



# Soil as genetic resources: an underestimated potential in the sustainable management of contaminated sites by persistent organic pollutants

**Intersol 2022**

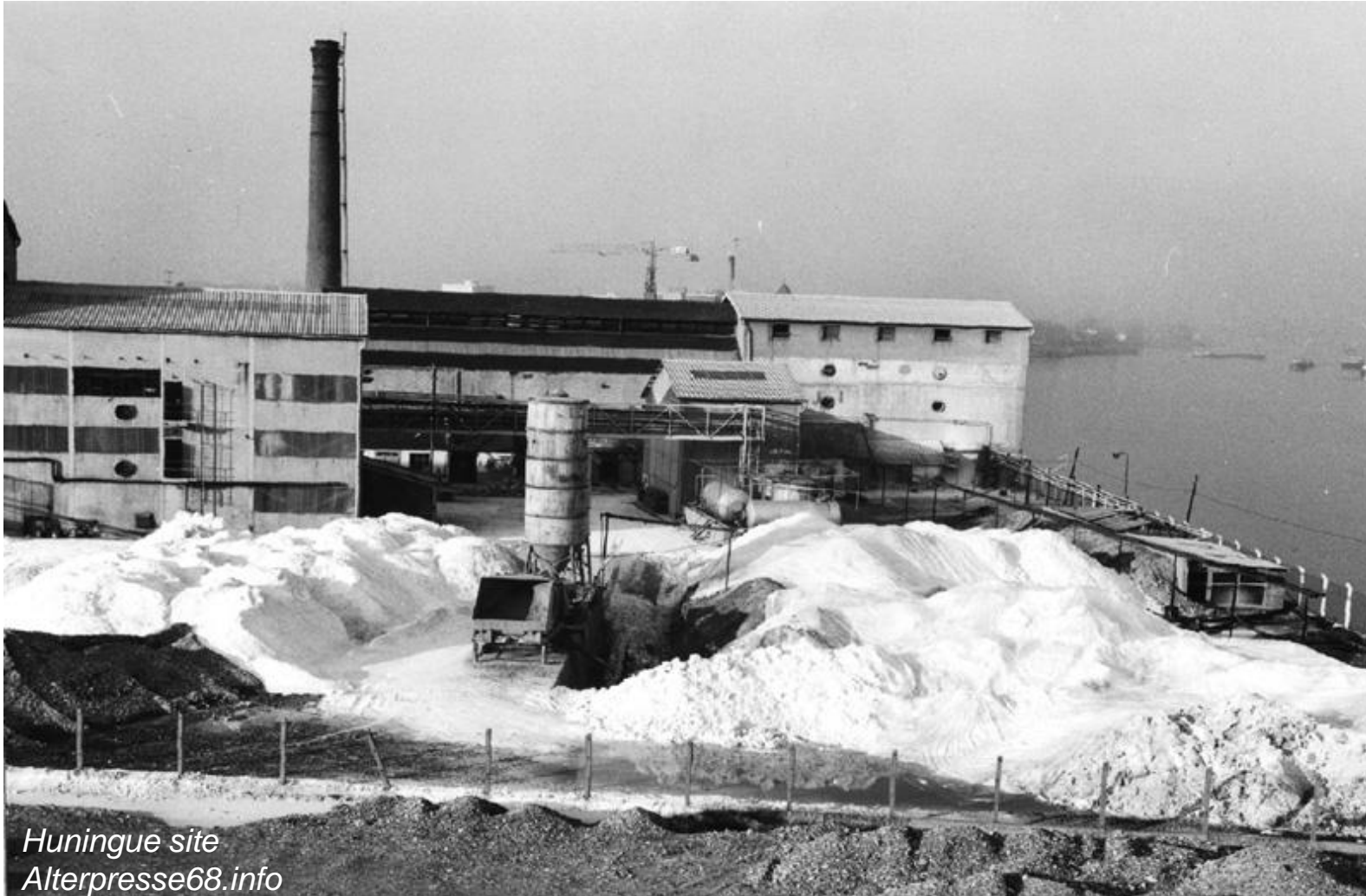
*Alexiane Godain, Céline Baguelin, Pauline Denaud, Sébastien Kaskassian,  
Cédric Malandain, Thierry Ruffenach*



