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Universitat Autònoma de Barcelona

14th International HCH and Pesticides Forum

LAB STUDIES LEADING TO DECISION-MAKING FOR *IN SITU* BIOREMEDIATION OF ORGANOHALIDES

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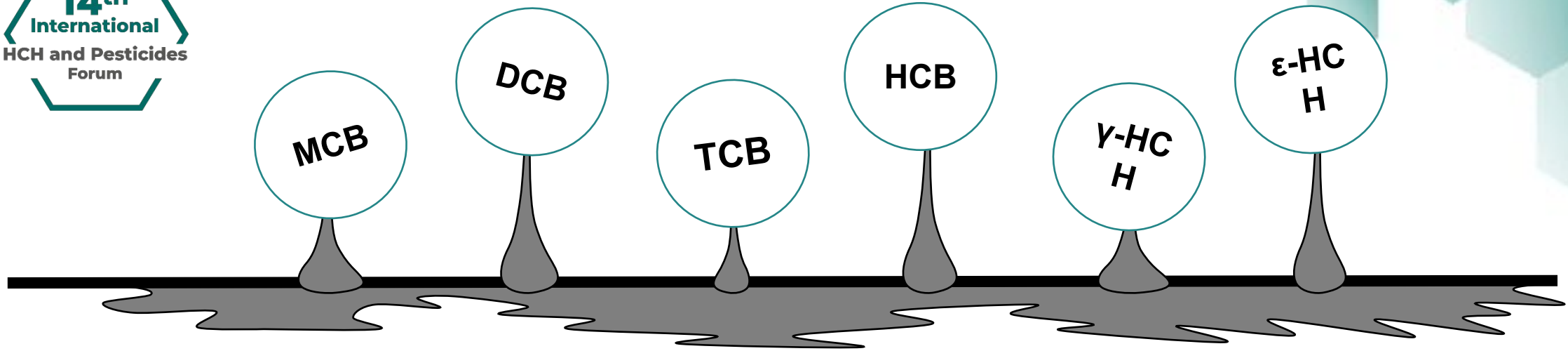
Introduction



Physical barriers

Soil-vapor extraction (SVE)

Permeable reactive barriers (PRB)

Excavation

Pump & Treat (P&T)

In-situ thermal desorption (ISTD)

Surfactants

In-situ chemical reduction (ISCR)

Dual-phase extraction (DPE)

In-situ chemical oxidation (ISCO)

Bioremediation

Biology to the rescue!

Anaerobic bacteria

Aerobic bacteria

- Energy conservation
- Carbon source
- Co-metabolic

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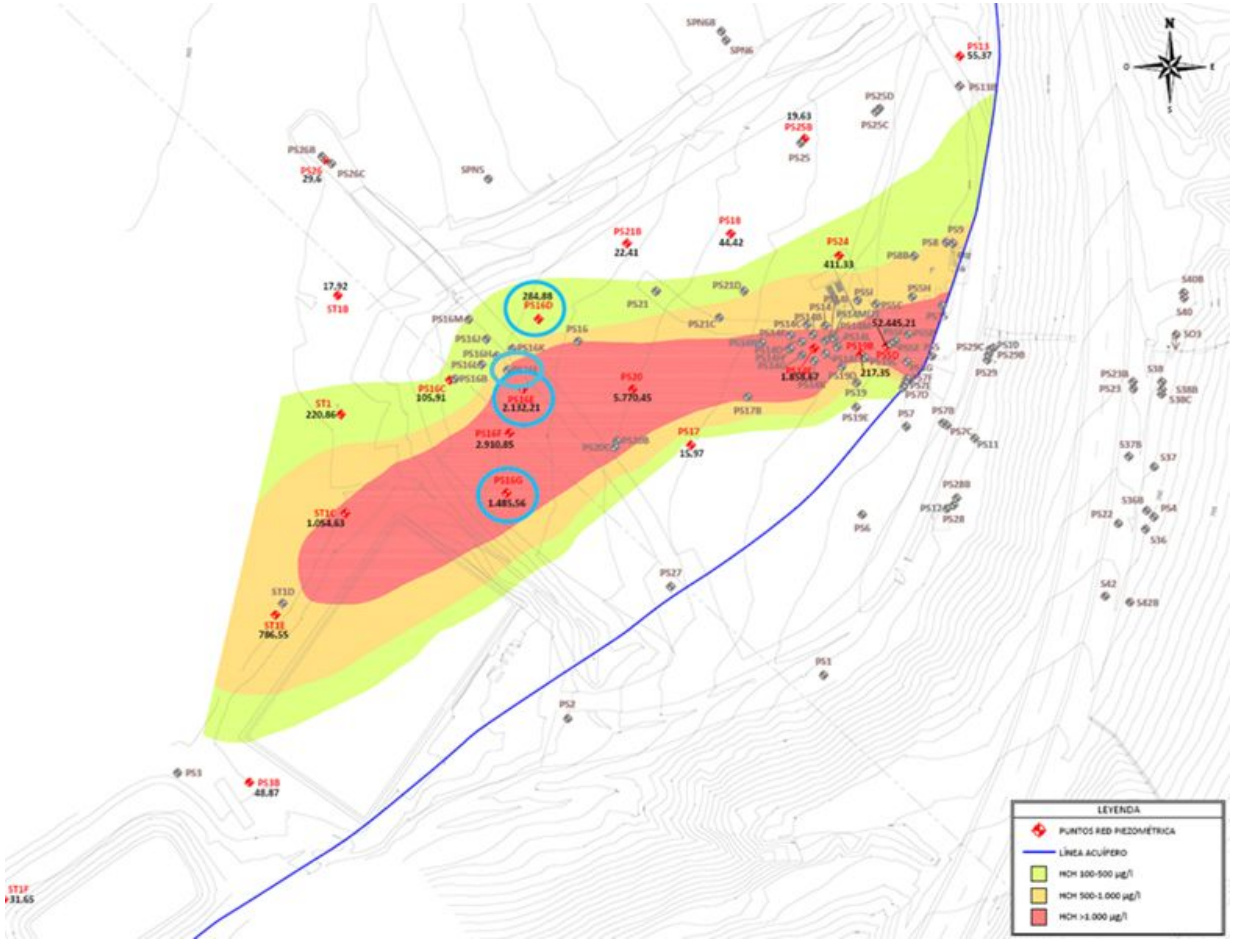
To study the feasibility of bioremediation in the polluted groundwater of Sardas (Spain) by testing different conditions to select the best treatment to apply at a pilot scale



Parameters	Sampling wells		
	PS16E	PS16D	PS16G
Depth (m)	13	12	12
Temperature (°C)	13,23	12,67	13,03
pH	6,76	6,84	6,91
Redox potential (mV)	-53,4	35,7	116,7
DO (mg/L)	2,99	2,96	5,46
Sum-HCH (µg/L)	1968,1	764	50,7
1,3,5-TCB (µg/L)	16,3	15,2	0,5
1,2,4-TCB (µg/L)	703,9	259,1	5,7
1,2,3-TCB (µg/L)	116,9	37,9	2,1
1,3-DCB (µg/L)	99,4	286,2	5,5
1,4-DCB (µg/L)	1028	1418	23,4
1,2-DCB (µg/L)	997,2	909,9	20,7
MCB (µg/L)	6620,8	8255,8	191,2
Benzene (µg/L)	216	140	4,4

Set up of the first microcosms

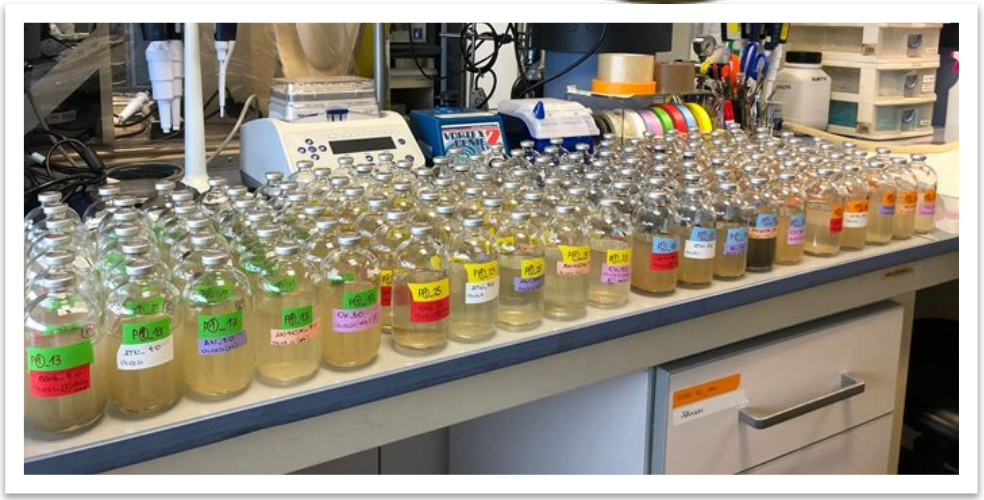
To determine the biodegradation potential and under which conditions

