

REMTECH
Europe

APPLICATION OF BIO-TRAPS[®] FOR SITE DIAGNOSTICS

CLAUDIO SANDRONE – BAW S.R.L.

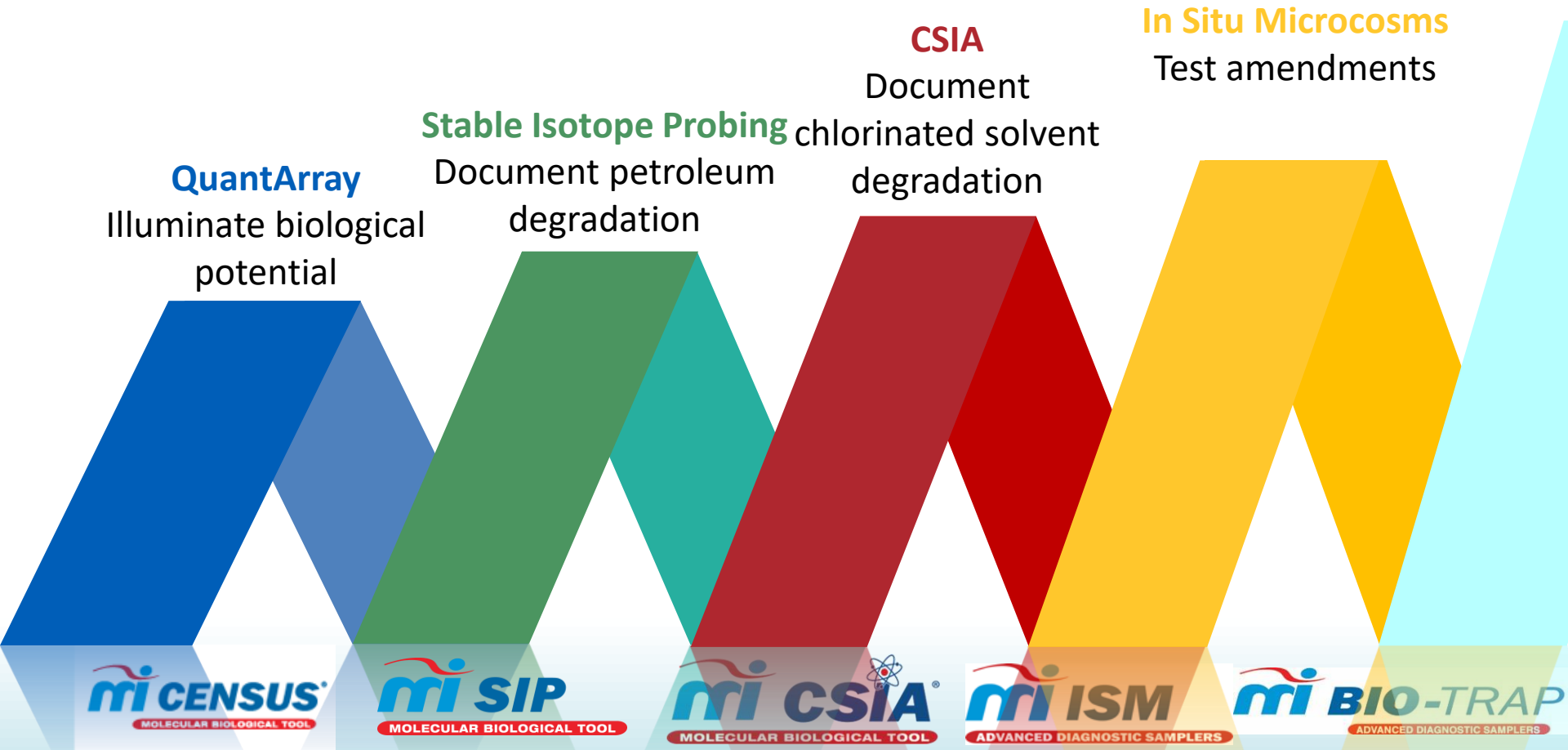
INNOVATIVE CHARACTERIZATION TECHNOLOGIES

18 September 2019

RemTech Expo 2019 (18, 19, 20 September) FerraraFiere

www.remtechexpo.com

27 YEARS OF INNOVATION



Bio-Trap[®]
Samplers

In Situ
Microcosms

Stable Isotope
Probing

Case
Studies

Database



www.microbe.com

Bio-Trap Samplers[®]



What is a Bio-Trap[®]?