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# HCH waste management

## Lindane production site



**600,000tons**  
of Lindane have been  
produced across the  
world leading to about

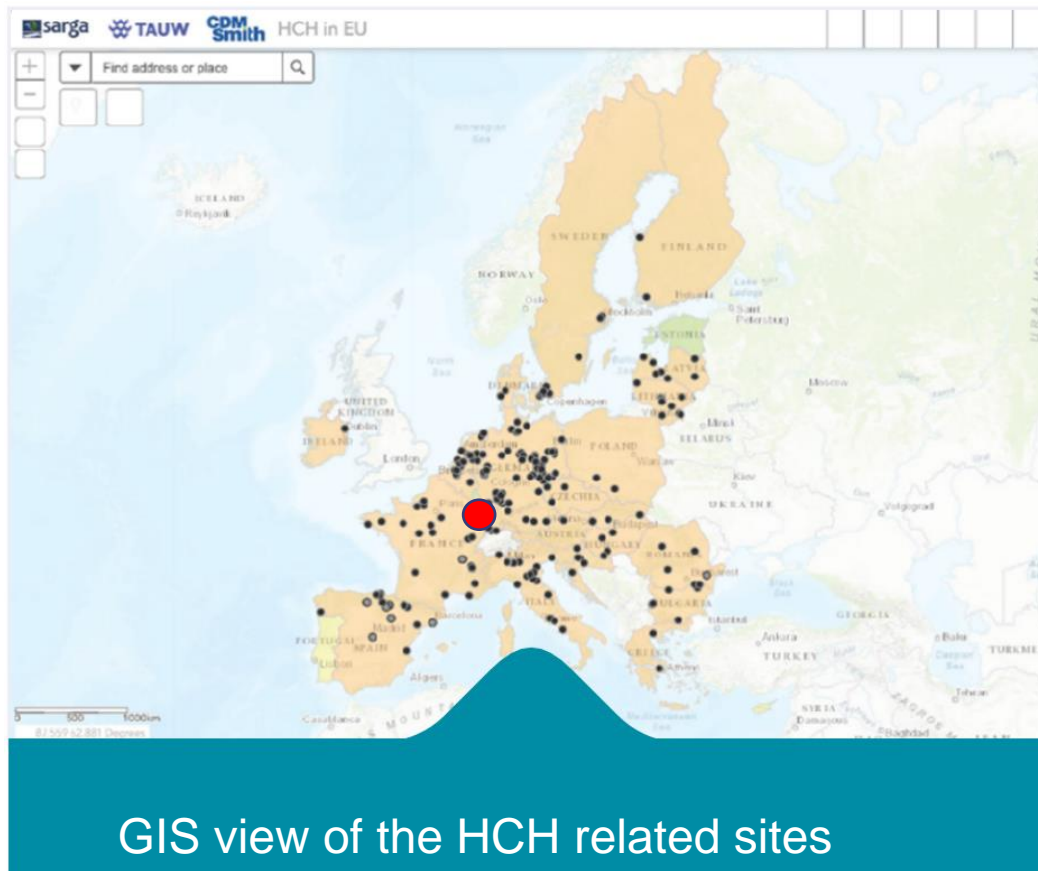
**4.8 million tons**  
of HCH waste generated