3 Wells:

PS16E → 13°C and 25°C

PS16D → 13°C

PS16G → 13°C



Natural Attenuation (ATN) → 85 d

Aerobic biostimulation (OX) → 85 d

Anaerobic biostimulation (AN) → 85 d

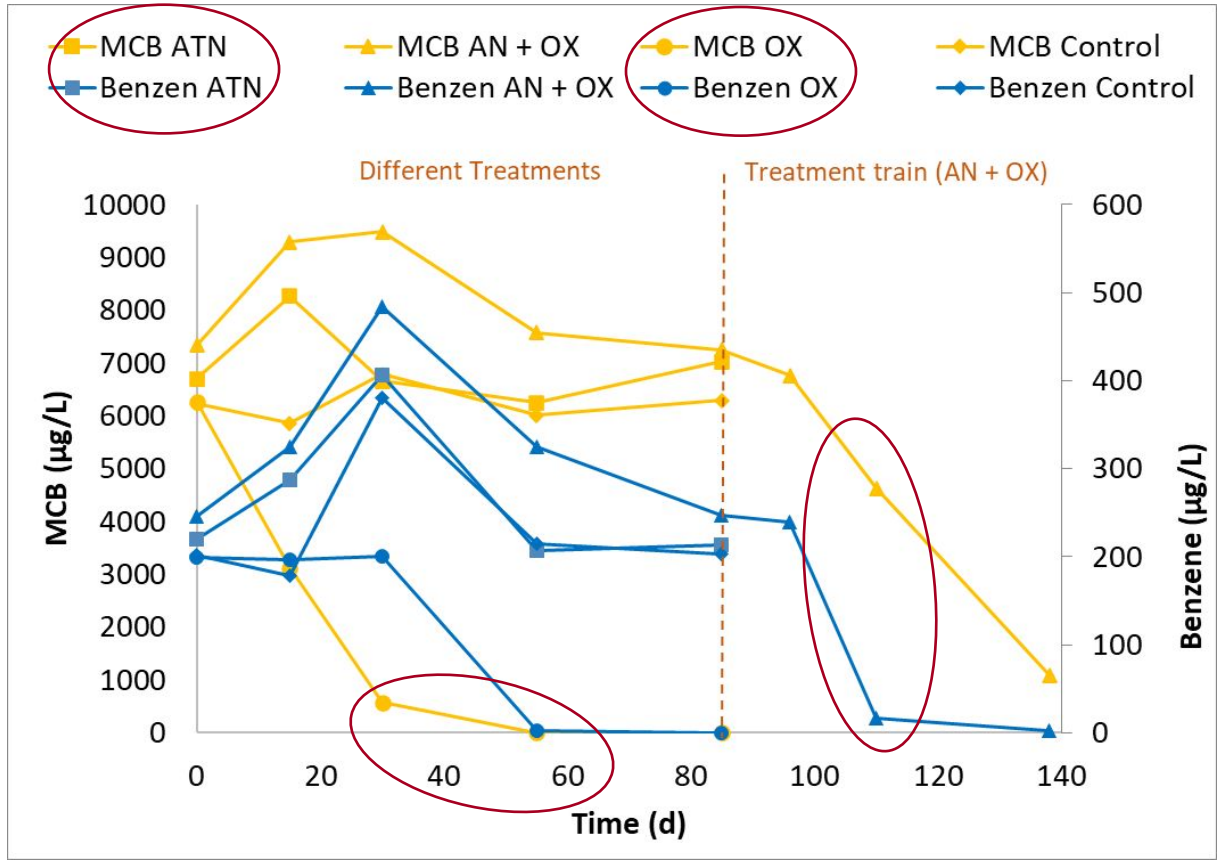
Treatment train (AN+OX) → 85 d + 53 d

16s rDNA Sequencing

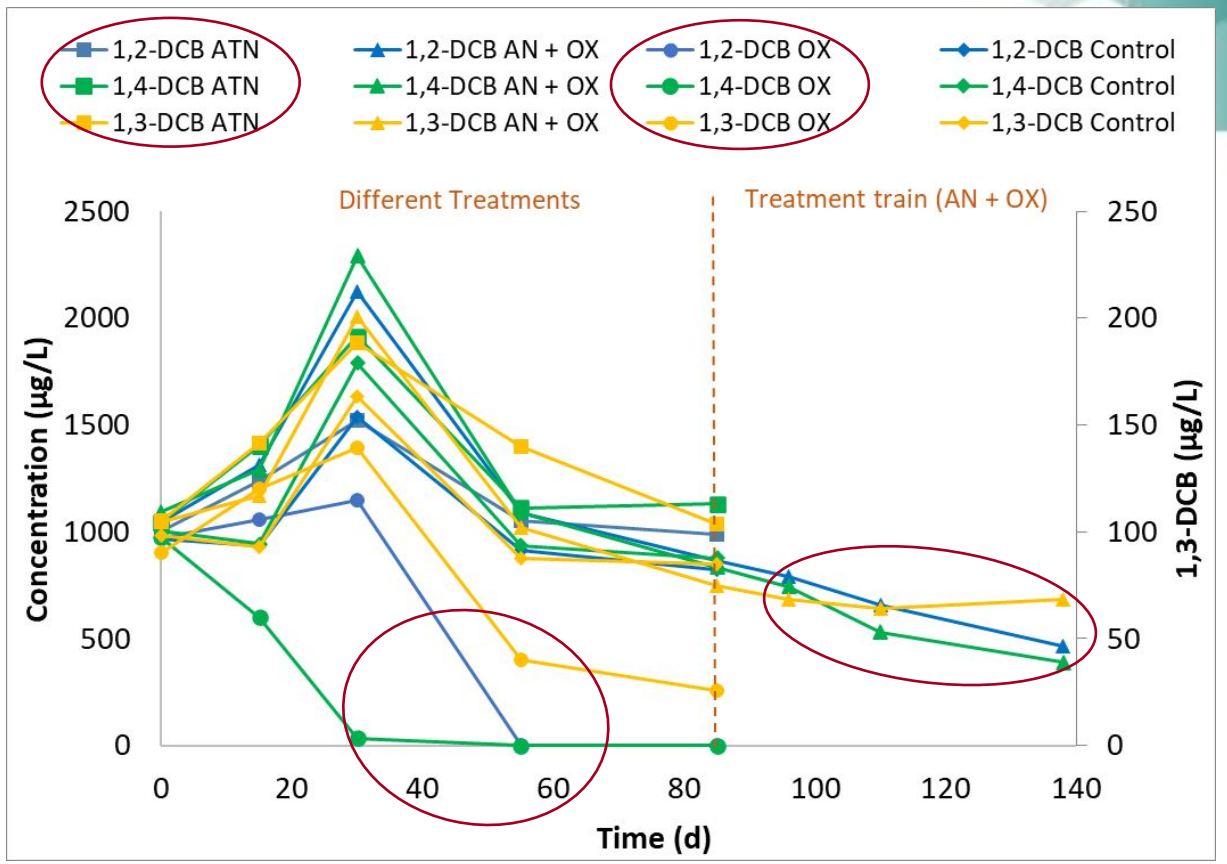
Results: Comparing different treatments

PS16E Well at 13°C

Benzene and MCB



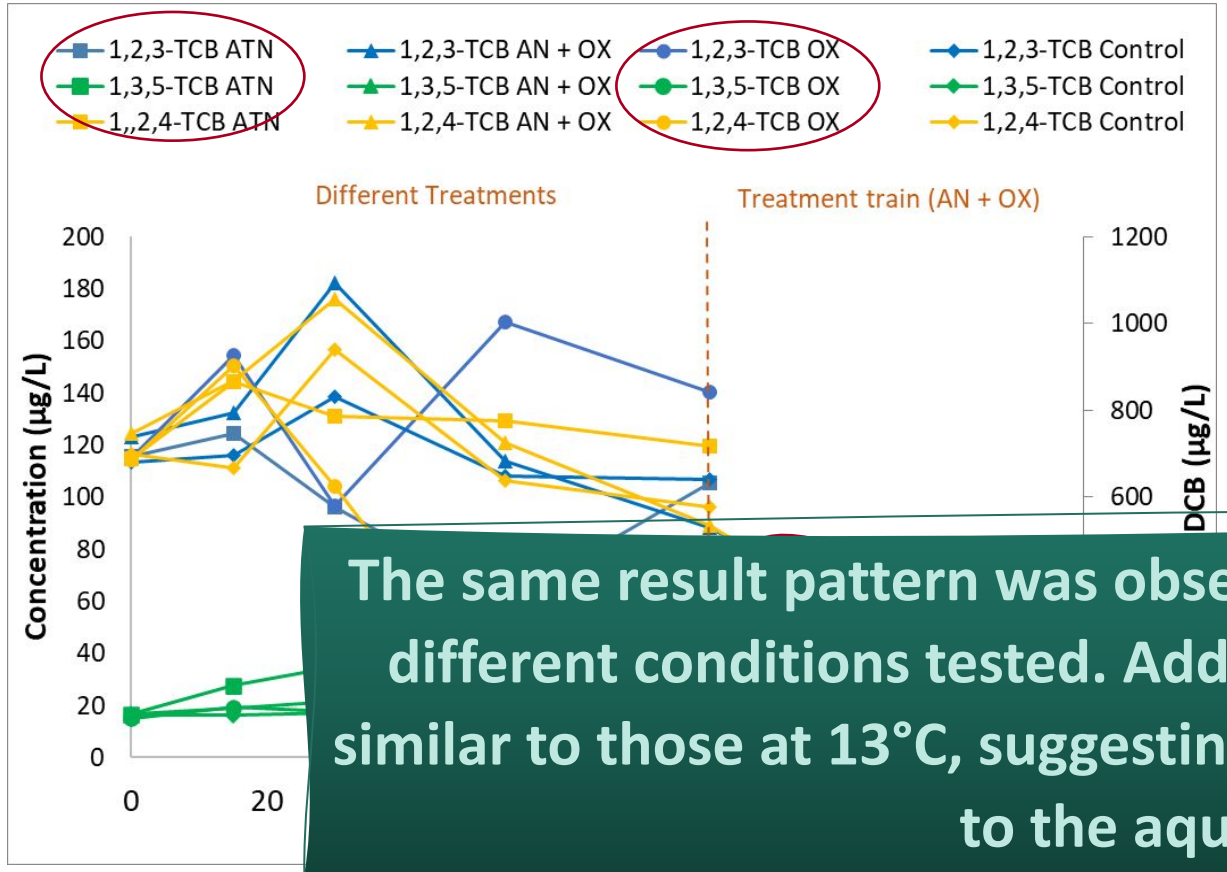
DCB Isomers



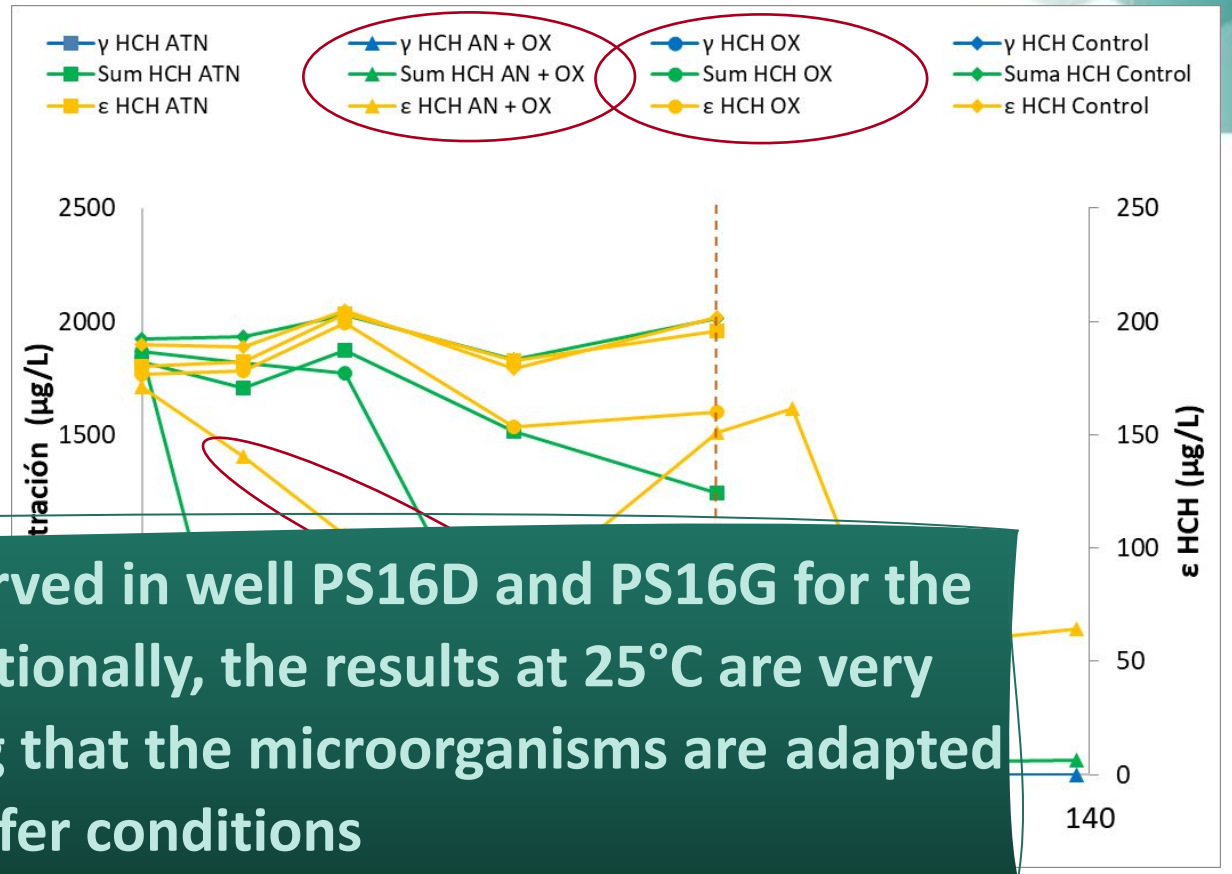
Results: Comparing different treatments

PS16E Well at 13°C

TCB Isomers



