

## ALGICIDAL ACTIVITY

### TEST SUBSTANCE

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

**Remarks:** The 3M production lot number was not noted. The test sample is FC-203. Current information indicates it is a mixture of 1.34% PFOS, 35% diethylene glycol butyl ether, 37.85% water, 20% ethylene glycol, 2.66 % Sultone foamer, 3% sodium octyl sulfate, 0.1% sodium lauryl sulfate, and 0.05% tolyltriazole.

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

### METHOD:

**Method:** Standard No. 22, Relating to Swimming Pool Water Treatment Chemicals and/or Processes (National Sanitation Foundation)

**Type (test type):** Acute Static

**GLP:** No

**Year study performed:** 1974

**Species:** *Chlorella pyrenoidosa* and *Phormidium inundatum*

**Source:** In-house cultures maintained at Industrial Bio-test Laboratories, Inc., Northbrook, IL.

**Element basis:** Cell populations calculated from hemocytometer counts, turbidimetric measurement.

**Exposure period:** 14 days

**Statistical Methods:** Not given.

**Analytical monitoring:** Not given.

### **Test Conditions:**

**Algal Nutrient Medium:** Allen's nutrient medium.

**Stock and test solution preparation:** Algal solutions prepared by blending algae with 100 mL of Allen's medium using a Waring Blender. One mL aliquots of the test substance were handled as 100% active and diluted with distilled water and added to Allen's medium. One mL of adjusted cell suspension was added to each test solution, for a final volume of 100 mL.

**Exposure vessels:** Flasks containing final volume of 100 mL.

**Number of replicates:** 1

**Initial cell loading:**  $7.5 \times 10^4$  cells/mL for *Chlorella*,  $12.5 \times 10^4$  cells/mL for *Phormidium*

**Exhibit  
2714**

State of Minnesota v. 3M Co.,  
Court File No. 27-CV-10-28862

3M\_MN00436466

**Number of concentrations:** six plus a blank control for *Chlorella*, five plus a blank control for *Phormidium*.

**Lighting:** Not given

**Subculture:** Inoculated bottles subcultured to fresh medium after one day incubation, with final readings made after 14 days inoculation. Three dilutions of the test substance were subcultured (1:5, 1:10, and 1:100).

**Water Chemistry:** Not given.

## RESULTS

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**Nominal concentrations:** Bk control, 1:5, 1:10, 1:100, 1:150, 1:1000, and 1:10000 dilution for *Chlorella pyrenoidosa* study. Bk control, 1:5, 1:10, 1:100, 1:1000, and 1:10000 dilution for *Phorinidium inundatum* study.

### Results of the Study with *Chlorella pyrenoidosa*

Sample (dilution)	Gross Appearance (Color)	Turbidity (absorbance)
Control	green	>2.00
1:5	none	<0.01
1:10	none	<0.01
1:100	none	<0.01
1:150	Slight green tint	0.03
1:1000	green	>2.00
1:10000	green	>2.00
1:5 subculture	none	<0.01
1:10 subculture	none	<0.01
1:100 subculture	none	<0.01

### Results of the Study with *Phorinidium inundatum*

Sample (dilution)	Gross Appearance (Color)	Turbidity (absorbance)
Control	green	>2.00
1:5	none	<0.01
1:10	none	<0.01
1:100	none	<0.01
1:1000	green tint	>2.00
1:10000	green tint	>2.00
1:5 subculture	none	<0.01
1:10 subculture	none	<0.01
1:100 subculture	none	<0.01

Element values are based on dilutions of nominal concentrations.

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

#### CONCLUSIONS

Algicidal rather than algistatic action of the test substance was obtained.

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

#### DATA QUALITY

Reliability: Klimisch ranking = 3. Study lacks description and records of the method followed. The sample purity was not properly characterized and the study lacks analytical confirmation of the amount of fluorochemical proportion in the solution.

#### REFERENCES

Test was conducted by Industrial Bio-Test Laboratories, Inc., of Northbrook, IL at the request of the 3M Company, St. Paul, MN, 1974.

#### OTHER

**Last changed:** 6/27/00

*Neonic* ~~FC-203~~  
FC-203

*Industrial* **BIO-TEST** *Laboratories, Inc.*

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MICROBIOLOGY DEPT.

FDA REG. NO. 14-14813

REPORT TO

3M COMPANY

ALGICIDAL ACTIVITY OF  
TWO SAMPLES LABELED "A" AND "B"  
<sub>202</sub> <sub>203</sub>

JANUARY 23, 1974

IBT NO. 633-04473

3M\_MN00436469

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REPORT TO

3M COMPANY

ALGICIDAL ACTIVITY OF  
TWO SAMPLES LABELED "A" AND "B"

JANUARY 23, 1974

IBT NO. 633-04473

I. Introduction

At the request of 3M Company, tests were conducted to determine the algicidal activity of two samples identified as "A" and "B". Two species of algae, Chlorella pyrenoidosa and Phormidium inundatum, were used in the evaluation.