Passive sampling tool for microbes

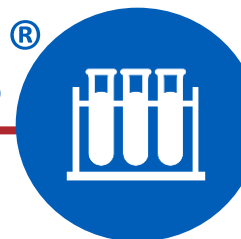
Collects **active** microorganisms

Integrated sample vs. “snapshot”

Use with any of the molecular tools

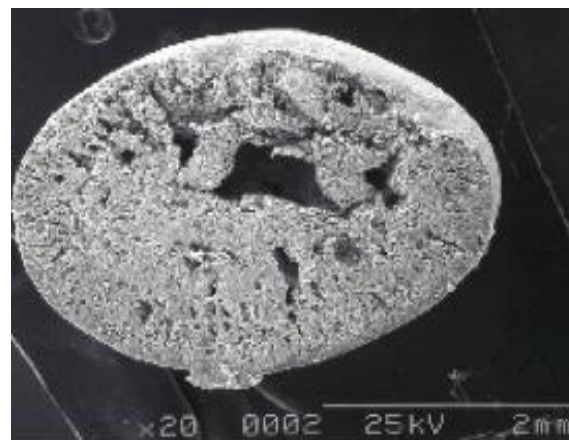


Bio-Trap Samplers[®]



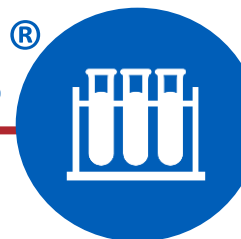
How do Bio-Traps work?

- 3-4 mm in diameter
- 25% Nomex and 75% PAC
- 74% porosity
- 600 m² of surface area/g
- Heat sterilized at 270°C
- Colonized by native microbes



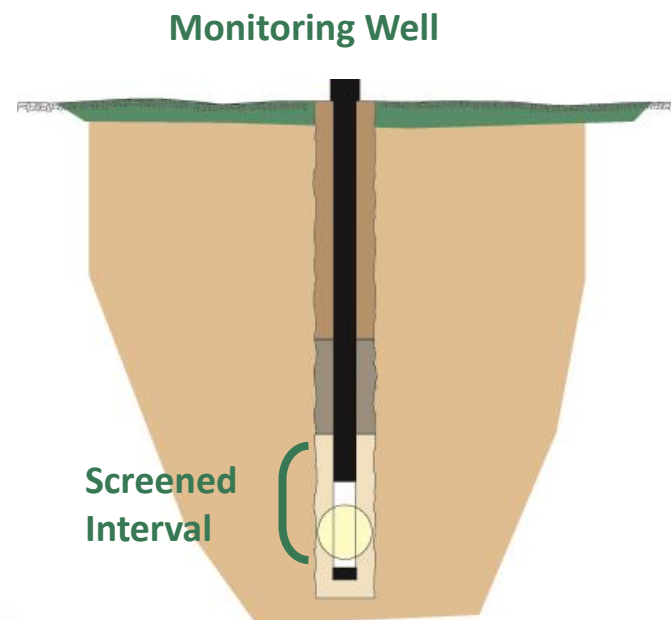
mi.

Bio-Trap Samplers[®]



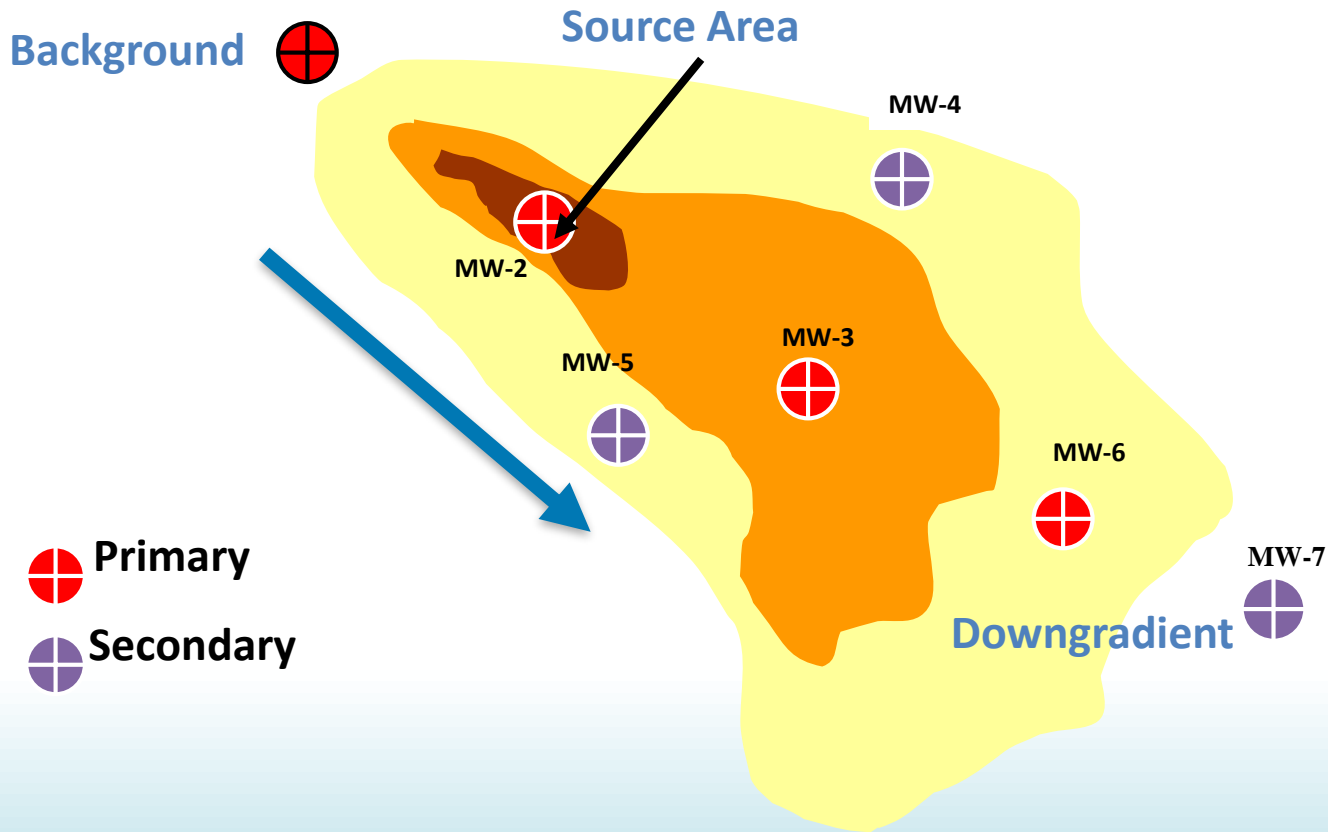
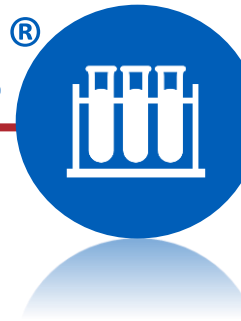
How are they deployed?