HCH Isomers



The same result pattern was observed in well PS16D and PS16G for the different conditions tested. Additionally, the results at 25°C are very similar to those at 13°C, suggesting that the microorganisms are adapted to the aquifer conditions

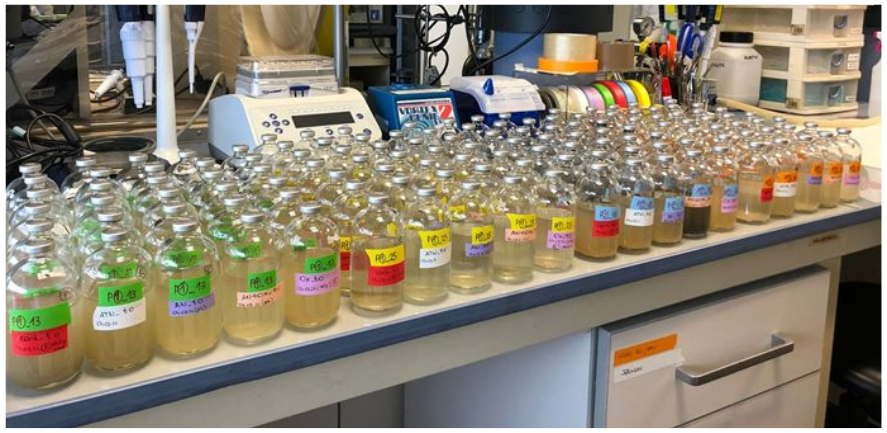
Set up of the second microcosms

To study the effect of nutrients on the degradation rate

2 Wells:

PS16E → 13°C

PS16I → 13°C



Aerobic biostimulation (OX) → **15 d**

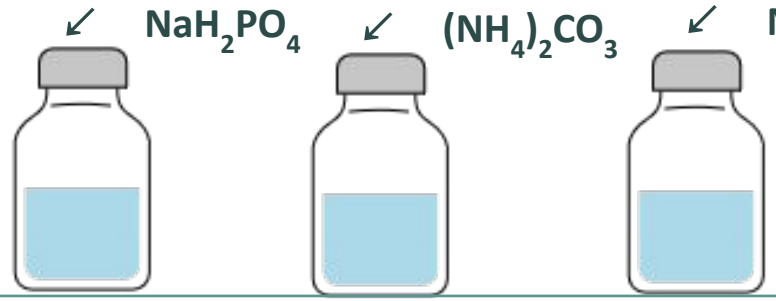
↙ NaH_2PO_4 ↙ $(NH_4)_2CO_3$ ↙ $NaH_2PO_4 + (NH_4)_2CO_3$

