APPLICATION OF MICROTITER-BASED OXYGEN SENSORS TO ASSESS MICROBIAL COMMUNITIES

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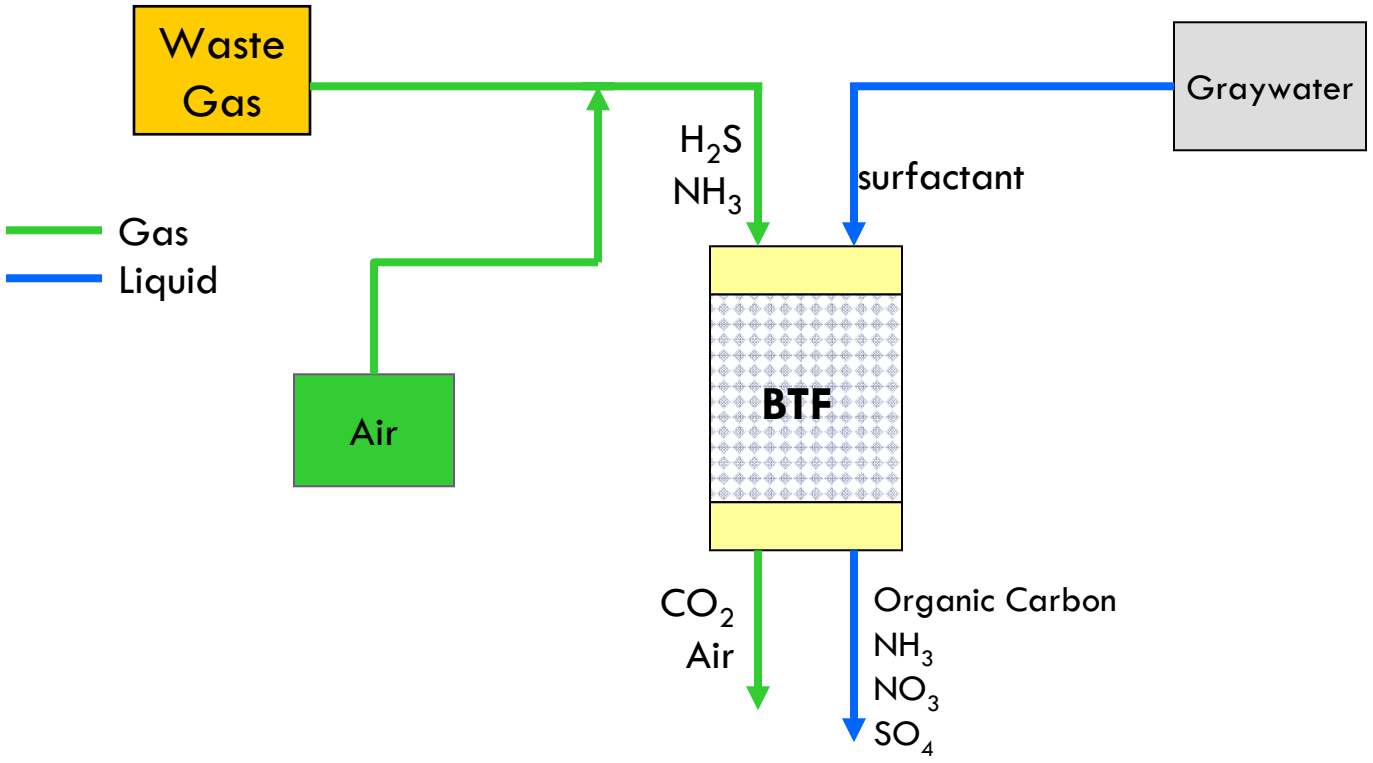
Overview

- Project description
- Description of oxygen sensor
- Applications
  - Surfactant biodegradation pathway
  - Spatial variation of functional groups of microorganisms in a biotrickling filter