*Huningue site  
Alterpresse68.info*



# European project: HCH in EU

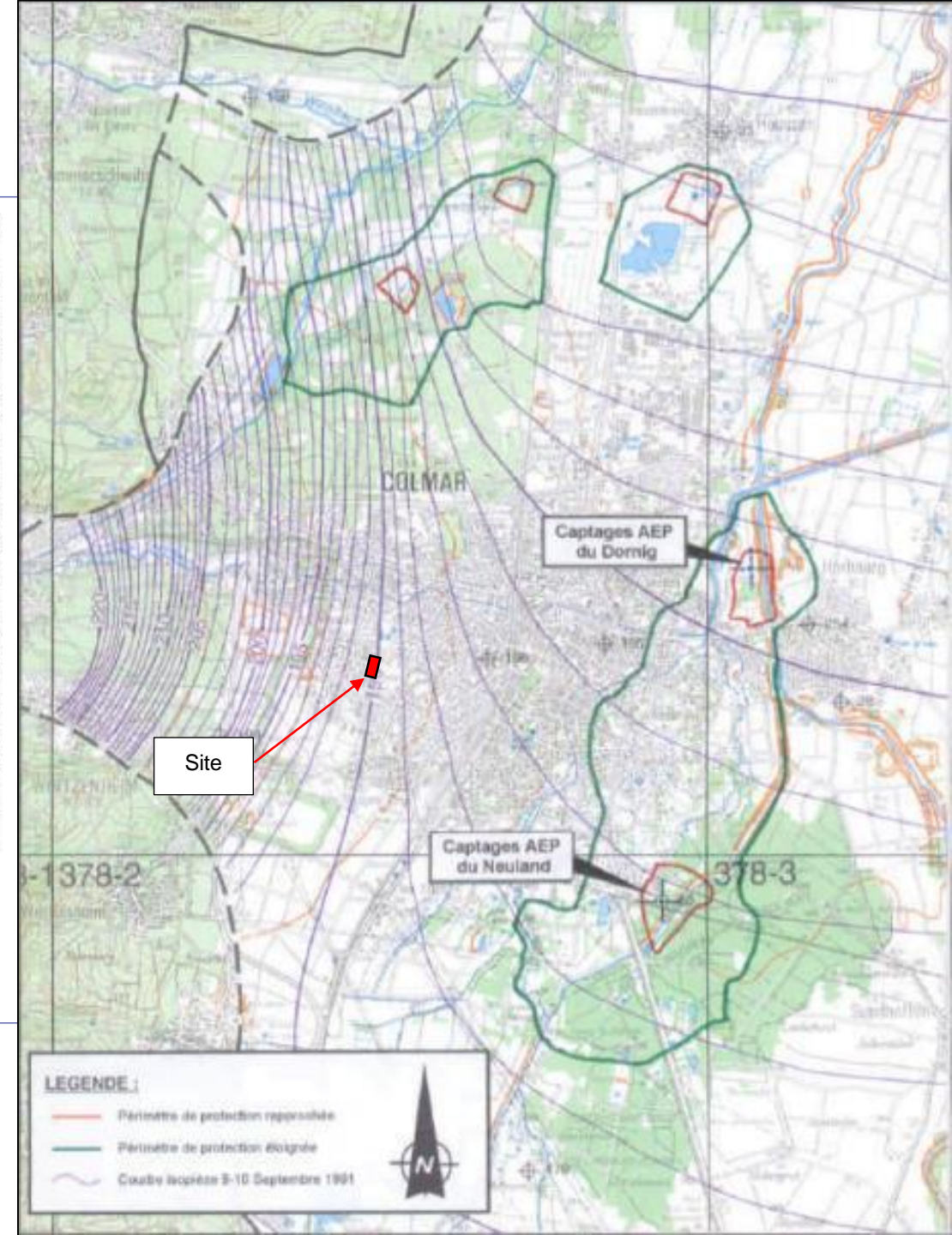