Anaerobic biostimulation (AN) → **15 d**

↙ NaH_2PO_4 ↙ $(NH_4)_2CO_3$ ↙ $NaH_2PO_4 + (NH_4)_2CO_3$

Treatment train (AN+OX) → **15 d + 15 d**

↙ NaH_2PO_4 ↙ $(NH_4)_2CO_3$ ↙ $NaH_2PO_4 + (NH_4)_2CO_3$



16s rDNA Sequencing

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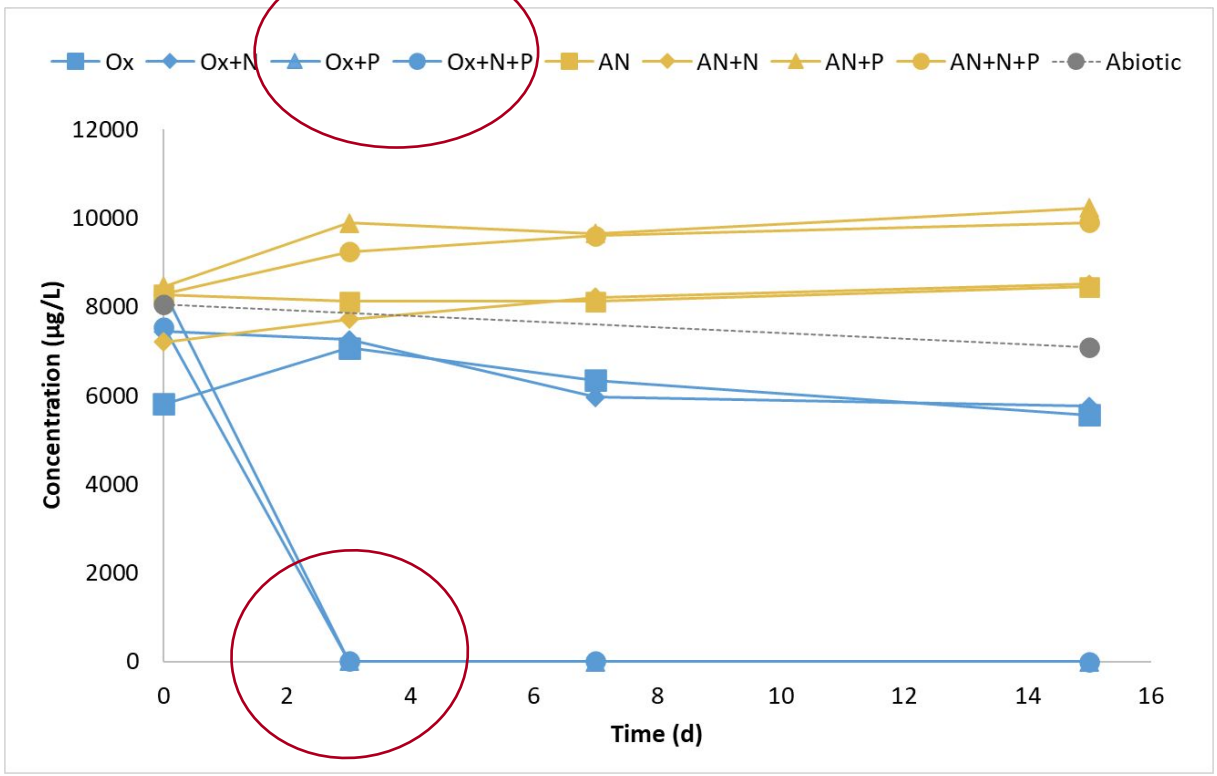
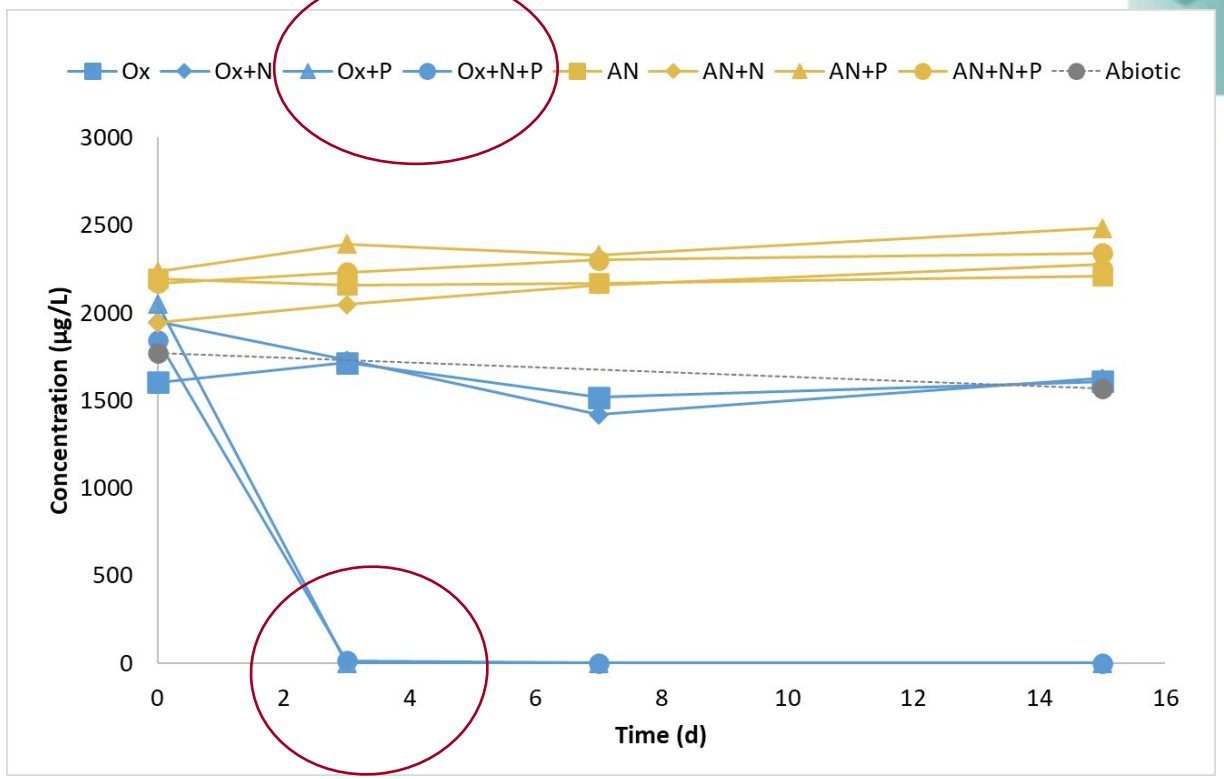
Results: Comparing AEROBIC and ANAEROBIC biostimulation