Biotrickling Filter for Graywater and Waste Gas Treatment



Biological Conversions

- Surfactant removal from liquid phase (Pert Plus Kids)

$$C_{18}H_{37}ONCH_2NCH_2OHCH_2COONa + 33O_2 \rightarrow 24CO_2 + 24H_20$$

$$2CH_3(CH_2)_{10}CH_2(OCH_2CH_2)_3OSO_3Na + 51O_2 \rightarrow 36CO_2 + 37H_2O$$

- NH_3 gas solubilized to liquid phase where nitrification will occur

$$2NH_3 + 6O_2 \rightarrow 2NO_3 + 6H^+$$

- H_2S oxidation to liquid SO_4

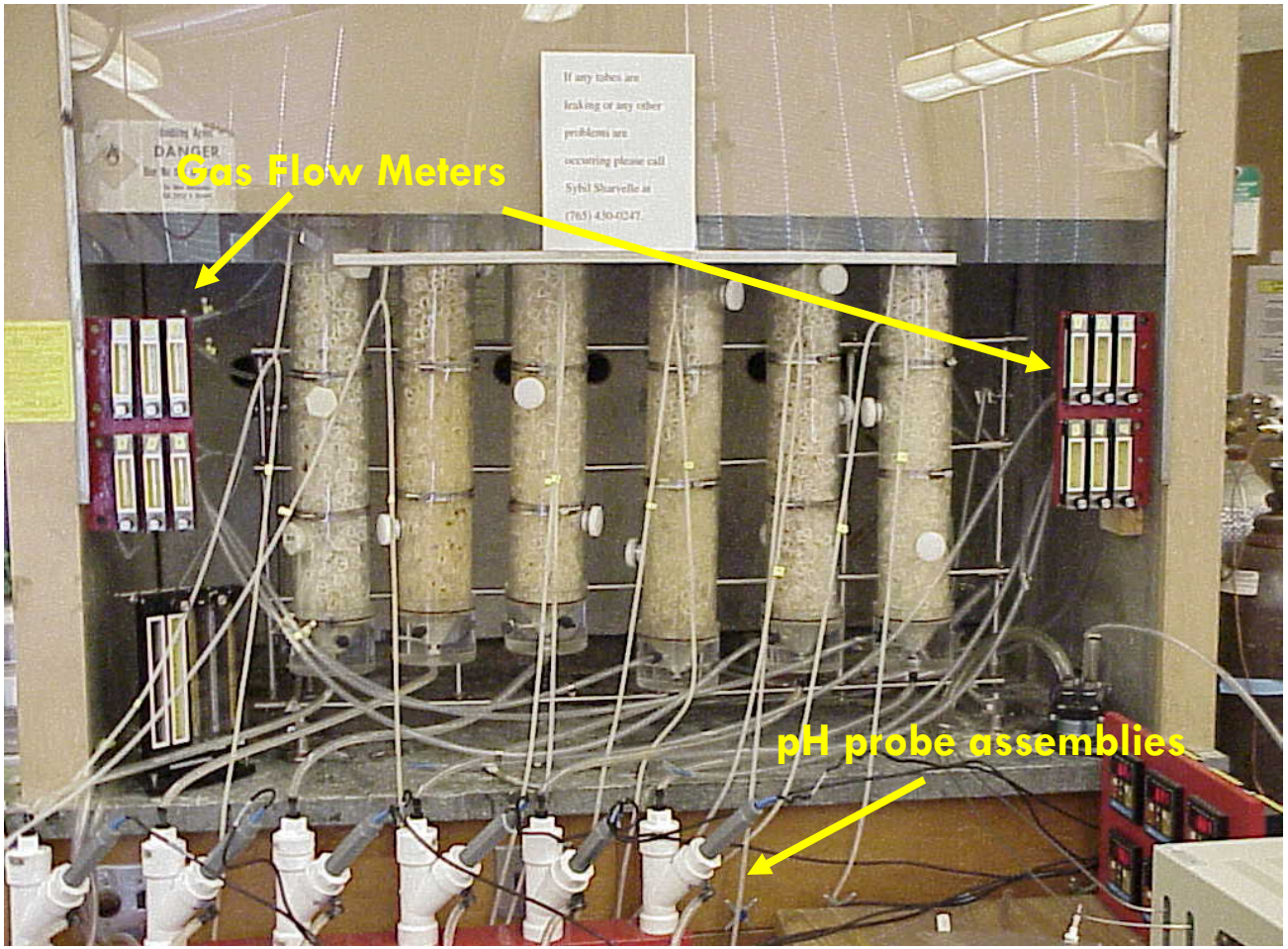
$$H_2S + 2O_2 \rightarrow SO_4 + 2H^+$$

