-hours of exposure.

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, it lacks characterization of the test substance purity and analytical confirmation of test substance concentrations. No results were given for the reference compound.

REFERENCES

This study was conducted by the 3M Company, Environmental Laboratory, St. Paul, MN, Lab Request number E1282.

OTHER

Last changed: 5/24/00

003856

LR E1282
Date 4-20-87
Analyst SAB

ENVIRONMENTAL LABORATORY
ACTIVATED SLUDGE MIXED LIQUOR

Type Test Activated Sludge Respiration Inhibition OECD #209

Source Metro Waste Treatment Plant St Paul, MN

Date Obtained 4-20-87 Date Used 4-20-87

Mixed Liquor Suspended Solids (MLSS) 2.7 g/L Adjustment None g/L

pH Initial = 7.8 Final = 8.0

Comments _____

Test Substance

Sample Description FC-126 Lot 390

Sample pH of highest conc. 1000 mg/L = 6.6 Adjustment All test solns. to 7.0

Comments Individual weights

Reference Substance

Sample Description 3,5-Dichlorophenol

Sample pH 7.3 Adjustment None

Comments Stock Solution, 500 mg/L

Exposure Conditions

Test Duration 3-Hours

Test Temperature 20-21 °C

Comments Test ran with LR E1282

003857

Environmental Lab - Work Sheet

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Sample Description

Activated Sludge Respiration
FC-126 Lot 390 Inhibition Test OECD #209Date: 4-20-87Analyst: SAB- white powdery
Solid

Test Solutions Preparation

Test Conc. mg/L	(1) mg/5cc ml	ml DI water Added/5cc ml	ml synthetic sewage feed Added/5cc ml	ml microbial Inoculum Added/5cc ml	Total mixture vol. ml
100	50	+ 284	+ 16	+ 200	= 500
180	90	+ 284	+ 16	+ 200	= 500
320	160	+ 284	+ 16	+ 200	= 500
560	280	+ 284	+ 16	+ 200	= 500
1000	500	+ 284	+ 16	+ 200	= 500
Blank (2) Controls	0	284	+ 16	+ 200	= 500

(1) Individual weights. Appeared to dissolve completely

Initial pH highest conc. tested, 1000 mg/L, = 6.6

Adjusted all test solns. to pH 7.0 with 1N NaOH

(2) Control I shared with LR E1281-1, FC-44x, Lot-101 Test, as
Control II

DI water used:

Distilled 4-16-87, Aeration started 4-17-87 pH = 6.7

ENVIRONMENTAL LABORATORY

ACTIVATED SLUDGE RESPIRATION INHIBITION TEST OECD #209

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RAW DATA

Test Substance: FC-126 Lot 390Source of Activated Sludge Mixed Liquor: Metro Waste Treatment Plant St Paul, MNDate Received: 4-20-87Date Used: 4-20-87Analyst: Juan A. Beach30 - MIN. EXPOSURE

DISSOLVED OXYGEN CONCENTRATIONS mg/L

Test Substance Concentration (mg/L)	TIME-MINUTES											Temp °C
	0.5	1.0	2.0	3.0	4.0	5.0	6.0	7.0	8.0	9.0	10.0	
CONTROL I	6.6	6.2	5.5	4.8	4.1	3.3	2.6	1.9	1.2	0.5 8:51		21
100	5.9	5.5	4.8	4.1	3.4	2.7	2.0	1.3	0.5			21
180	5.3	4.9	4.2	3.5	2.8	2.1	1.3	0.6	0.5 7:08			21
320	5.3	4.9	4.2	3.5	2.9	2.2	1.6	0.9	0.5 7:31			21
560	5.5	5.2	4.5	3.9	3.3	2.7	2.1	1.5	0.9	0.5 8:42		20
1000	6.1	5.8	5.3	4.9	4.4	3.9	3.4	2.9	2.4	1.9	1.5	20
CONTROL II	6.7	6.4	5.7	5.0	4.3	3.6	2.9	2.2	1.5	0.7 4:20		21

3 - HOUR EXPOSURE

DISSOLVED OXYGEN CONCENTRATIONS mg/L

Test Substance Concentration (mg/L)	TIME-MINUTES											Temp °C
	0.5	1.0	2.0	3.0	4.0	5.0	6.0	7.0	8.0	9.0	10.0	
CONTROL I	7.4	7.2	6.7	6.2	5.6	5.1	4.6	4.1	3.5	3.0	2.5	21
100	6.9	6.8	6.3	5.8	5.3	4.8	4.3	3.8	3.3	2.8	2.2	21
180	8.0	7.9	7.5	7.0	6.5	6.1	5.6	5.1	4.6	4.1	3.6	20
320	8.4	8.3	7.9	7.5	7.1	6.7	6.2	5.8	5.3	4.9	4.4	20
560	8.0	7.9	7.6	7.2	6.7	6.3	5.9	5.4	4.9	4.5	4.0	21
1000	8.5	8.5	8.3	8.0	7.7	7.4	7.1	6.7	6.4	6.1	5.7	20
CONTROL II	7.4	7.2	6.7	6.1	5.6	5.1	4.6	4.0	3.5	3.0	2.4	21

pH of Activated Sludge Mixed Liquor Initial = 7.8

Final = 8.0

pH of highest concentration tested Initial = 7.4

Final = 8.1

Test Temperature 20-21°C

see pg 1 for
soln. prep.

003859

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

LR E1282
Date 4-20-87
Analyst SLB

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Test Substance EC-126 Lot 390

30 - MINUTE EXPOSURE

Test Substance Concentrations (mg/L)	Oxygen Uptake Rate R	R Corrected to 20°C	% Inhibition in Respiration Rate (1)
CONTROL I	0.7238	0.6913	-
100	0.7083	0.6765	1.1
180	0.7197	0.6874	-
320	0.6655	0.6356	7.0
560	0.6056	0.6056	11
1000	0.4848	0.4848	29
CONTROL II	0.7081	0.6763	-

Percent Difference in Controls (2) 2.2 %

EC50 (95 % Confidence Limits) mg/L 320, 560, 1000 mg/L

EC50 = 2010
(1350-5690)
mg/L

3 - HOUR EXPOSURE

Test Substance Concentrations (mg/L)	Oxygen Uptake Rate R	R Corrected to 20°C	% Inhibition in Respiration Rate (1)
CONTROL I	0.5255	0.5019 0.4938	-
100	0.5055	0.4828	4.2
180	0.4800	0.4800	4.7
320	0.4333	0.4333	14
560	0.4400	0.4202	17
1000	0.3145	0.3145	38
CONTROL II	0.5297	0.5059	-

Percent Difference in Controls (2) 0.8 %

EC50 (95% Confidence Limits) mg/L 320, 560, 1000 mg/L

EC50 = 1680
(1150-4720)
mg/L

See pg 4 for stock test soln. prep. SLB
4-20-87

$$(1) \% \text{ Inhibition} = \left(\frac{RC_1 + RC_2}{2} \right) - R_S$$

$$\left(\frac{RC_1 + RC_2}{2} \right)$$

Temp. Corrected to 20°C:

$$\frac{R_{T,obs}}{\theta^{T-20}} = \text{mg/L per min. at } 20^\circ\text{C}$$

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

Where: R = Oxygen Uptake Rate (mg O₂/L/Min.),
S = Test Substance

C₁ = Control I C₂ = Control II

θ = 1.047
x = T_{obs} - 20°C

003860

The two BK Control respiration rates should be within 15% of each other.