- Purge monitoring well
- Suspend from top of casing
- Deploy within the screened interval
 - At depth of interest
- If water fluctuates, suspend from a float



mi.

Bio-Trap Samplers[®]



In Situ Microcosms



What treatment should be selected?

Each ISM unit represents a treatment option

MNA

BioStim

BioAug

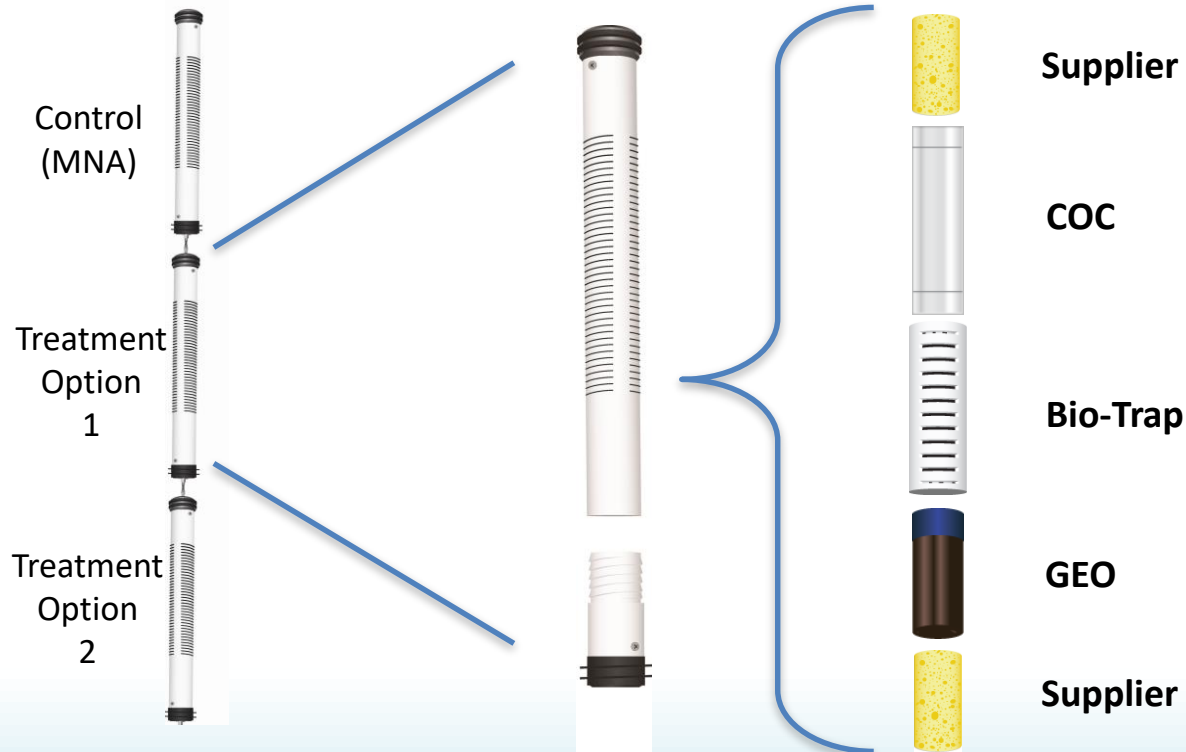
Each unit contains a set of passive samplers