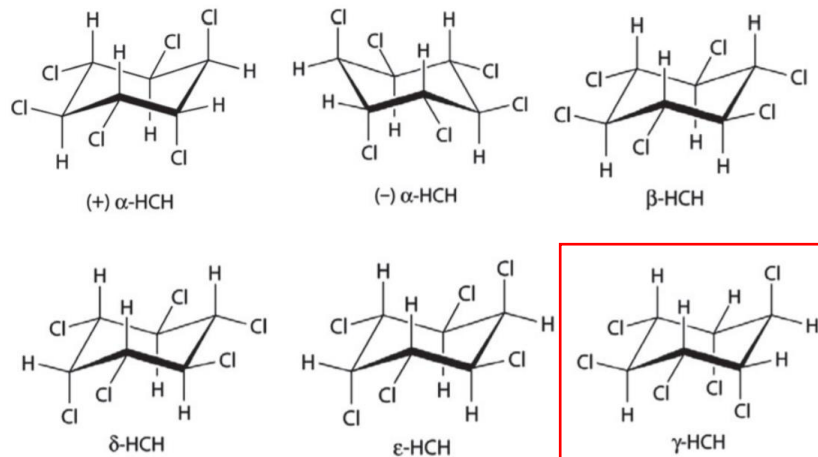
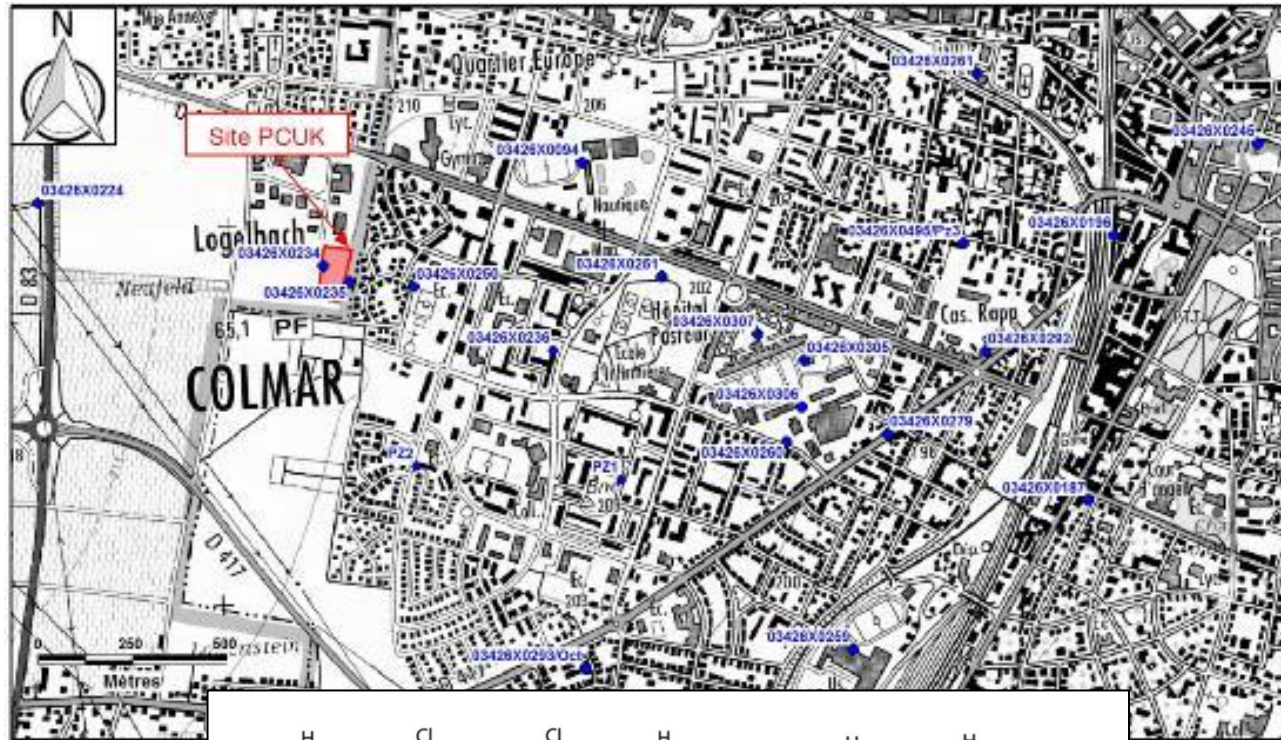
## Purpose of the project