PS16E Well at 13°C

MCB



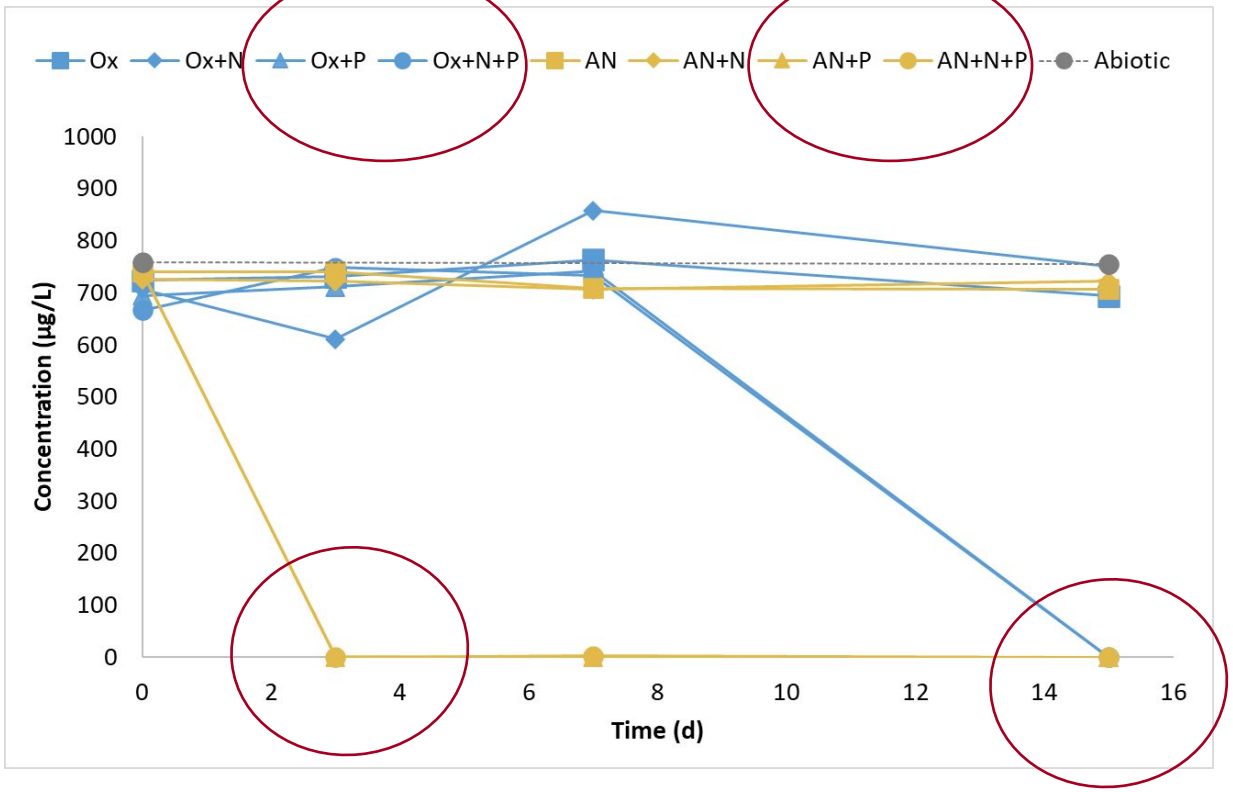
1,4-DCB



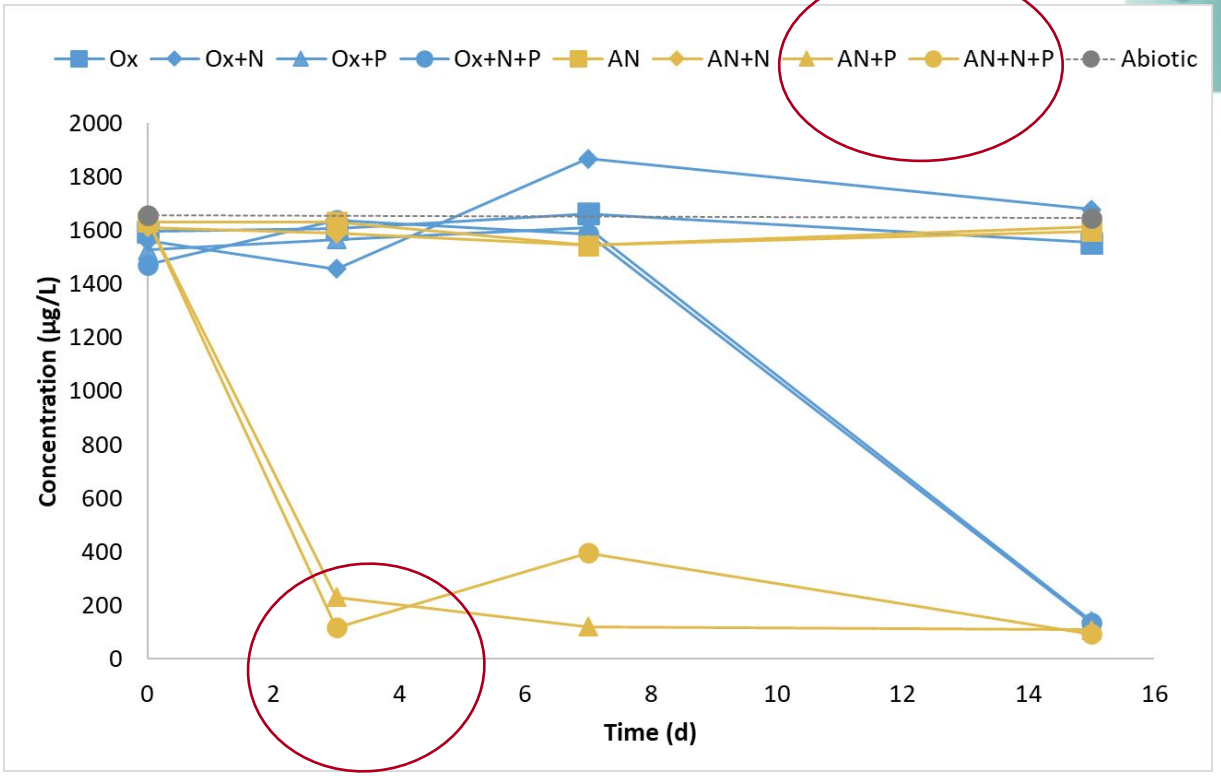
Results: Comparing AEROBIC and ANAEROBIC biostimulation

PS16E Well at 13°C

γ-HCH



Sum-HCH



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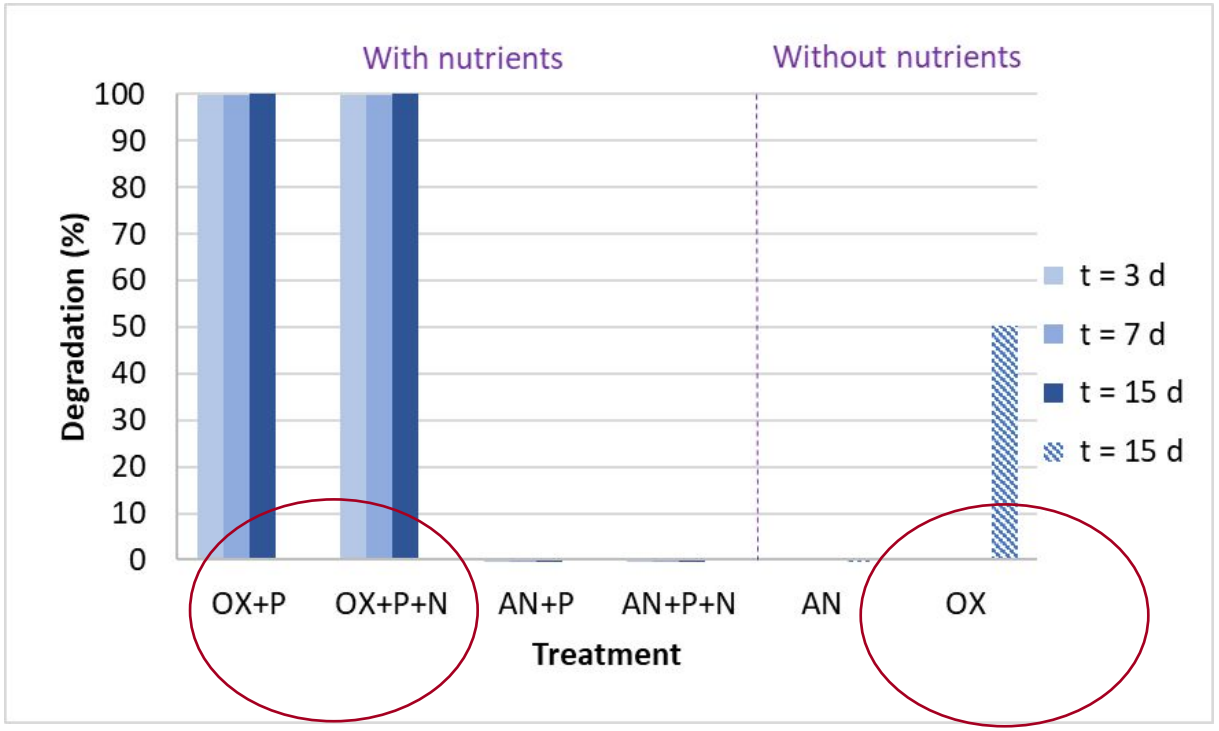
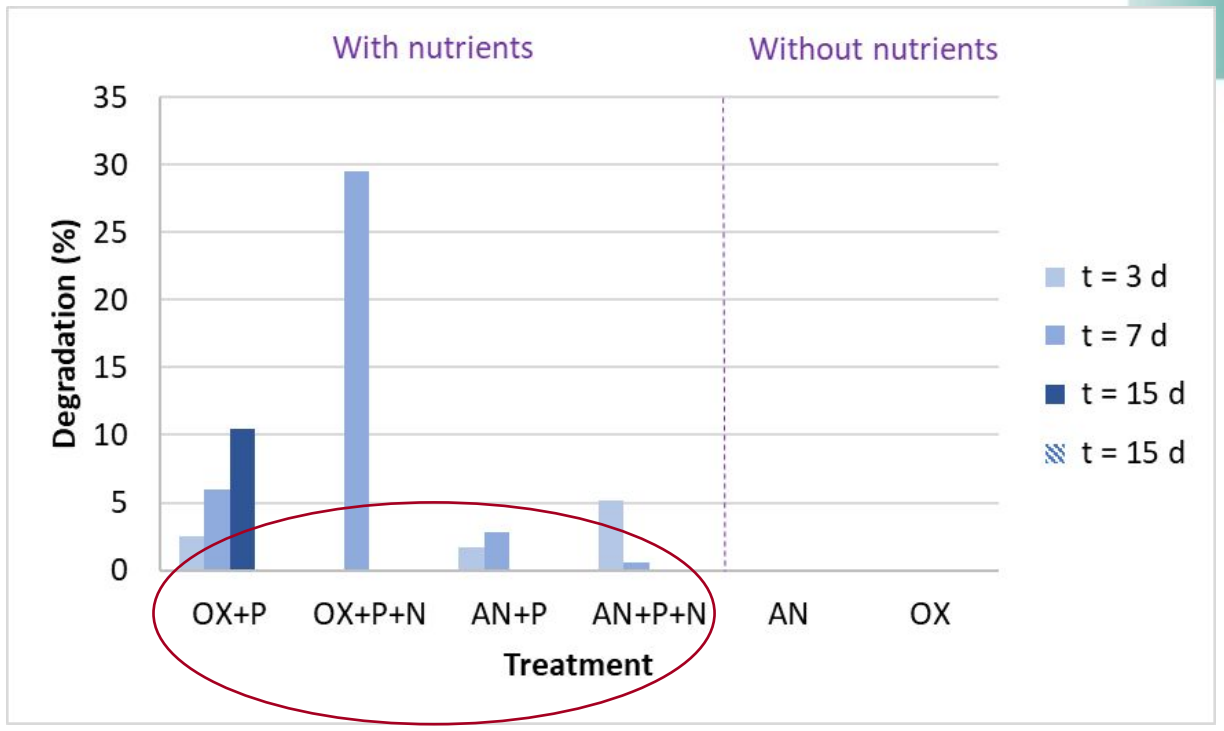
Results: Microcosms combined