Deployment for 60-90 days

Recovered and shipped to the lab for analysis



In Situ Microcosms





CHEMISTRY



GEOCHEMISTRY



MICROBIOLOGY

In Situ Microcosms



Electron Donors

- Vegetable oil
- Molasses
- HRC
- EOS
- Lactate
- And more

Electron Acceptors

- Oxygen (PermeOx, ORC)
- Nitrate
- Iron
- Sulfate

Stable Isotope Compounds (^{13}C)

- Benzene
- Toluene
- p-Xylene
- MTBE
- TBA
- Naphthalene
- Chlorobenzene
- And more

Case Study

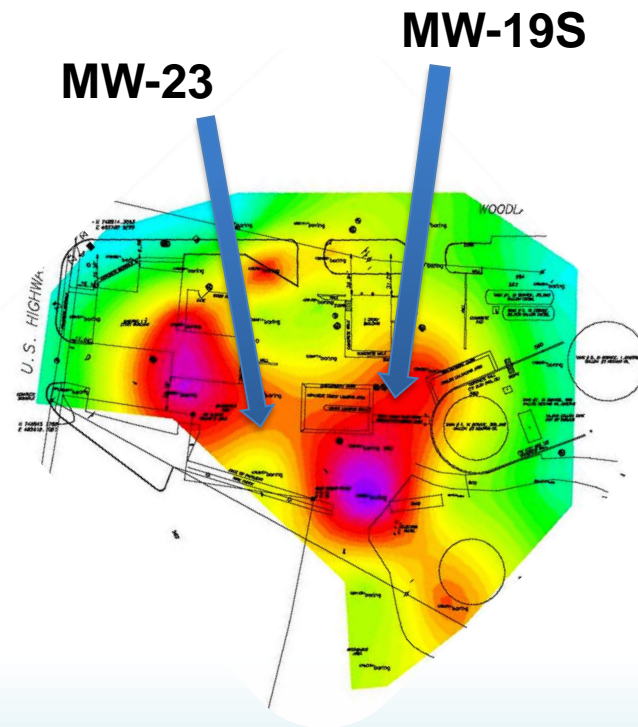


Will addition of sulfate as an electron acceptor stimulate anaerobic BTEX biodegradation?

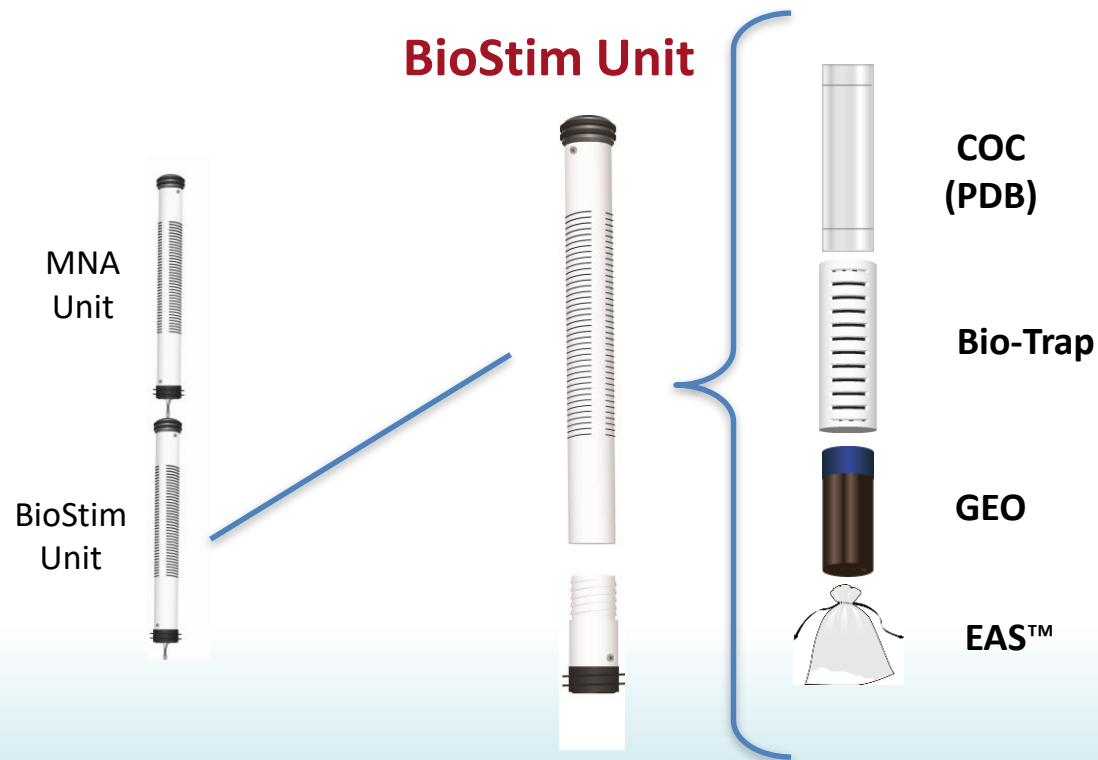
ISM deployed in two monitoring wells

Each assembly consisted of an MNA unit and a BioStim unit amended with EAS™

Following a 60 day in well deployment period, ISM units were recovered for analysis