Graywater Simulant

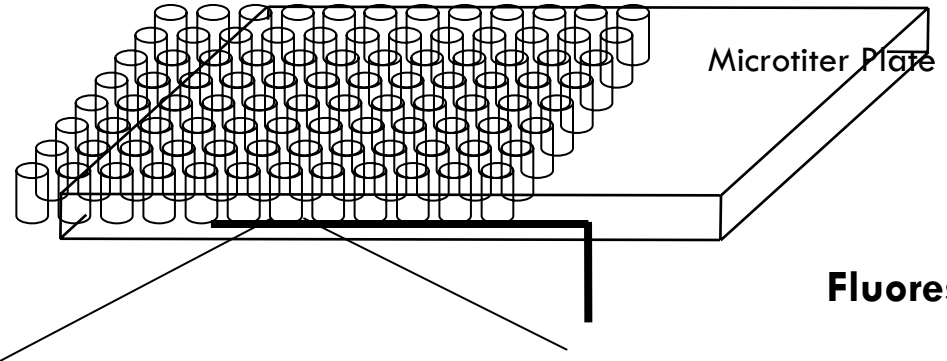
Constituent	*Concentration (mg/L)*	*NASA Potable Water Standard (mg/L)*
Surfactants:		
sodium laureth sulfate	73	0.5[a]
disodium cocoamphodiacetate	62	0.5[a]
polyalcohol ethoxylate	69	0.5[a]
Inorganics:		
sodium	74	-
chloride	55	200
potassium	51	-
bicarbonate	9	-
ammonium	5	0.5
Organics:		
urea	87	0.5
lactic acid	58	0.5
acetic acid	38	0.5
formic acid	22	0.5
propionic acid	20	0.5

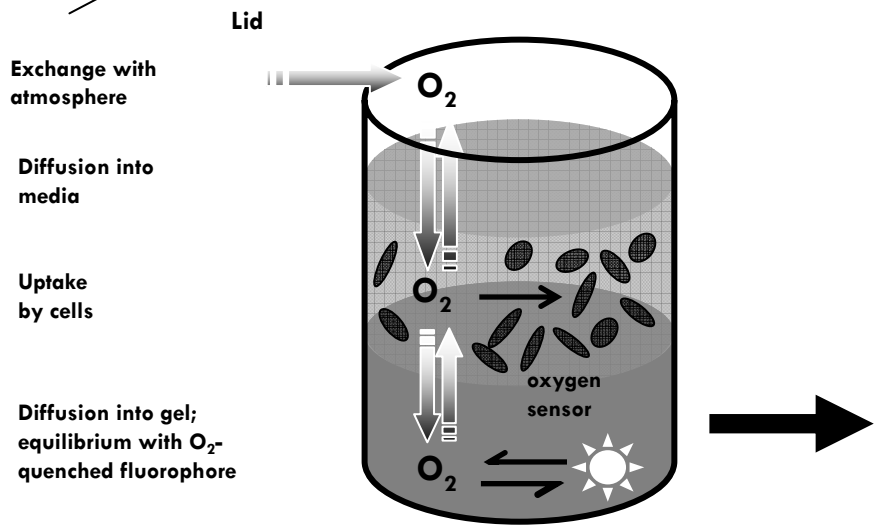
a – EPA Standard for foaming constituents