Comparison between PS16E well WITH and WITHOUT nutrients at 13°C

MCB



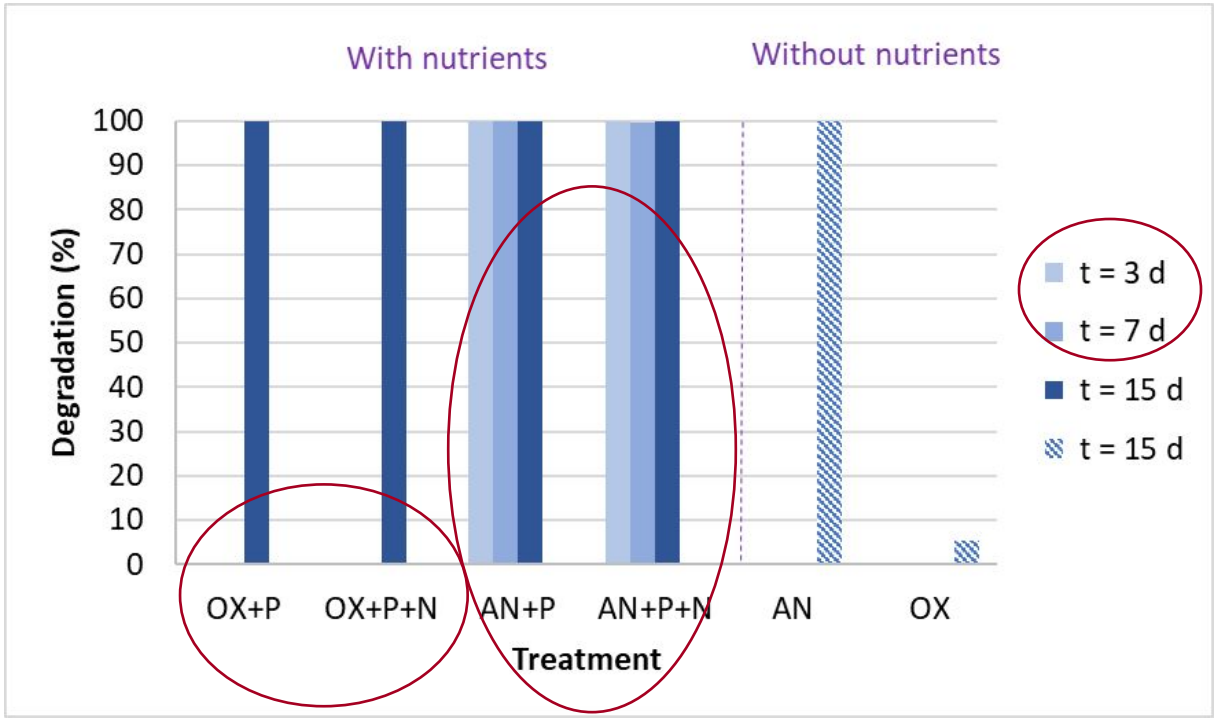
1,3,5-TCB



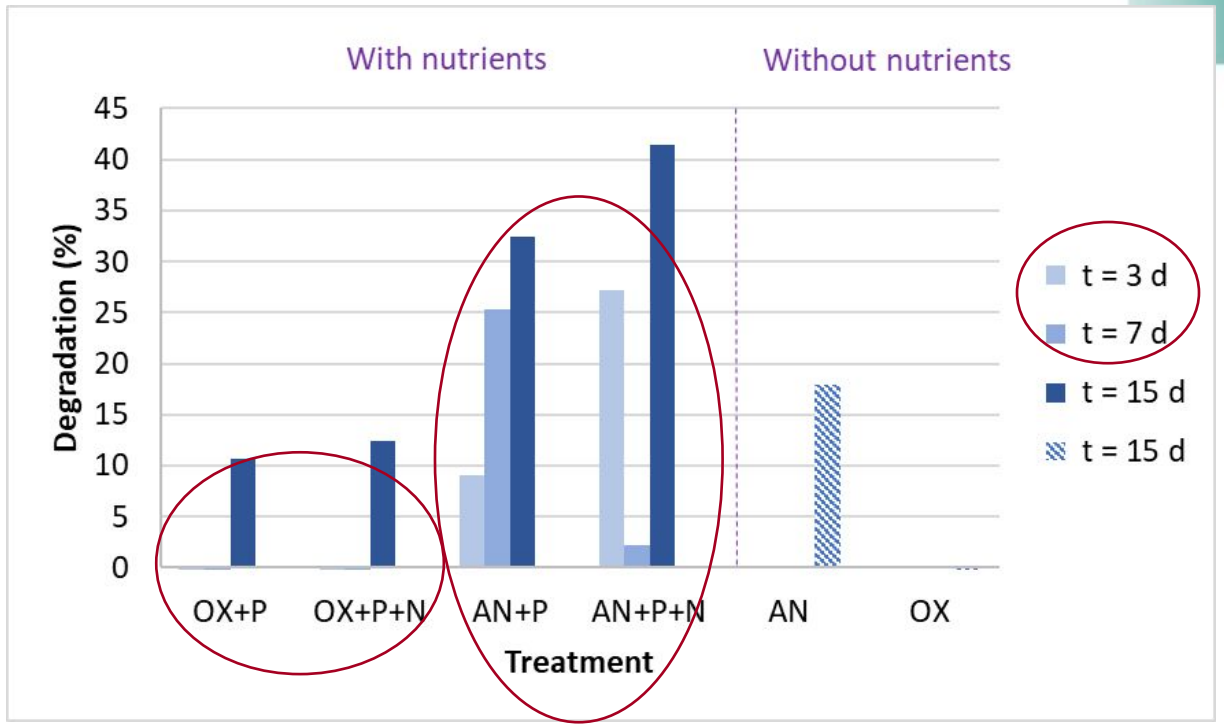
Results: Microcosms combined

Comparison between PS16E well WITH and WITHOUT nutrients at 13°C

γ-HCH

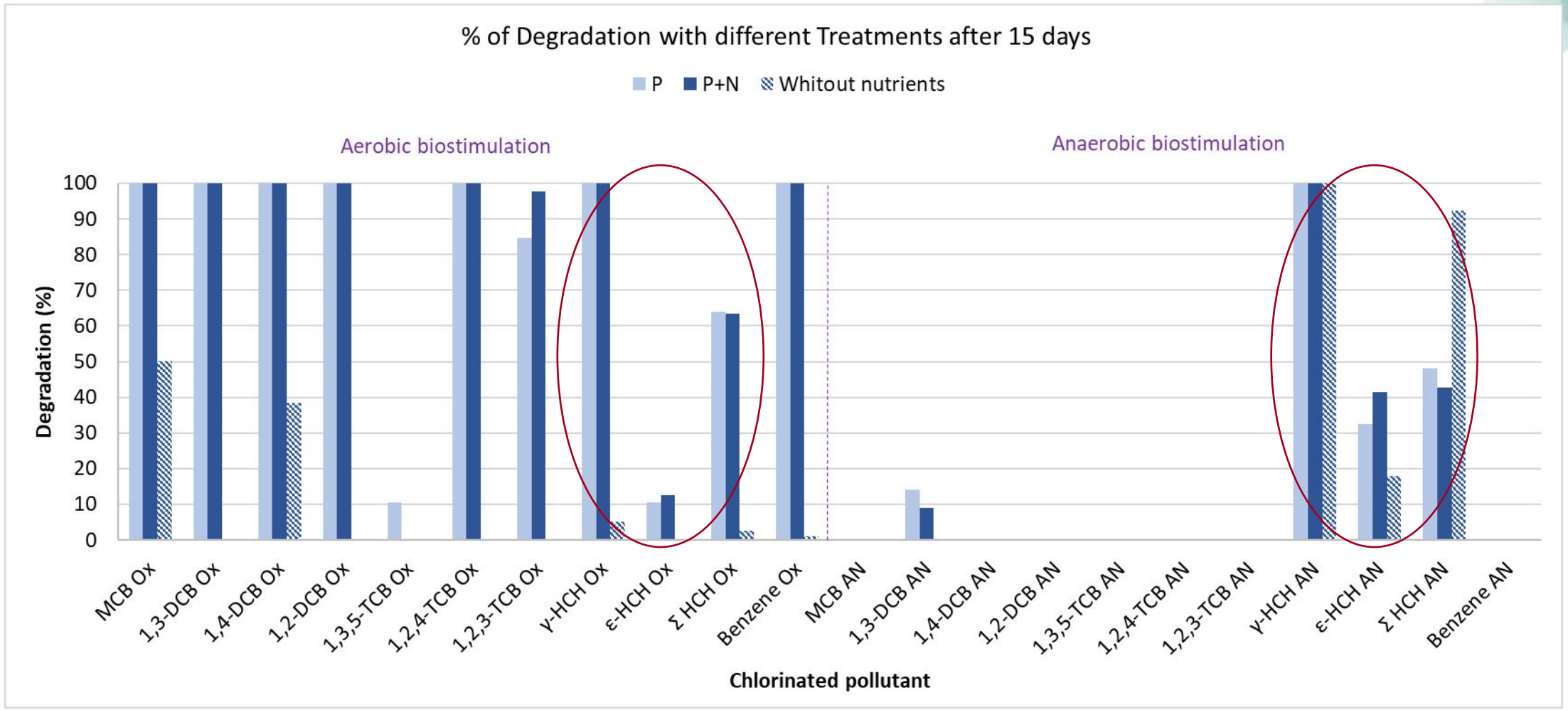


ε-HCH



Results: Microcosms combined overview

Comparison between PS16E well WITH and WITHOUT nutrients at 13°C



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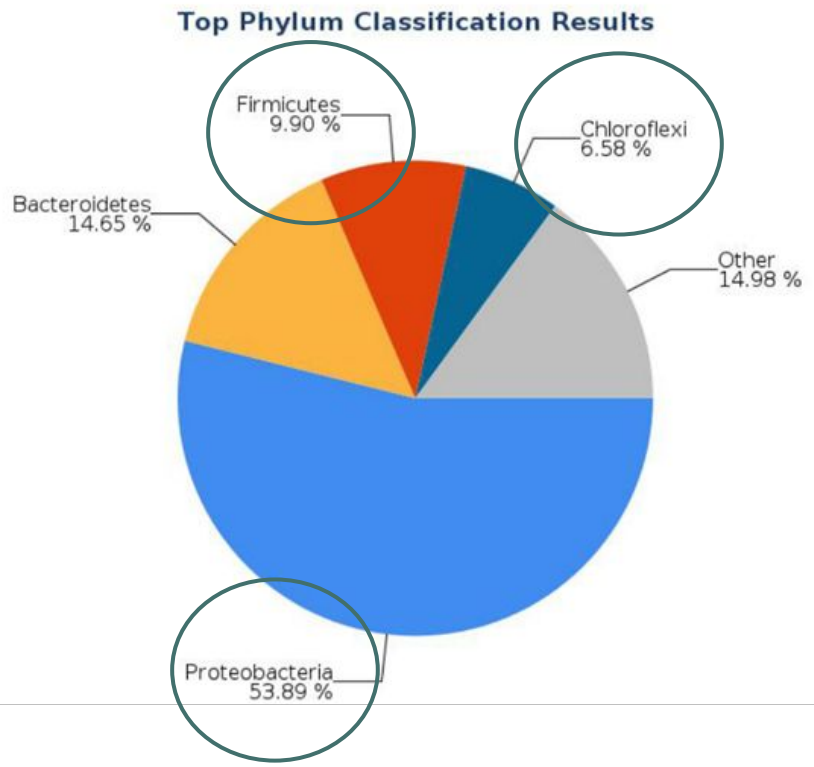