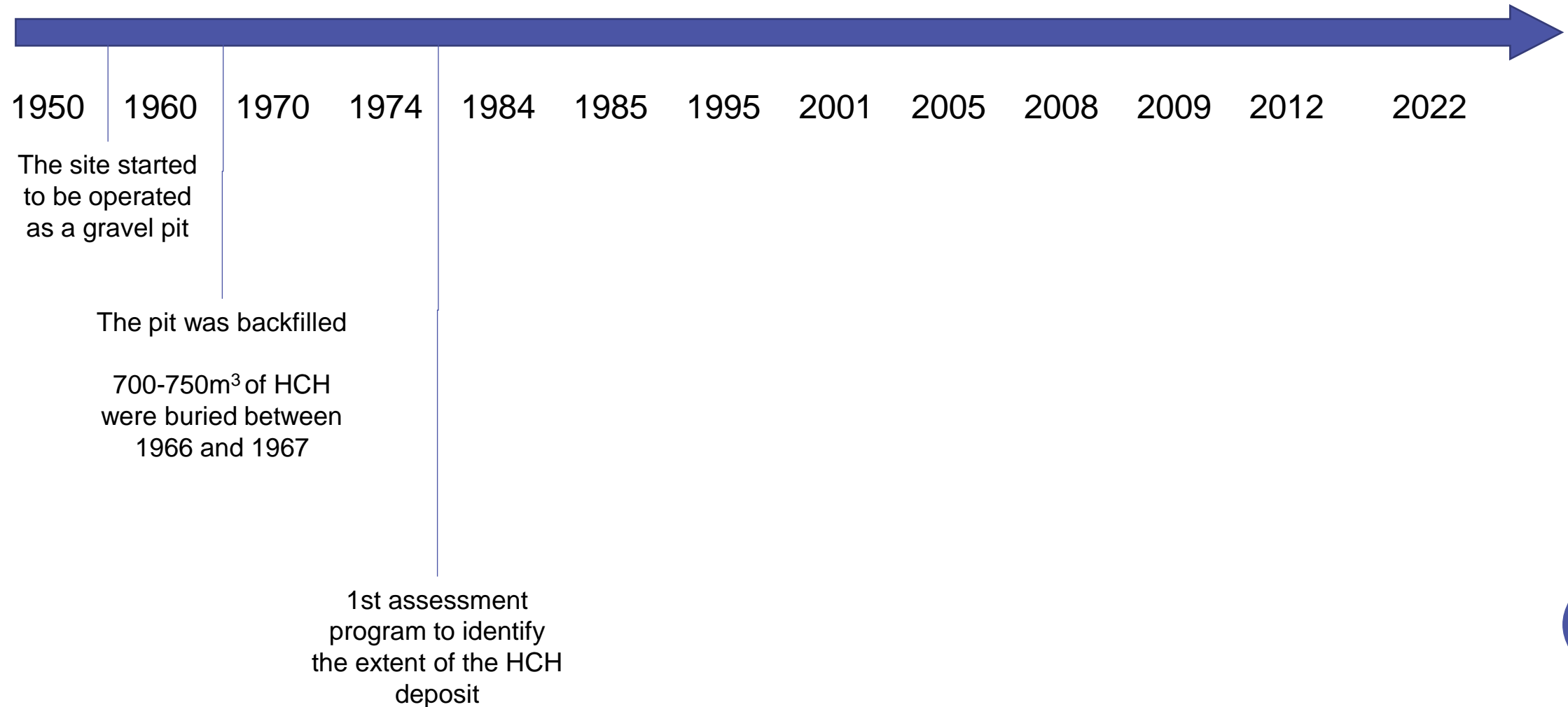
- The European Parliament has asked the European Commission to initiate the project 'HCH in the EU'.
- This project has two objectives:
  - Perform an inventory of the regulation of Lindane production, and technical HCH in Europe.
  - Assist the authorities (technical and organizational support) with the sustainable management of six HCH contaminated sites.