II. Summary

Two samples, identified as "A" and "B", were submitted on November 20, 1973, by 3M Company. The algicidal test was initiated on November 30, 1973, using two species of algae, Chlorella pyrenoidosa Wisc. 2005, Phormidium inundatum Wisc. 1093.

The test was conducted according to a method outlined and prepared by the National Sanitation Foundation, Standard No. 22, Relating to Swimming Pool Water Treatment Chemicals and/or Processes. The turbidimetric determination conformed to the procedure recommended by Fitzgerald.\*

A 1:100 dilution of Sample "A" or "B", effectively controlled the proliferation of Chlorella sp. and Phormidium sp. for a 14-day period. Cidal rather than static action of the candidate algicides was obtained.

\* Fitzgerald, G. P. Bioassay for Algicidal versus Algistatic Chemicals, Water and Sewage Works, 110-296 (1963)

Respectfully submitted,

INDUSTRIAL BIO-TEST LABORATORIES, INC.

Report prepared by:

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January 23, 1974  
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### III. Procedure

#### A. Preparation of Inoculum

Two species of algae, Chlorella pyrenoidosa Wisc. 2005, and Phormidium inundatum Wisc. 1093, were used as the test organisms. Each organism was subcultured at weekly intervals in Allen's nutrient medium for three weeks, prior to inception of the test.

The suspensions of Chlorella and Phormidium were adjusted to contain  $7.5 \times 10^6$  and  $12.5 \times 10^6$  cells/ml respectively. They were prepared by shaking the algae growth in the flasks, to which was added 100 ml of Allen's medium and blenderized for one minute in a Waring Blender. Cell populations were calculated from hemocytometer counts.

#### B. Preparation of Sample

One ml aliquots of test samples handled as 100% active were diluted in distilled water and added to Allen's medium for a final concentration ranging from 1:5 to 1:10,000. These concentrations were achieved after the addition of 1 ml of the adjusted algae cell suspension for a final volume of 100 ml. The numbers of Chlorella and Phormidium cells used were  $7.5 \times 10^4$  and  $12.5 \times 10^4$  respectively. Six control flasks, two per species, were also prepared. All inoculated bottles were subcultured to fresh medium after one day incubation. Final readings for the presence of algal growth in all bottles were made fourteen days after inoculation.

IV. Results

The results of the growth examination and turbidimetric measurement of all flasks inoculated with Chlorella pyrenoidosa and Phormidium inundatum used for the algicidal tests are presented in Table I and II respectively.



TABLE I

Results of the Gross and Turbidimetric Examination of Chlorella pyrenoidosa

## Algicidal Test

Sample "A" (dilution)	Gross Appearance (color)	Turbidity (absorbance)
1:5	none	< 0.01
1:10	none	< 0.01
1:100	none	< 0.01
1:150	slight green tint	0.03
1:1,000	green	> 2.00
1:10,000	green	> 2.00
1:5 subculture	none	< 0.01
1:10 subculture	none	< 0.01
1:100 subculture	none	< 0.01
Control	green	> 2.00
<u>Sample "B"</u>		
1:5	none	< 0.01
1:10	none	< 0.01
1:100	none	< 0.01
1:150	slight green tint	0.03
1:1,000	green	> 2.00
1:10,000	green	> 2.00
1:5 subculture	none	< 0.01
1:10 subculture	none	< 0.01
1:100 subculture	none	< 0.01
Control	green	> 2.00

1 = 0.001  
 10 = 0.01  
 100 = 0.01  
 1000 = 0.01

TABLE II

Results of the Gross and Turbidimetric  
Examination of Phorinidium inundatum

## Algicidal Test

Sample "A" (dilution)	Gross Appearance (color)	Turbidity (Absorbance)
1:5	none	< 0.01
1:10	none	< 0.01
1:100	none	< 0.01
1:1,000	green tint	> 2.00
1:10,000	green tint	> 2.00
1:5 subculture	none	< 0.01
1:10 subculture	none	< 0.01
1:100 subculture	none	< 0.01
Control	green	> 2.00
<u>Sample "B"</u>		
1:5	none	< 0.01
1:10	none	< 0.01
1:100	none	< 0.01
1:1,000	green tint	> 2.00
1:10,000	green tint	> 2.00
1:5 subculture	none	< 0.01
1:10 subculture	none	< 0.01
1:100 subculture	none	< 0.01
Control	green	> 2.00