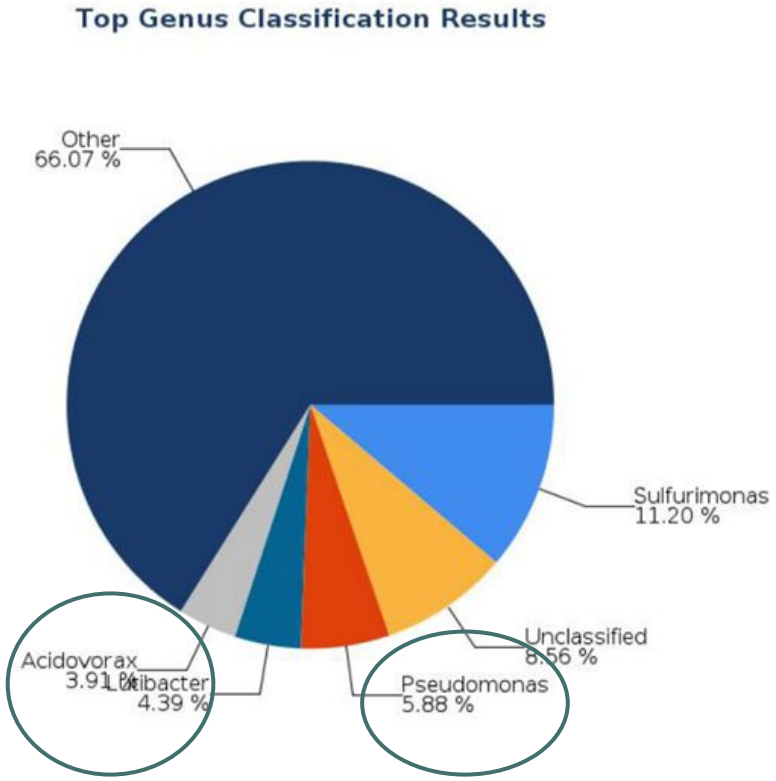
Results: Study of microorganisms on the microcosm experiments

PS16E at 13°C

Initial stage





In the initial sample, phyla of bacteria that can perform the degradation of chlorobenzenes under both aerobic and anaerobic conditions are already detected.

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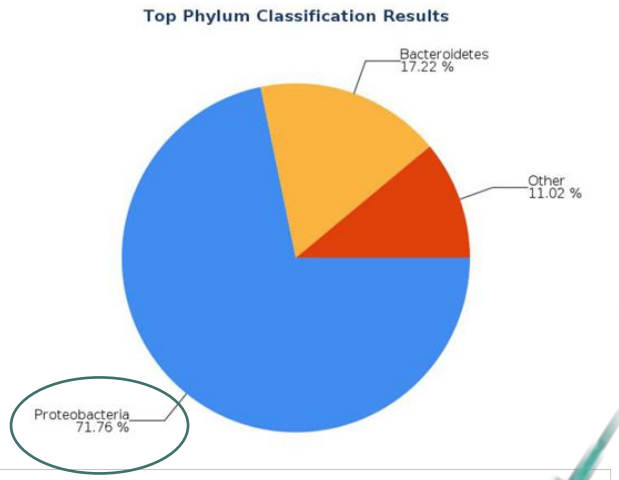
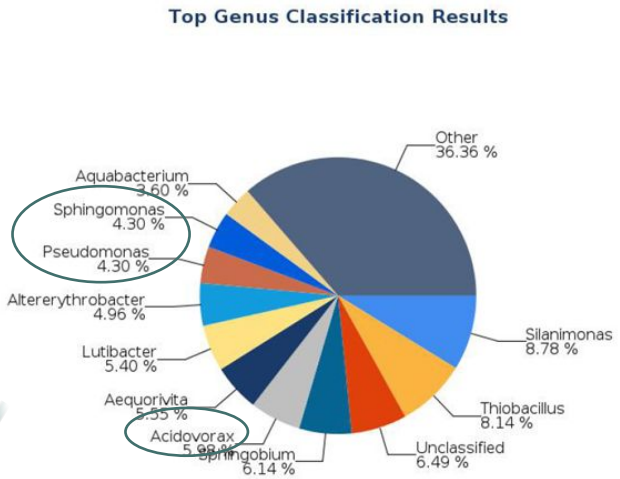
Results: Study of microorganisms on microcosm experiments

