Task ID**:** 425.015
Task Title**:** Reductive Dehalogenation of Perfluoroalkyl Surfactants in Semiconductor Effluents.
Deliverable**:** Report on the susceptibility of PFOS and related perfluoroalkyl surfactants to microbial reductive dehalogenation.

Summary Abstract:
PFOS and related perfluorinated alkyl surfactants (PFAS) are important components in a number of semiconductor operations. Although more environmentally benign chemistries are under development, commercial alternatives to PFOS for critical photolithography uses are still lacking. To date neither biodegradation or chemical degradation of PFOS under ambient conditions has been observed due to the stability of covalent C─F bonds and the lack of reactive substituents in the molecule. In the quest for PFOS biotransformation, microbial reductive dehalogenation should be considered. Reductive dehalogenation is an important mechanism contributing to the biodegradation of highly chlorinated compounds *(1)*. Certain microorganisms can rapidly utilize the organohalogens as electron acceptors in an energy yielding reaction known as halorespiration. Cometabolic reductive dehalogenation is also known to occur due to the chemical reactivity of common occurring organo-metallic enzyme cofactors present in anaerobes. Bioreactor technology with halorespiring biofilms can convert highly chlorinated solvents such as perchloroethylene at high volumetric rates. Although microbial degradation of various organofluorines compounds is well documented *(2)*, no studies have been published to date considering the biological reductive defluorination of PFOS and related perfluoroalkyl surfactants. The main objective of the research reported here is to investigate the feasibility of microbial reductive dehalogenation for the removal of PFOS and related long chain perfluorinated compounds in semiconductor effluents.

Technical Results and Data:
Microbial degradation of PFOS and perfluorobutane sulfonate (PFBS, a fluorinated derivative proposed as an environmentally-benign alternative to PFOS) was evaluated in anaerobic shaken batch bioassays. The experiments were inoculated with sediments or sludge samples obtained from a variety of sources, including inocula previously exposed to fluorinated compounds (eg. biosolids from wastewater treatment plants receiving PFOS-containing semiconductor wastewaters). Hydrogen gas was supplied as primary electron-donating substrate for the halorespiring microorganisms and it was replenished periodically by flushing the headspace of the bioassays with H_2/CO_2 gas (80/20, v/v). Sludge controls (no fluorinated compound added) and abiotic controls (microorganisms killed by autoclaving) were run in parallel to determine background fluoride concentrations and to account for the loss of fluorinated compounds by mechanisms other than microbial dehalogenation (eg. surfactant removal by sorption to the biomass). Assessment of reductive dehalogenation relied on the monitoring of fluoride release using an ion-selective electrode. Samples of the culture medium were also withdrawn

periodically for the analysis of fluorinated compounds by ion chromatography with suppressed conductivity detection. Samples of the headspace were analyzed by GC-MS to examine the possible formation of volatile fluorinated metabolites. Selected liquid samples were also investigated using ^{19}F-nuclear magnetic resonance (F-NMR, Fig. 1) and liquid chromatography tandem mass spectrometry (LC/MS-MS), two different techniques which can serve, respectively, to detect changes in the fluorine substitution pattern and to identify putative microbial metabolites of the parent compounds.

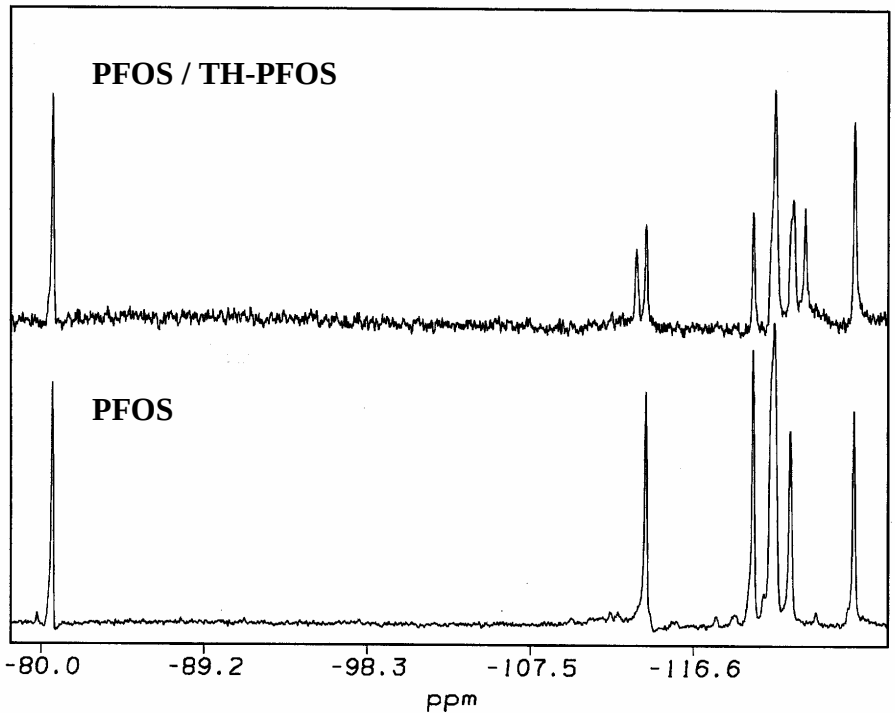

Fig. 1. *F-NMR spectra of solutions containing perfluorooctane sulfonate (PFOS) / tetrahydro-PFOS (TH-PFOS) at 1:1 molar ratio and PFOS alone. The peak at -80 ppm can be assigned to the internal standard.*

The highly oxidized character of perfluorinated compounds is expected to make them more prone to microbial attack in anaerobic reducing environments as compared to aerobic conditions. Nonetheless, no evidence has been obtained that the perfluorinated compounds assayed are biodegraded by any of the inocula tested after more than 12 to 18 months of incubation under anaerobic conditions. These results appear to confirm the outstanding resistance of PFOS to microbial attack. Literature studies have also shown that PFOS is not degraded under aerobic or sulfate-reducing conditions.

It is interesting to note that PFBS, a compound proposed as an alternative to PFOS, was also found to be highly resistant to microbial degradation under reductive conditions. While the shorter alkyl chain is known to reduce the tendency of PFBS to accumulate in biological matrices, the perfluorinated compound appears to share its persistent character with PFOS.

Technical PFOS is a mixture of linear and branched structural isomers, with the latter making up 20 to 30% of the total mass. Work conducted in the frame of this study has shown that branched PFOS isomers are more susceptible to biomimetic reductive dehalogenation than the linear PFOS isomer. These results suggest that branching may facilitate the microbial degradation of perfluorinated compounds. Future efforts will assess the microbial transformation of branched PFOS isomers in anaerobic environments.

References

1. **Smidt, H., de Vos, W. M.** Anaerobic microbial dehalogenation. *Annu. Rev. Microbiol.* **58**, 43-73 (2004).
2. **Natarajan, R. et al.** Microbial cleavage of C-F bond. *J. Fluorine Chem.* **126,** 425-436 (2005).