Case Study

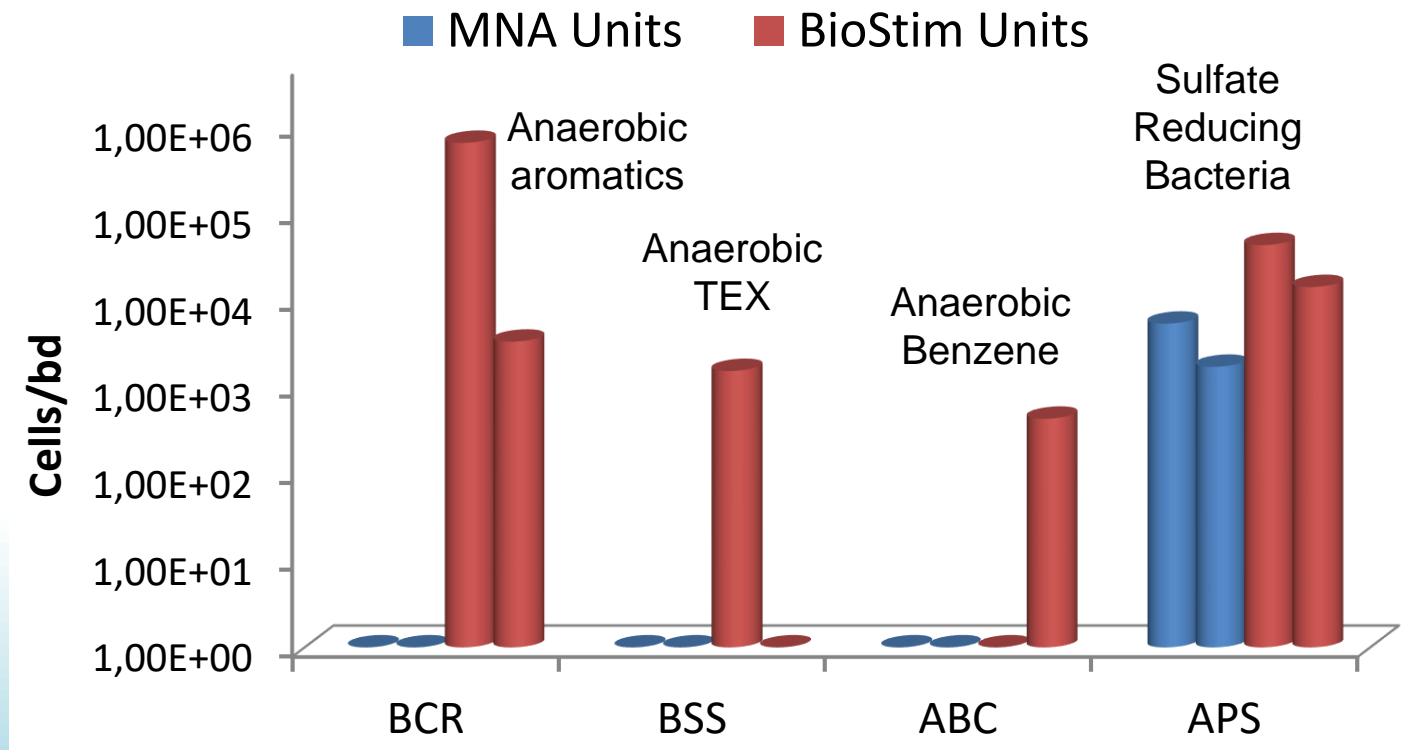


mi.

Case Study



Anaerobic BTEX – MNA vs BioStim Units



mi.

Stable Isotope Probing



99% ^{13}C

Specially produced “heavy” compounds

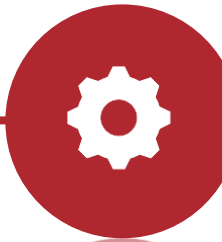
- Natural compounds are 99% ^{12}C
- Same characteristic as original
- Behave same as the natural compound

Tracer

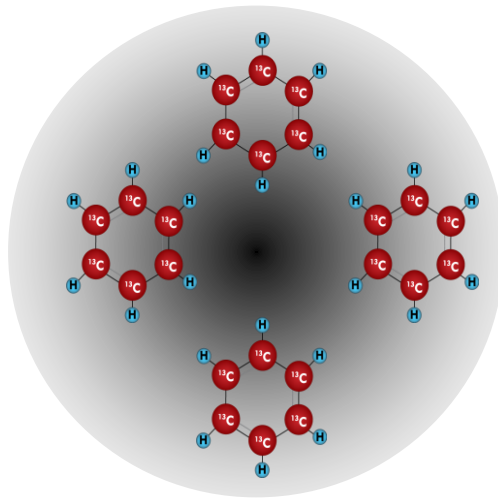
Used as a “tracer” to increase our understanding of the contaminant fate