# PCUK site in Wintzenheim (FR)

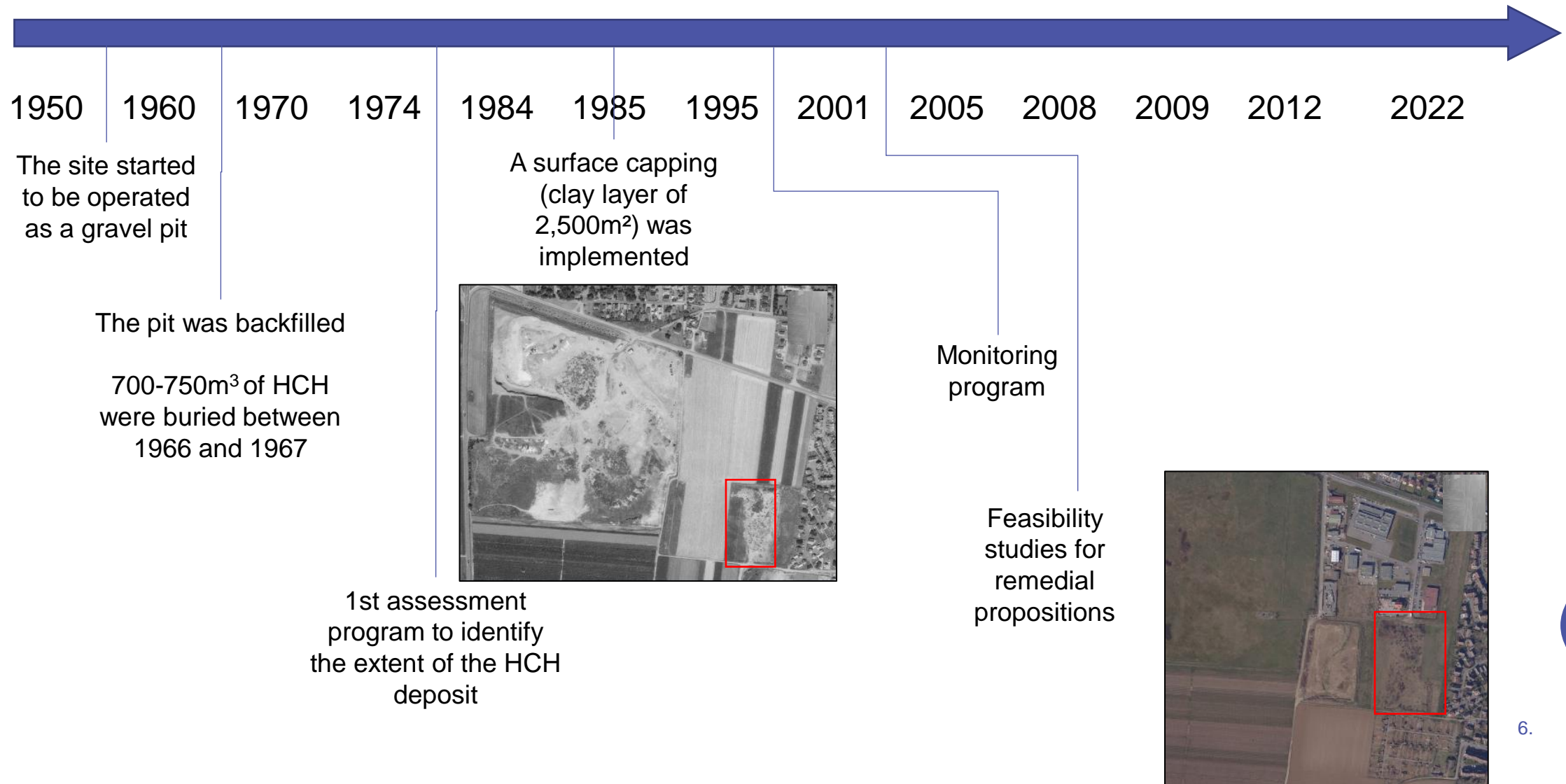


# Site history

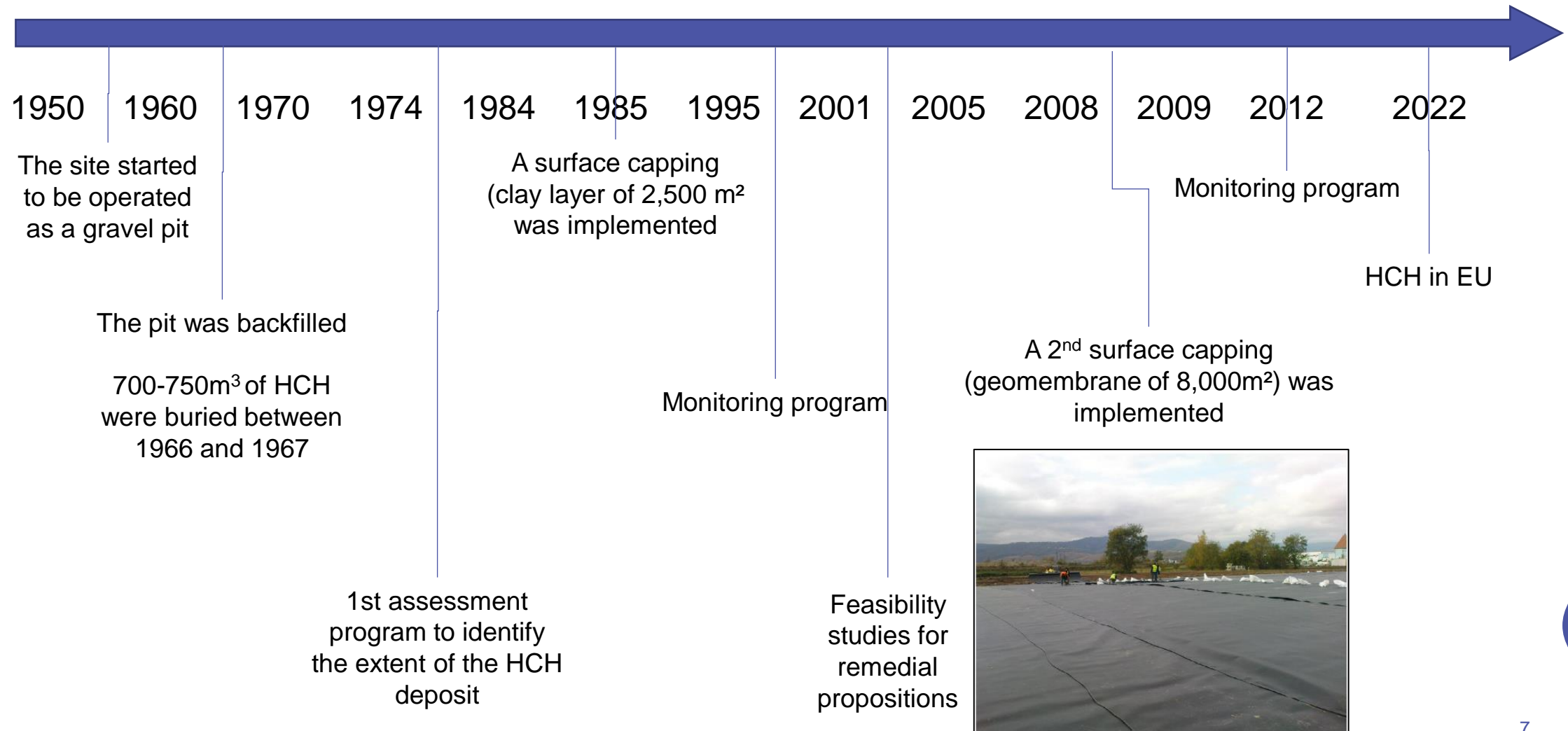




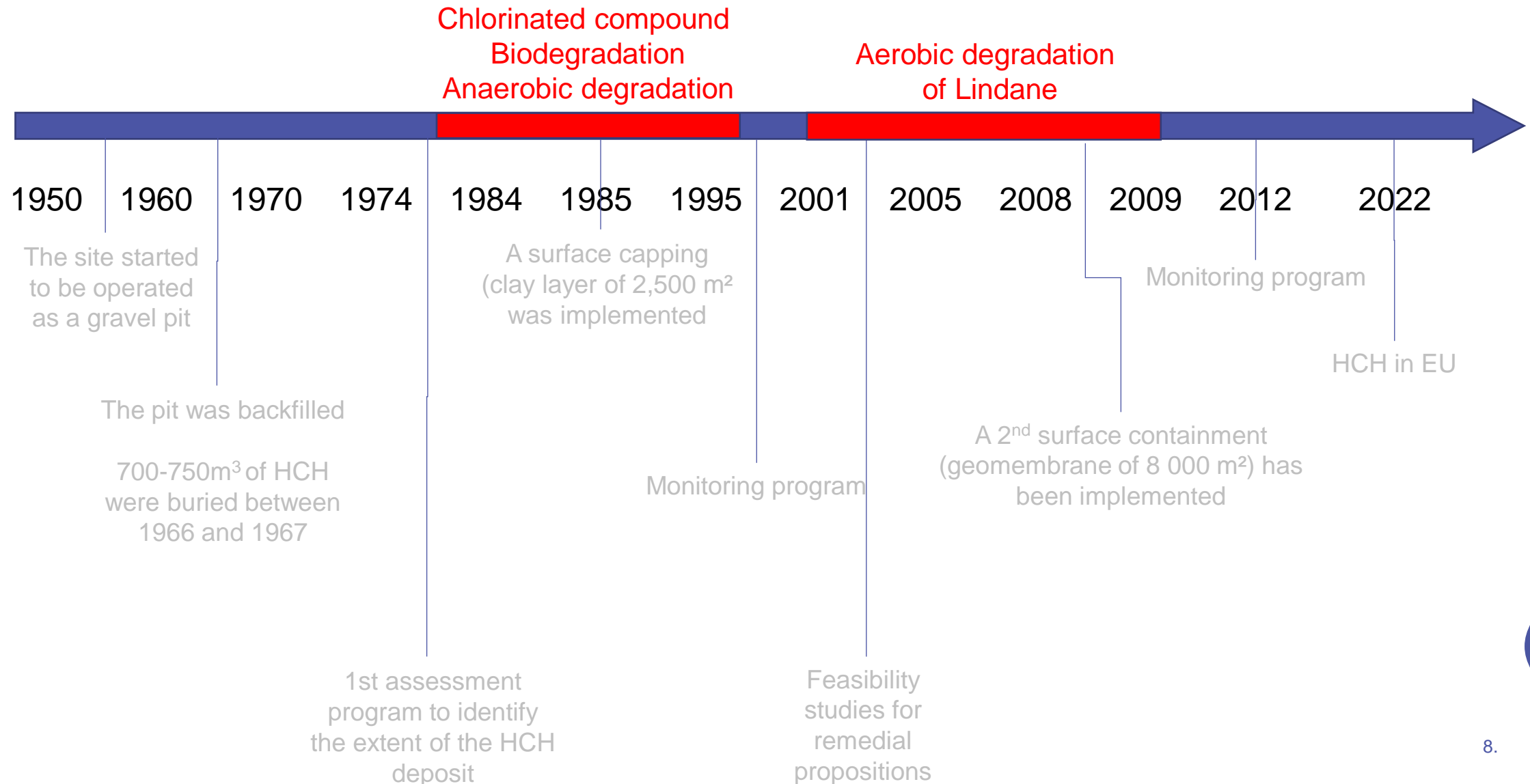
# Site history



# Site history



# Site history vs. scientific knowledge