Six Replicate Bench Scale Reactors



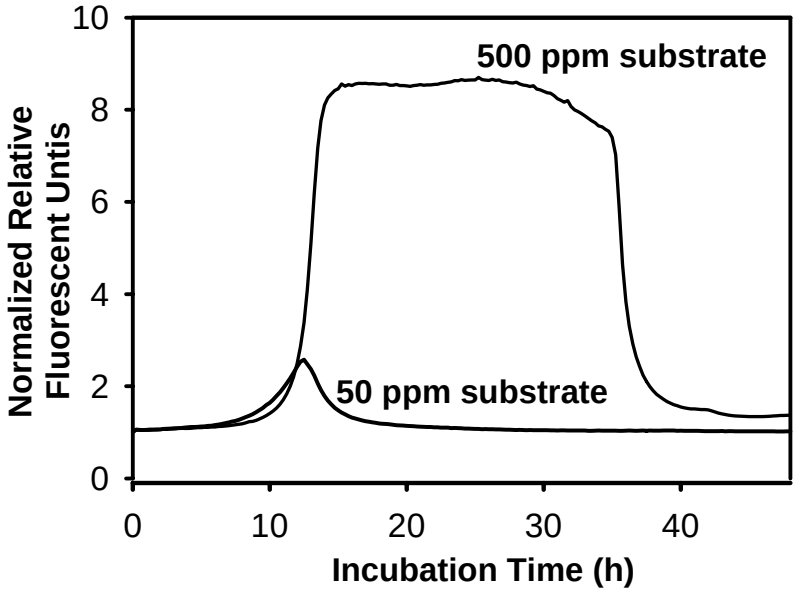
Microtiter Based Oxygen Sensors





Individual Well Layout

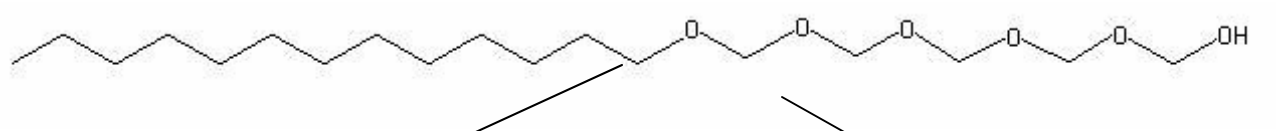
Fluorescence Response



Surfactant Biodegradation Pathways

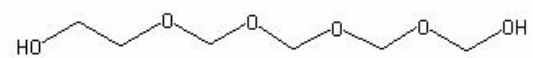
PAE (polyalcohol ethoxylate)

(polyalcohol ethoxylate)

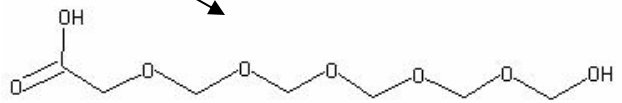


hydrophobe-phile scission

alkyl chain degradation

(alkyl chain) + 

(PEG chain)



(carboxylalkyl-PEG)

$HO(CH_2CH_2O)_nCH_2COOH$

(carboxilic acids)

acidic C_2 units

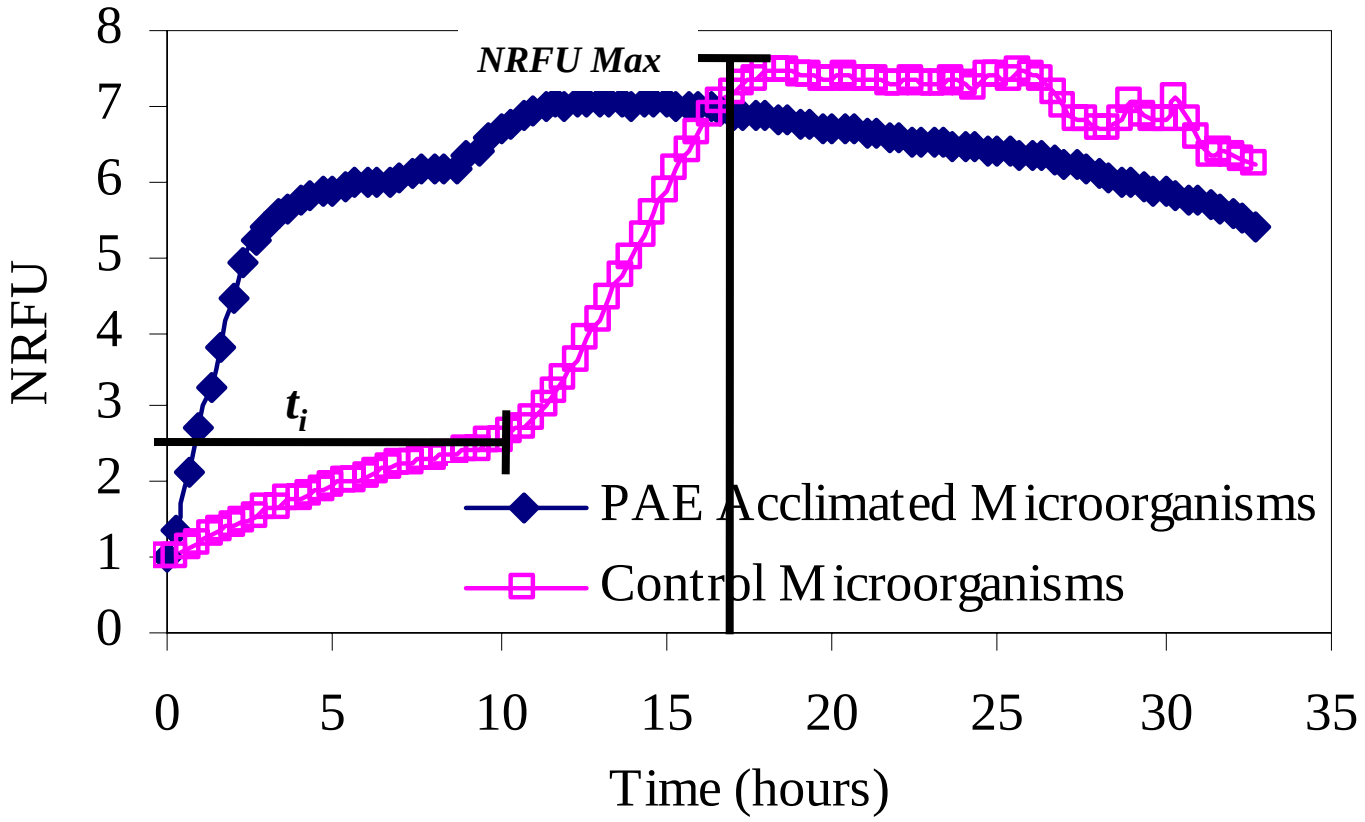
CO_2 + biomass

(Steber and Wierich, 1985)

