ACUTE EASTERN OYSTER SHELL DEPOSITION

TEST SUBSTANCE

Identity: Perfluorooctanesulfonate; may also be referred to as PFOS or FC-95. (1-Octanesulfonic acid, 1,1,2,2,3,3,4,4,5,5,6,6,7,7,8,8,8-heptadecafluoro-, potassium salt, CAS # 2795-39-3)

Remarks: The test substance is a white powder. Sample was taken from 3M lot number 217. Sample was stored under ambient conditions prior to testing. Purity determined to be 90.49% by LC/MS, ^{1}H-HMR, ^{19}F-NMR and elemental analyses techniques.

METHOD

Method: OPPTS 850.1025
Type: Static acute
GLP: Yes
Year completed: Study completed 1999. Report completed 2000
Species: *Crassostrea virginica*
Supplier: P. Cummins Oyster Company, Inc., Baltimore, MD
Shell grinding: Prior to test initiation, recently deposited shell at the rounded (ventral) end was removed using a small electric grinder. Care was taken to remove the shell rim uniformly to produce a smooth, rounded, blunt profile.
Analytical monitoring: PFOS measured at 0, 48, 96-hours
Exposure period: 96-hours
Statistical methods: Shell growth inhibition was calculated for each treatment group as the percent reduction in shell growth relative to mean shell growth in the negative control. The EC50 value was estimated by visual inspection of shell growth inhibition data. The shell growth data was evaluated for normality and homogeneity of variances using the Chi-Square test and Bartlett's test, respectively. Dunnett's test was used to identify treatment groups which had a statistically significant ($\leq$0.05) reduction in shell growth as compared to the control.
Test oyster age: unknown
Length: 33.8 (27.8-41.5) mm,
Pretreatment: None

Test conditions:

Dilution water: Natural seawater diluted to a salinity of 20$^{o}/_{oo}$ with well water
Dilution water chemistry (during the 4-week period immediately preceding the test):
salinity: 21 (20-21) $^{o}/_{oo}$

pH: 8.1 (8.0-8.2)
TOC: < 1.0 mg/L

Stock and test solution preparation: Primary stock prepared in dilution water at 9.1 mg/L and mixed for ~24 hours prior to use. After mixing, primary stock solution appeared clear and colorless with some white particulate material suspended throughout the solution. It was proportionally diluted with dilution water to prepare the four additional test concentrations. All test solutions appeared clear and colorless. Due to the relatively low solubility of PFOS in natural seawater, the highest concentration attainable with this matrix is approximately 3.3 mg/L.
Concentrations dosing rate: Once
Exposure vessels: 52L polyethylene aquaria containing approximately 40L of test solution; water depth approximately 21 cm. Each chamber was continuously stirred to circulate the supplemental algae diet using an electric paddle mixer.
Feeding: Algal cells (*Thalassiosira pseudonana, Skeletonema* sp., *Chaetoceros* sp., *and Isochrysis* sp.) were provided to supplement naturally occurring algae and to maximize oyster growth rates during the test.
Number of replicates: one
Number of oysters per replicate: twenty
Number of concentrations: five plus a negative control
Water chemistry during the study:
Dissolved oxygen range (0 – 96 hours):
6.6 – 7.6 mg/L (control exposure)
6.1 – 7.7 mg/L (3.0 mg/L exposure)
pH range (0 – 96 hours)
7.6 – 8.1 (control exposure)
7.6 – 8.1 (3.0 mg/L exposure)
Test temperature range (0 – 96 hours)
22.2 – 22.3°C (control exposure)
21.8 – 22.7°C (3.0 mg/L exposure)

Method of calculating mean measured concentrations: arithmetic mean

RESULTS

Nominal concentrations: Bk control, 1.2, 2.0, 3.3, 5.5, 9.1 mg/L
Measured concentrations: <LOQ, 0.36, 0.40, 1.3, 1.9, 3.0 mg/L
Element value: 96-hour EC_{50} = >3.0 mg/L (C.I. not calculable)
96-hour NOEC = 1.9 mg/L
All element values based on mean measured concentrations

Statistical evaluation of shell growth:
EC_{50} values could not be calculated due to insufficient shell growth inhibition at the highest attainable concentration.

Analytical Methodology: Analyses of test solutions were performed at Wildlife International Ltd. using high performance liquid chromatography with mass spectrometric detection (HPLC/MS). When determining the concentration of the test substance in the test solutions, the same and most prominent peak response for perfluorooctanesulfonate was used. No attempt was made to quantify on the basis of individual isomeric components. The LOQ (limit of quantitation) was 0.115 mg/L in this study. Samples collected at test initiation had measured values from 28 to 46% of nominal. Measured values for samples taken at 48-hours ranged from 15 to 41% of nominal. Measured values for samples taken at 96-hours ranged from <LOQ to 52% of nominal.

Summary of analytical chemistry data:

Nominal Test Concentration, mg/L	Measured Duplicate Values at 0, 48, and 96-hours, Respectively, mg/L	Mean Measured Concentration, mg/L	Percent of Nominal
Negative Control	All < LOQ	<LOQ	-
1.2	0.331, 0.353, 0.341, 0.429, <LOQ, <LOQ	0.36	30
2.0	0.622, 0.633, 0.299, 0.313, 0.249, 0.257	0.40	20
3.3	1.36, 1.15, 0.924, 0.878, 1.58, 1.72	1.3	39
5.5	2.42, 2.53, 2.02, 2.24, 1.45, 0.970	1.9	35
9.1	3.39, 3.44, 3.01*, 3.74, 3.57, 1.99, 2.19	3.0	33

*3 replicates analyzed at time 0

Biological observations after 96-hours: Oysters in the negative control and all PFOS treatment groups appeared normal and healthy throughout the exposure period.

Shell deposition and shell growth inhibition at test termination:

Mean Measured Concentration, mg/L	Shell Deposition Mean ± SD, mm	Percent Inhibition in Shell Growth
Negative Control	2.67 + 0.824	-
0.36	2.50+ 0.933	6.4
0.40	2.40 + 0.820	10
1.3	2.51 + 0.919	6.0
1.9	2.13 + 0.804	20
3.0	1.91 + 0.591	28

Mortality of controls: None

CONCLUSIONS

The potassium perfluorooctanesulfonate 96-hour EC_{50} for the Eastern Oyster was determined to be > 3.0 mg/L, the highest concentration tested and the practical limit of solubility in unfiltered seawater. The 96-hour no effect concentration was 1.9 mg/L.

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

DATA QUALITY

Reliability: Klimisch ranking = 1

REFERENCES

This study was conducted at Wildlife International Ltd., Easton, MD at the request of the 3M Company.

OTHER

Last changed: 5/3/00