BIODEGRADATION

TEST SUBSTANCE

Identity: N-ethylperfluorooctane sulfonamidoethanol; may also be referred to as N-EtFOSE alcohol. (1-Octanesulfonamide, N-ethyl-1,1,2,2,3,3,4,4,5,5,6,6,7,7,8,8,8-heptadecafluoro-N-(2-hydroxyethyl)-, CAS # 1691-99-2).

Remarks: N-EtFOSE alcohol was a white powder originally from 3M production lot number 30107. It was HPLC purified. An Interim Certificate of Analysis, reports the purity to be 99.9%. All results in this study were calculated assuming 100% purity.

METHOD:

Method: Based on EPA Guidelines OPPTS 835.3200
Test Type: Aerobic
GLP: No, but many GLP procedures followed.
Year Initiated: 2000
Year Completed: 2001
Contact time: 18 days

Inoculum: Activated sludge collected 7/31/00 from the aeration basin at the Metro Wastewater Treatment Plant, St Paul, MN. The mixed liquor suspended solids (MLSS) were determined to be 2,280 mg/L when first collected. The MLSS was stored at 4º C for approximately 5 weeks prior to being used for this study. The sludge was allowed to settle and the solids used for inoculum. The settled sludge constituted approximately 20% of the volume (~200 mL) of the MLSS used.
Test medium: Test flasks were prepared using a mineral salts medium defined in EPA Guideline OPPTS 835.3200. Methanol (1 mL per liter) was added per liter of mineral medium. Fifty mL of settled sludge was added per liter of mineral salts medium. Mineral medium plus sludge was prepared 9/7/00, while fresh mineral medium without sludge (abiotic controls) was prepared 8/10/00.
Study design:

Blank Sludge Controls	(mineral medium, inoculum)
Abiotic Controls	(mineral medium, N-EtFOSE alcohol)
Test Substance	(mineral medium, inoculum, N-EtFOSE alcohol)

Test vessels were set in duplicate. Additional quality control samples (blanks) were prepared and analyzed as appropriate.

Test concentrations: 2.856 mg/L.
Incubation condition:
Temperature: 25^{o}C +/- 3^{o}C
Agitation: ~200 rpm
Test vessels: Sterile 125 mL Nalgene polycarbonate culture flasks containing 25 mL of media

Dosing procedure: Test vessels were spiked with 6 µL of an 11,900 mg/L solution of N-EtFOSE alcohol in methanol yielding 2.856 mg/L.
Sampling Frequency: Days 0 and 18.
Analytical method: The day zero test vessels were prepared and immediately placed in a freezer that was maintained at –20^{o}C until analyzed. After 18 days, the test vessels

were removed from the incubator and frozen until final sample preparation by solid phase extraction (SPE). Following thawing, test vessel contents were adjusted to 1% acetic acid and then passed through a conditioned SEP-VAC C18 6cc SPE cartridge. Methanol was then added to the emptied culture flask, shaken vigorously and then passed through the SPE cartridge to extract adsorbed analytes. A second methanol wash was collected separately for analysis to ensure quantitative extraction.

Quantitative analysis was conducted on an HP1100 high performance liquid chromatograph with mass spectrometer detector (HPLC/MSD) system. The MSD was operated in electrospray ionization in negative ion mode using selected ion monitoring (SIM) for quantitation.

In addition to N-EtFOSE alcohol, the additional compounds quantified are specified below. In the case of the compounds that are potassium or ammonia slats, only the concentration of the fluorochemical anion was quantified and reported.

Compound Name
Acronym
Chemical Formula

2-(N-Ethyl Perfluorooctane sulfonamido) acetic acid
N-EtFOSAA
$C_8F_{17}SO_2N(C_2H_5)(CH_2COOH)$

2-(Perfluorooctane sulfonamido) acetic acid
M556
$C_8F_{17}SO_2NH(CH_2COOH)$

N-Ethyl Perfluorooctane sulfonamide
N-EtFOSA
$C_8F_{17}SO_2NH(C_2H_5)$

Perfluorooctane sulfonamide
FOSA
$C_8F_{17}SO_2NH_2$

Perfluorooctane sulfinate, potassium salt
PFOSulfinate
$C_8F_{17}SO_2^- K^+$

Perfluorooctanoate, ammonium salt
PFOA
$C_7F_{15}COO^- NH_4^+$

Perfluorooctane sulfonate potassium salt
PFOS
$C_8F_{17}SO_3^- K^+$

Reference substance: None. When results are compared to the previous 35-day study, the viability of the microbial inoculum is confirmed.

RESULTS

After 18 days, the analytical results[1] demonstrate that when exposed to municipal wastewater treatment sludge, 2.856 mg/L N-EtFOSE alcohol degraded well, with only 15.9% remaining. The major biotransformation product seen was N-EtFOSAA. Minor products were M556, N-EtFOSA, FOSA, PFOS, PFOSulfinate and PFOA. The results were in good agreement with the previous 35-day exposure study. Mass balance for N-EtFOSE alcohol test vessels ranged from 88 to 107%.

CONCLUSIONS

The results of this study confirm those found in the previous 35-day study. Biotransformation of 84.1% of 2.856 mg/L N-EtFOSE alcohol by aerobic microorganisms present in municipal sludge was demonstrated. The major metabolite identified was N-EtFOSAA. Minor metabolites include M556, N-EtFOSA, FOSA, PFOSulfinate, PFOS and PFOA. Mass balance between parent material and measured products was achieved, implying that all significant degradation products of N-EtFOSE alcohol under conditions studied were identified.

DATA QUALITY

Klimisch ranking = 2. The study was conducted as a non-GLP study but with the understanding that good data quality objectives be met.

Determination of analyte recovery from spiked sample matrices was not deemed necessary as they have been determined twice previously at several different concentrations, and in both instances recoveries were near 100% with some exceptions for M556. Methanol and sample (mineral medium) blanks contained no detected target analytes.

Analyses of the blank sludge controls (mineral media plus inoculum) at days 0 and 18 demonstrated that the inoculum source did not contain endogenous concentrations of test substance or its metabolites.

REFERENCES

Biodegradation Study Report, "*The 18-Day Aerobic Biodegradation Study of Perfluoroctanesulfonyl-Based Chemistries"*, Contract Analytical Project ID: CA097, February 23, 2001. Conducted at the request of the 3M Company by Pace Analytical Services, Inc., Minneapolis, MN.

OTHER

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

Last changed: 6/26/01