Preferential Uptake of Metabolites

- Batch Assays
  - Wastewater microbial consortium supplied with PAE as sole carbon source for at least 3 weeks
  - Control population cultured in tryptic soy broth
- Previously identified metabolites of PAE biodegration supplied to each culture in microtiter plate wells
  - More rapid uptake or a higher response indicates previous exposure to that metabolite
    - Identify likely metabolites

Response to PAE



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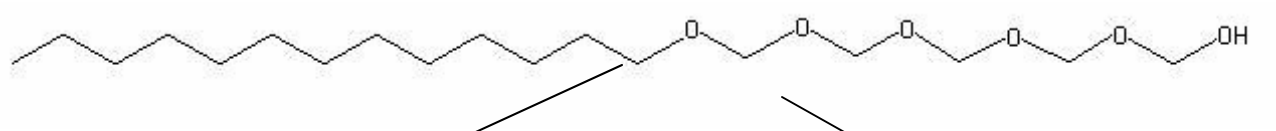

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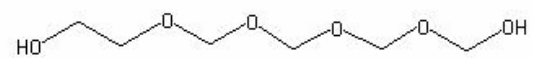
(alkyl chain) + 

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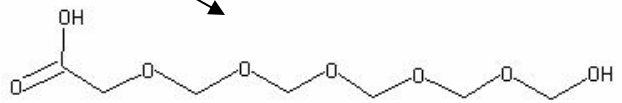

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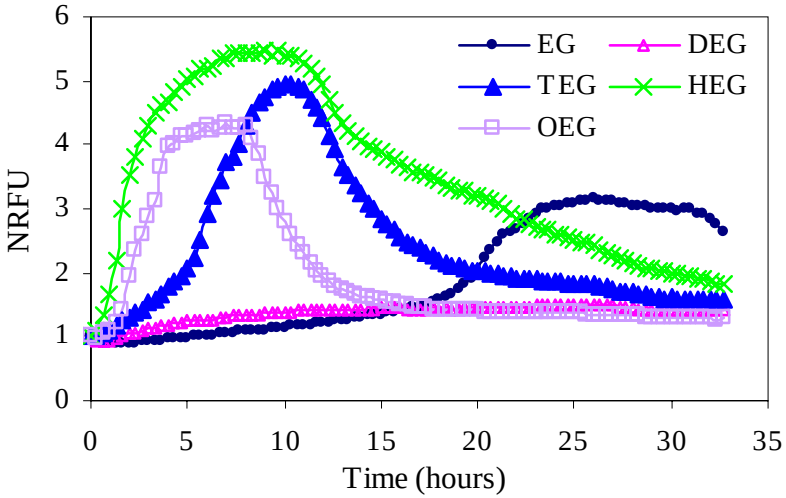
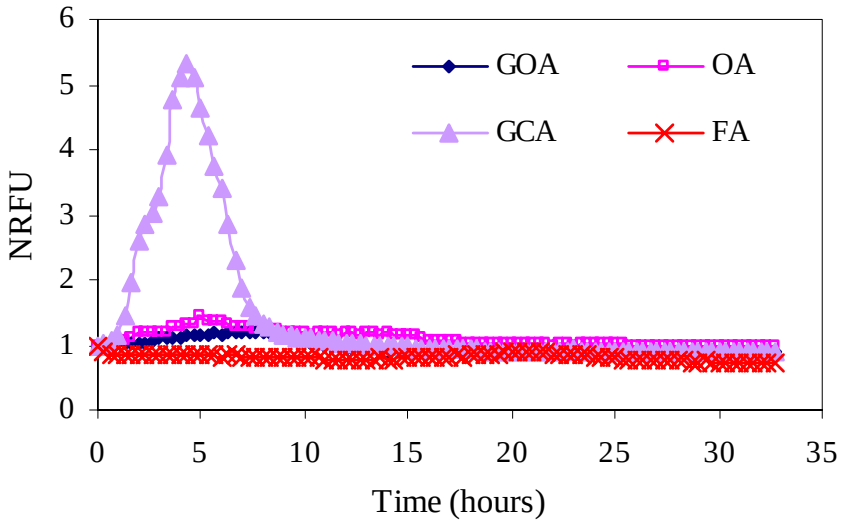
(Steber and Wierich, 1985)