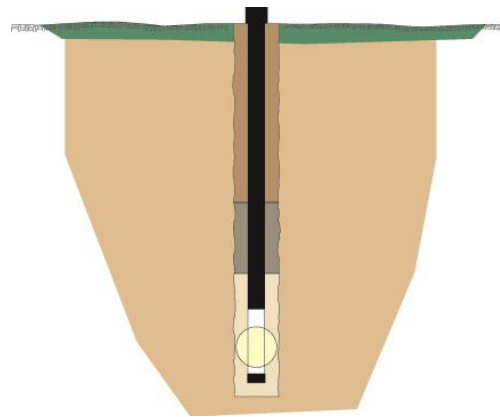
Stable Isotope Probing



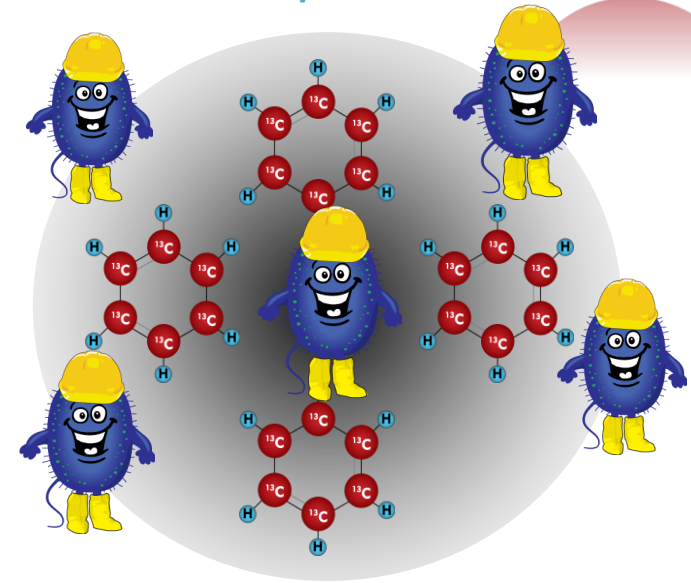
Bio-Sep Bead



Monitoring Well



Bio-Sep Bead



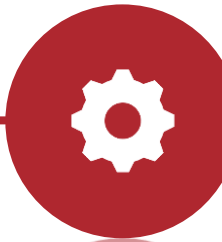
Native microbes colonize the beads within the trap



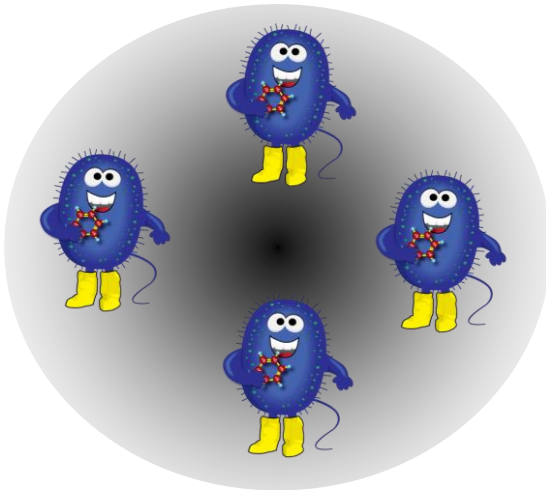
^{13}C compound is loaded onto the beads at the lab

Bio-Traps are deployed into the monitoring well for 30-60 days

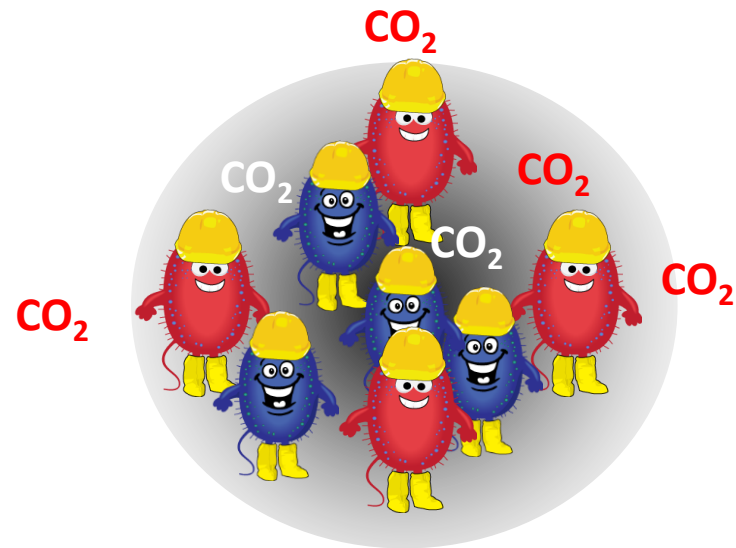
Stable Isotope Probing



Bio-Sep Bead



Some of the native microbes can utilize the ^{13}C compound on the beads



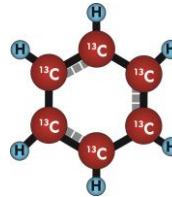
^{13}C is incorporated into new cells growing within the beads or into CO_2 being produced