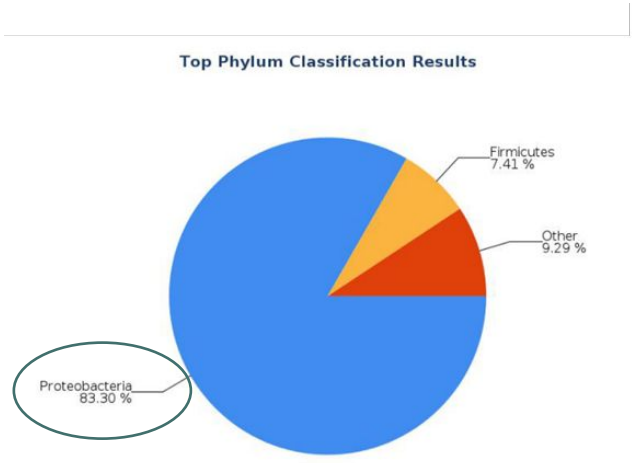
Aerobic biostimulation of PS16E at 13°C

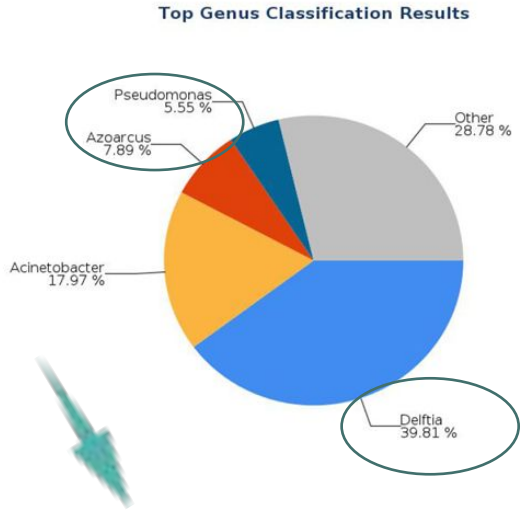
After 85 d without nutrients





After 15 d with P + N



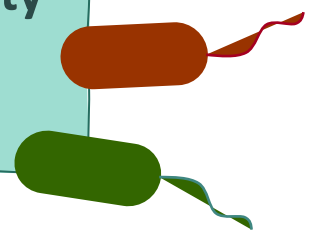


4,30% *Pseudomonas*

5,98% *Acidovorax*

4,30% *Sphingomonas*

Great bacterial diversity including potential aerobic degraders



5,55% *Pseudomonas*

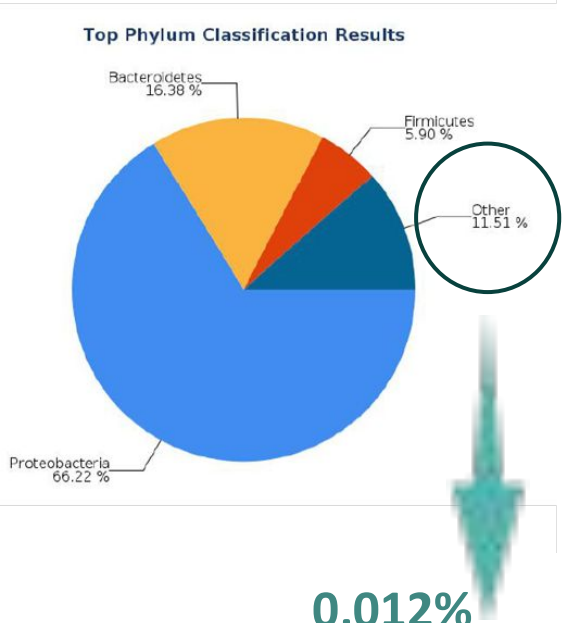
3,26% *Acidovorax*

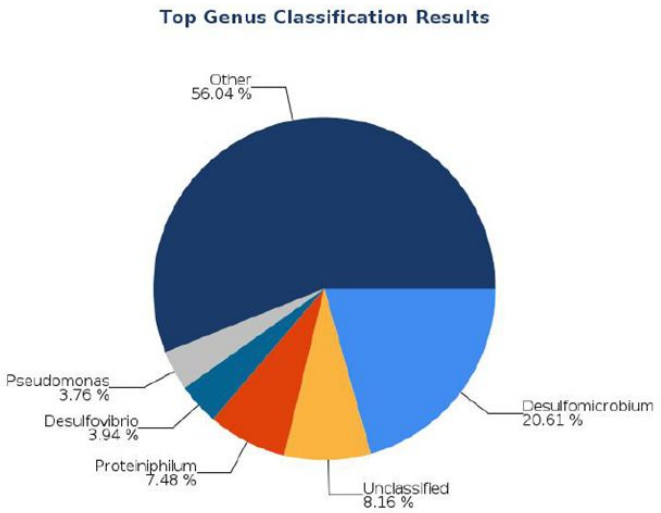
0,20% *Sphingomonas*

Results: Study of microorganisms on microcosm experiments

Anaerobic biostimulation of PS16E at 13°C

After 85 d without nutrients





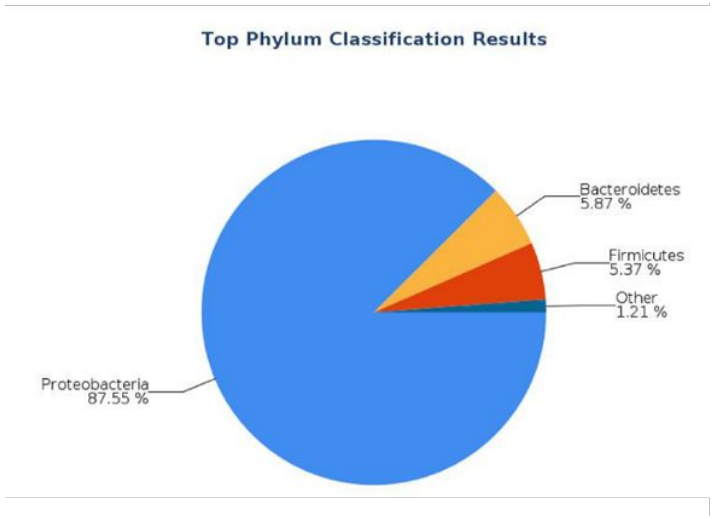
0,012%

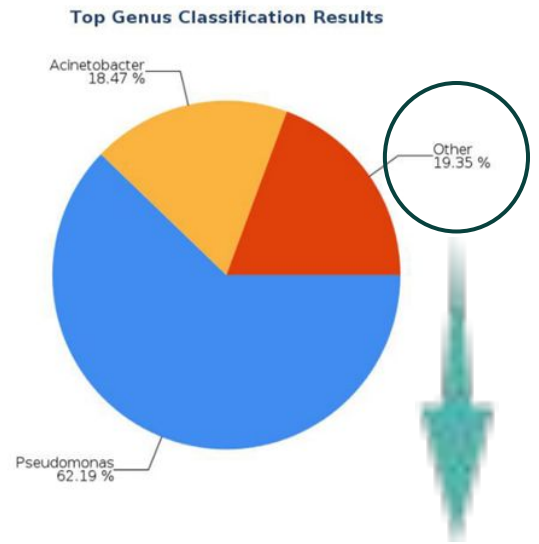
Dehalococcoides

0,095%

Dehalogenimonas

After 15 d with P + N





0,032%

Dehalogenimonas

0,004%

Dehalobacter

Slow growing microorganisms

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Conclusions

Bioremediation proved to be a suitable strategy to treat the alluvial groundwater of Sardas.

Natural attenuation conditions did not show significant biodegradation and therefore biostimulation is needed

Aerobic biostimulation degrades less chlorinated organochlorine contaminants more rapidly

HCH family are degraded faster in anaerobic conditions, and with a longer period and/or nutrients addition, also aerobically

Phosphorus biostimulation increases the degradation rate and achieves the total elimination of some contaminants in 3 days of cultivation

The addition of only nitrogen as biostimulant does not improve the degradation rate

The presence of chlorinated degrader microorganisms in aerobic and anaerobic conditions was detected

The strategy proposed for *in situ* pilot testing is aerobic biostimulation with oxygen and phosphorous addition

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THANK YOU FOR YOUR ATTENTION

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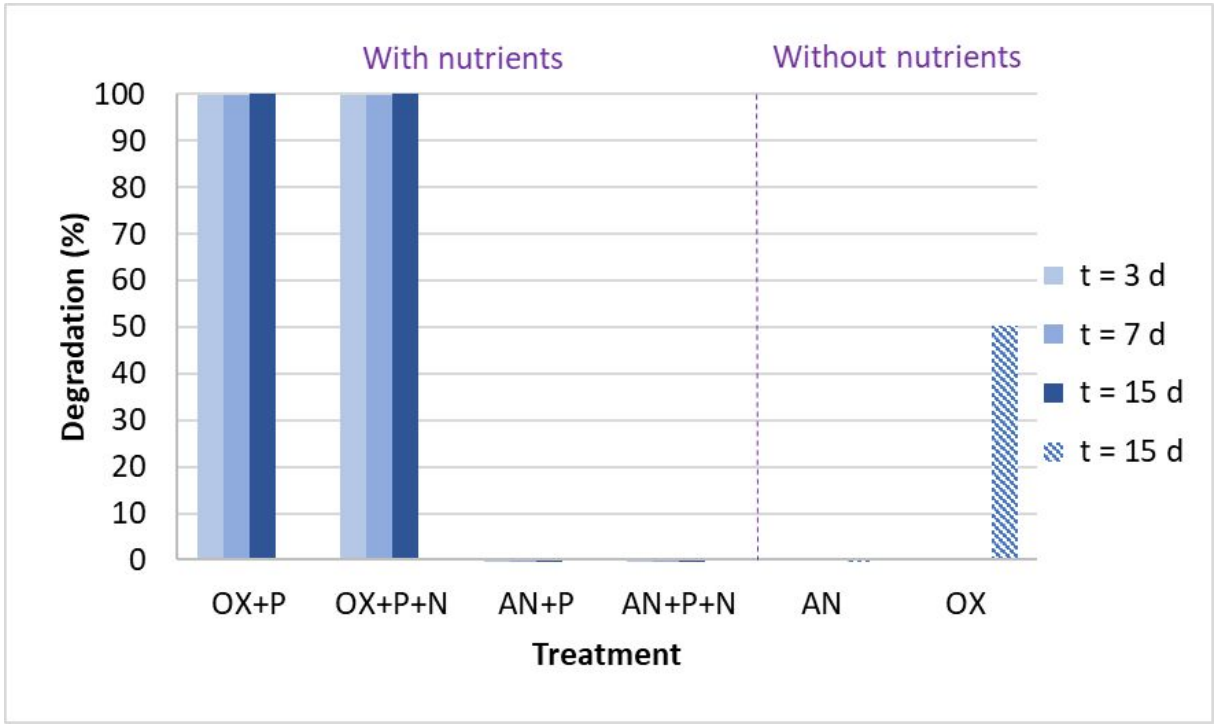
Results: Microcosms combined

Comparison between PS16E well WITH and WITHOUT nutrients at 13°C

MCB



γ-HCH