Response of PAE Acclimated Micororganisms to Various Substrates





EG – ethylene glycol
DEG – diethylene glycol
TEG – triethylene glycol
HEG – hexaethylene glycol
OEG – octaethylene glycol

GOA – glyoxalic acid
OA – oxalic acid
GCA – glycolic acid
FA – formic acid

PEG Biodegradation

$HO(CH_2CH_2O)_nCH_2CH_2OH$

↓

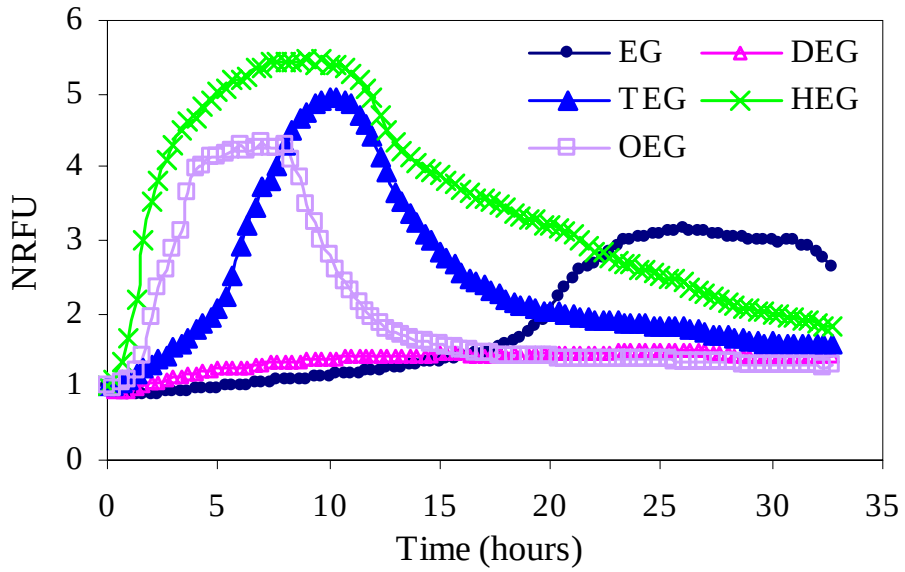
$HO(CH_2CH_2O)_nCH_2CH_2O$

↓

$HO(CH_2CH_2O)_nCH_2COOH$

↓

$HO(CH_2CH_2O)_{n-1}CH_2CH_2OH$



DEG and EG may accumulate

Summary of PAE Biodegradation Assays

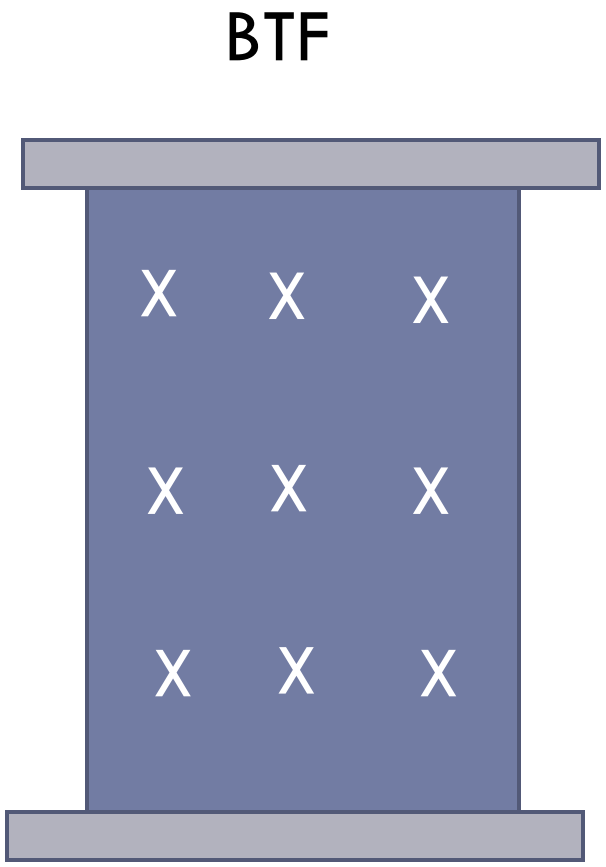
- Dominant pathway in studied systems is hydrophobe-phile scission as indicated by the formation of PEGs
- EG and DEG may accumulate
- Could be applied to bioreactor samples
  - Preliminary screening without intensive analytical methods development