Stable Isotope Probing



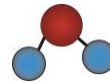
Bio-Trap SIP Analysis

Residual ^{13}C -Compound



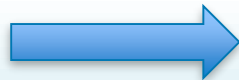
Local relative rate

$^{13}\text{C}/^{12}\text{C}$ Dissolved Inorganic Carbon



Mineralization
(C for energy)

$^{13}\text{C}/^{12}\text{C}$ of Biomarkers



PLFA
DNA
RNA

Metabolism
(C for growth)

CASE STUDY #1

Industrial Site in NJ

Impacted by finishing products (paints and coatings)

Leaking UST

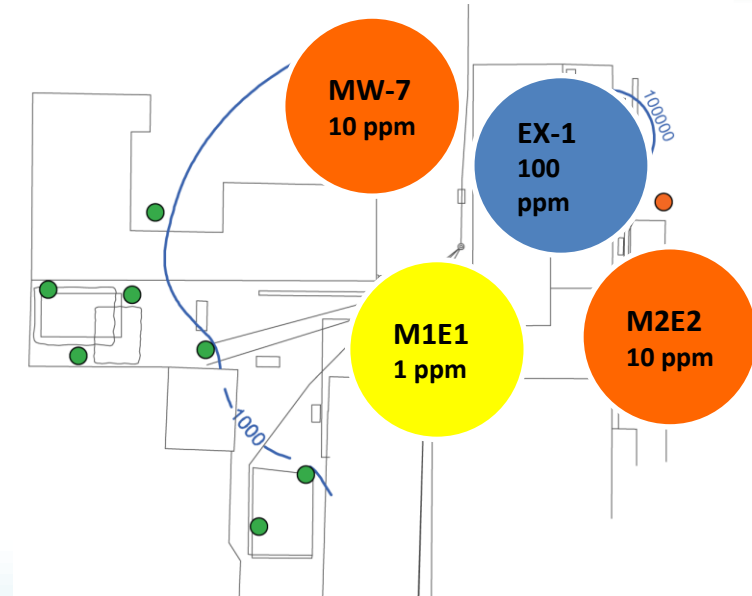
p-Xylene



SIP Case Study #1



- Bio-Traps loaded with ^{13}C p-xylene
- Deployed for 30 days in locations with varied concentration
 - 100, 10, and 1 ppm
- M2E2 was a control with no oxygen added
- Analyzed for SIP

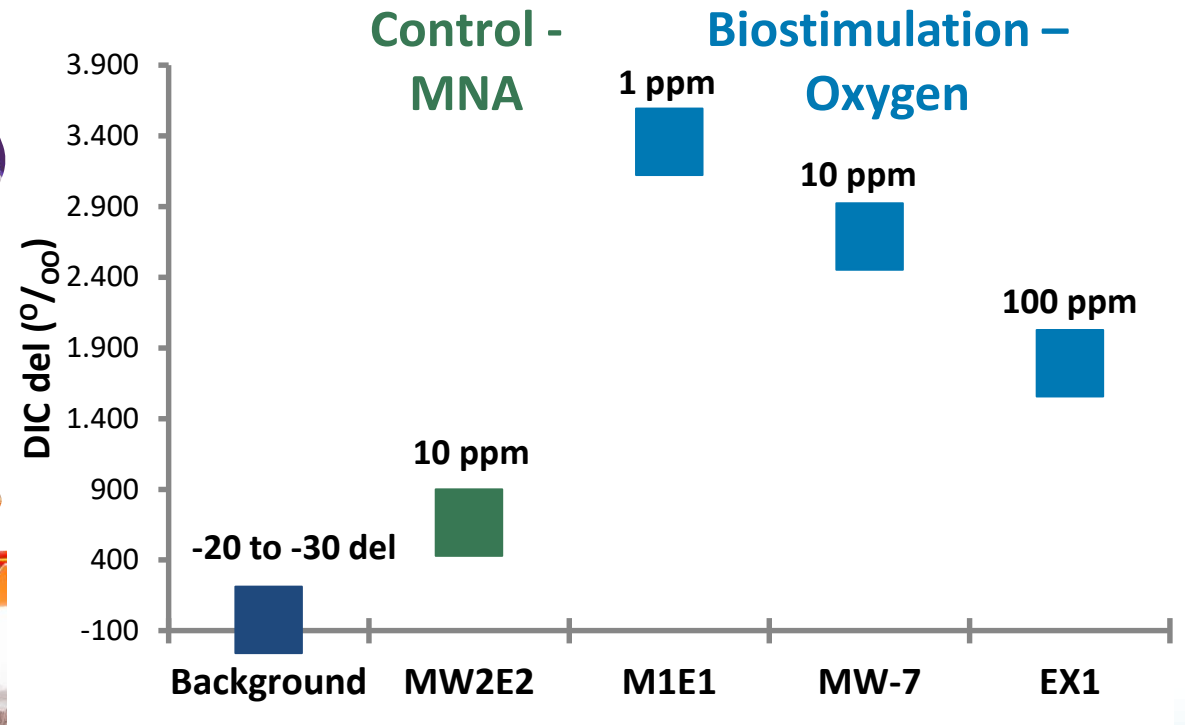




Bio-Trap Results – Relative Rate

Sample	Pre (mg/bd)	Post (mg/bd)	% Loss
<u>Control – MNA</u>			
M2E2 – 10 ppm	1.40	1.31	6 %
<u>Biostimulation – Oxygen</u>			
M1E1 – 1 ppm	1.40	0.84	40 %
MW-7 – 10 ppm	1.40	1.12	20 %
EX-1 – 100 ppm	1.40	1.17	16 %