Spatial Variation of Functional Groups in Biotrickling Filters

Experiment Description

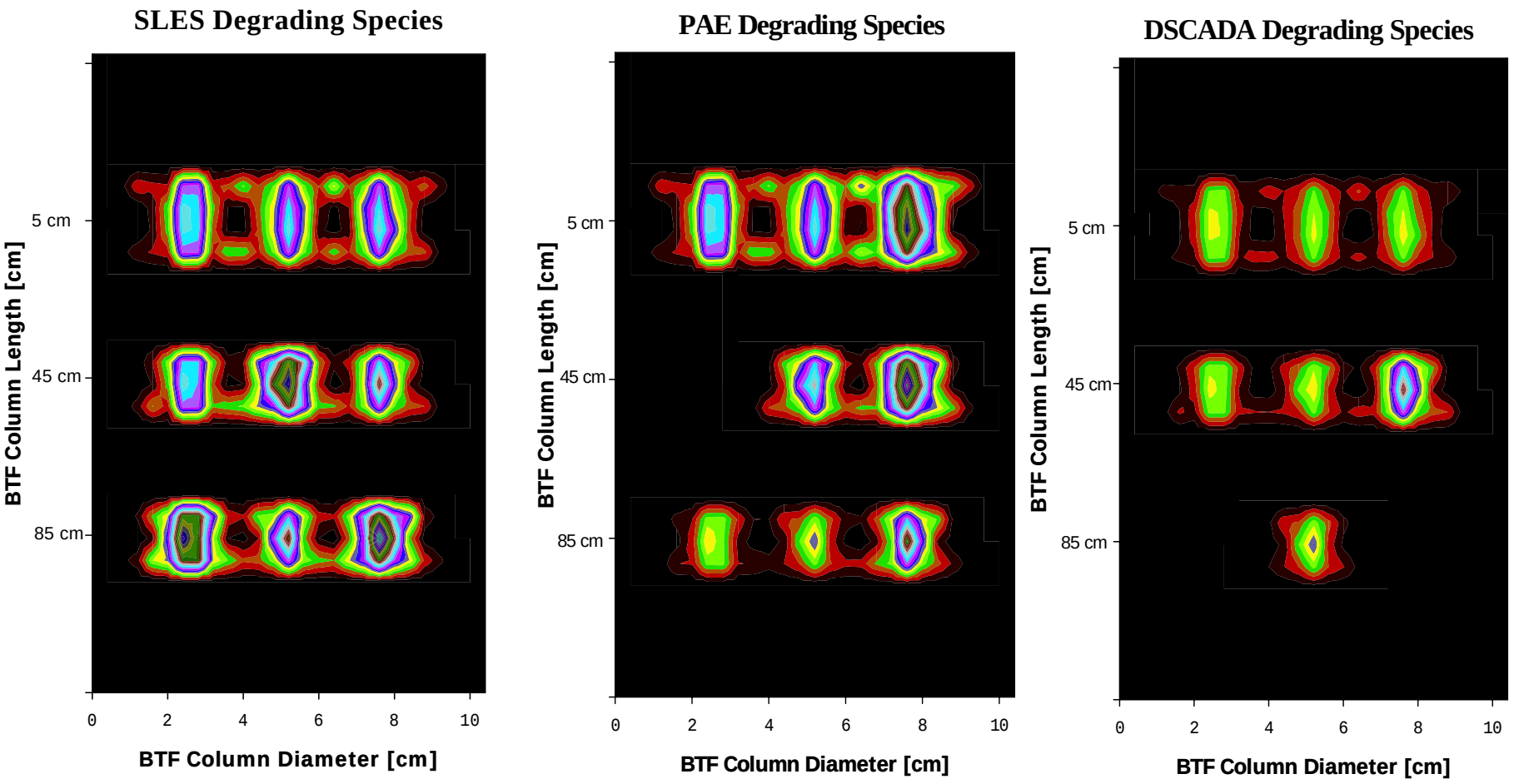
- Coexistence of diverse microbial community (heterotrophs and autotrophs)
  - Biodegradation of surfactants
  - $NH_3 \rightarrow NO_3$
  - $H_2S \rightarrow SO_4$
- Biological samples scraped with scalpel from biotrickling filter packing material
  - Placed in microtiter plate wells
- Surfactants, ammonia and sulfide supplied to sample wells

Reactor Sampling

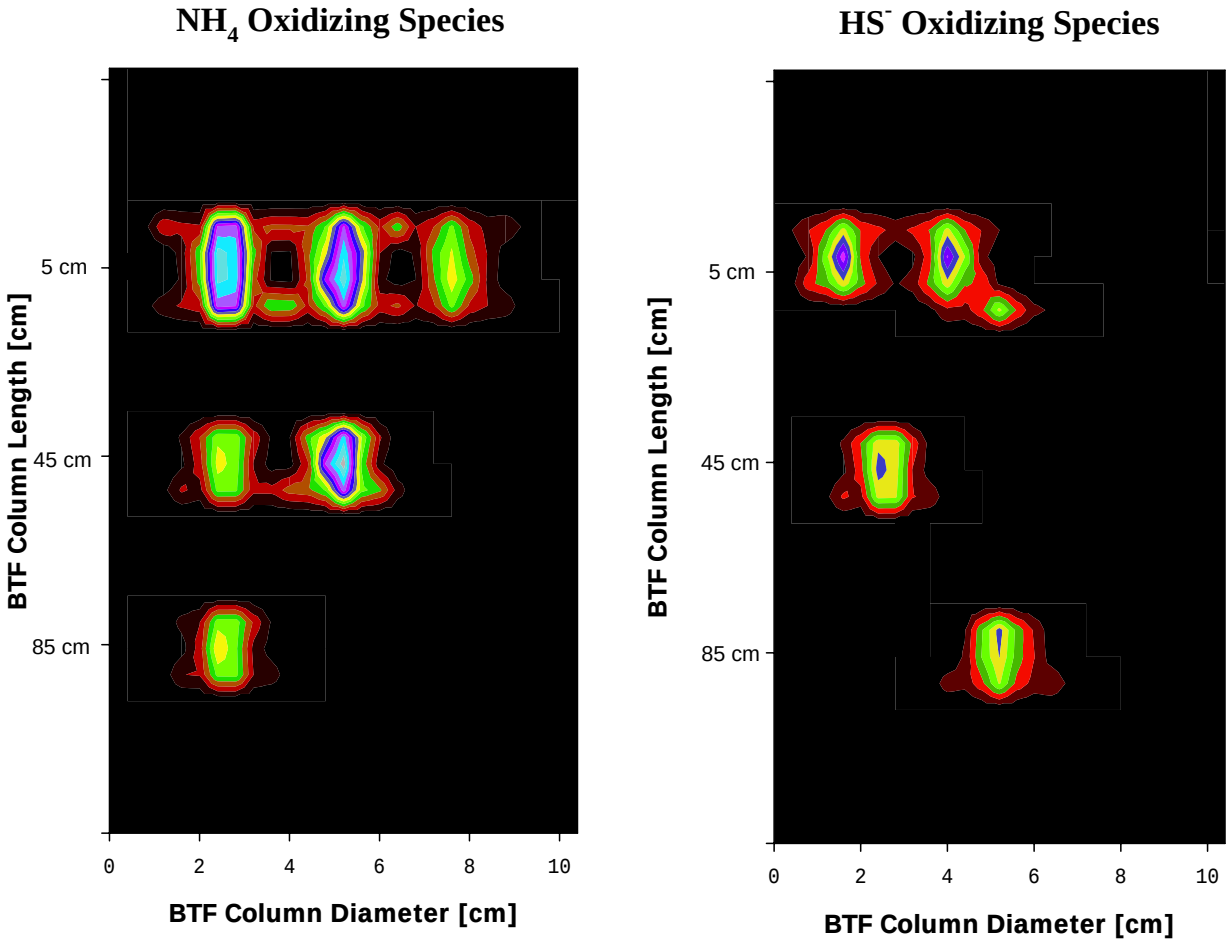


- 9 samples collected in each replicate reactor
- 3 replicate reactors

Gaussian Distributions



Gaussian Distributions



Summary of Spatial Variation Experiments

- Ammonia and sulfide oxidizing bacteria capable of coexisting with surfactant degraders in biotrickling filter
- DSCADA degraders not as well established as other surfactant degraders
- Ammonia and sulfide oxidizing bacteria located predominately at top of BTFs where contaminated gas entered

Overall Summary

- Oxygen sensors are a useful tool to elucidate information about biological processes in engineered and natural systems
  - Rapid analysis of a large number of samples

Questions