Bio-Trap Results - Respiration



Bio-Trap Results - Metabolism

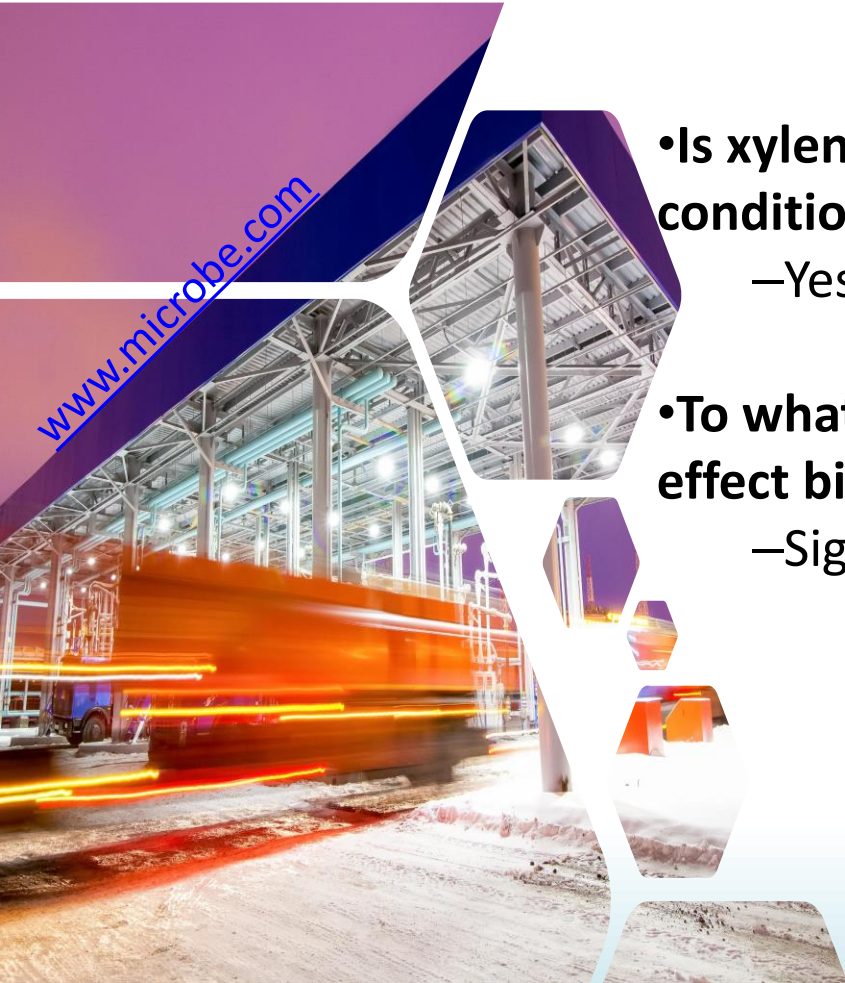
$^{13}\text{C}/^{12}\text{C}$ of Biomarkers

Biomass (cells/bead)

Del Values

Sample	Total	13C		Average	Minimum	Maximum
		Enriched	%			
<u>Control - MNA</u>						
M2E2	3.27E+05	2.15E+03	1%	+48	-50	+547
<u>Biostimulation – Oxygen</u>						
M1E1	2.88E+07	2.14E+06	7%	+6,288	+1,009	+10,764
MW-7	2.00E+07	6.24E+05	3 %	+1,624	+348	+3,878
EX1	6.77E+07	2.17E+06	3%	+1,739	+619	+3,521





• **Is xylene being degraded under the current conditions?**

– Yes see results of the control trap in M2E2

• **To what extent would biostimulation with oxygen effect biodegradation of xylene?**

– Significant stimulation

- Total biomass increased (large increases in PLFA with O_2)
- Higher levels of ^{13}C enrichment observed in PLFA
- Strong evidence of microbial respiration
- Relative rates faster in the wells receiving oxygen

MI DATABASE

Context. Driving
innovation.

MI Database



We use Google as a resource to learn more about a subject.

Use the MI database as a resource to add context to your data.



Over 40,000 samples from sites around the world

THANKS FOR THE ATTENTION,

Dr. Claudio Sandrone

BAW s.r.l. – Saluzzo (CN)

Ph: 334-5715645 – 0175/86642

e-mail: claudiosandrone@baw-env.it