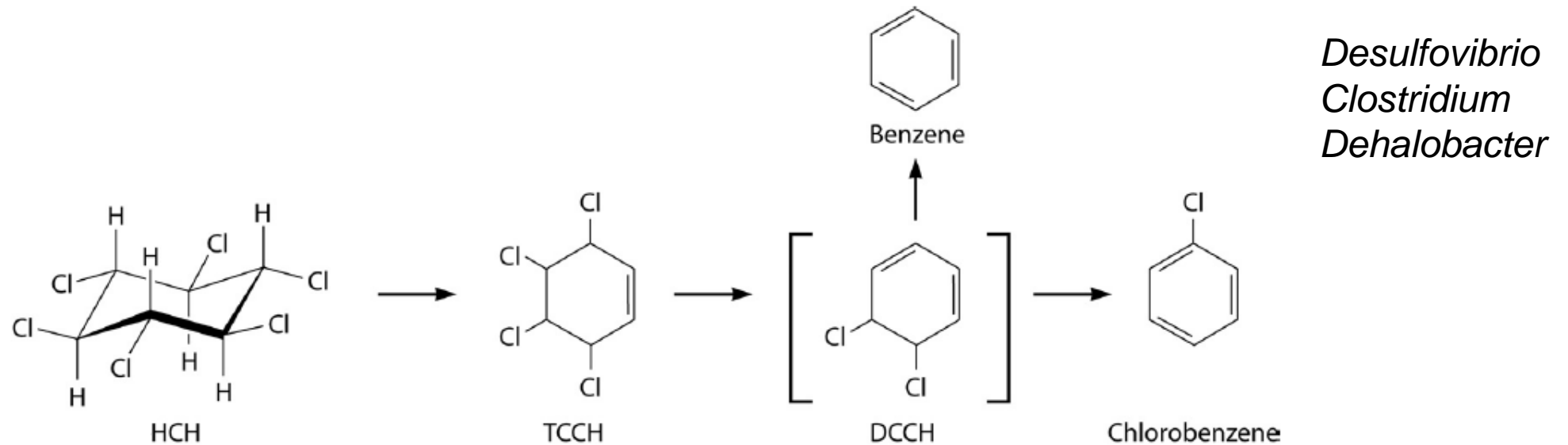


# PCUK site: Lindane plume extent





# Anaerobic biodegradation: benzene and chlorobenzene production



- Such as for chloroethene, HCH was first thought to be degraded by dehalorespiration
- **Lack of knowledge** about microbial species, and genes involved in the degradation process
- **The pathway is also poorly understood** but leads to the production of benzene and chlorobenzene

➡ **Difficulties are similar to those with chloroethene bioremediation**

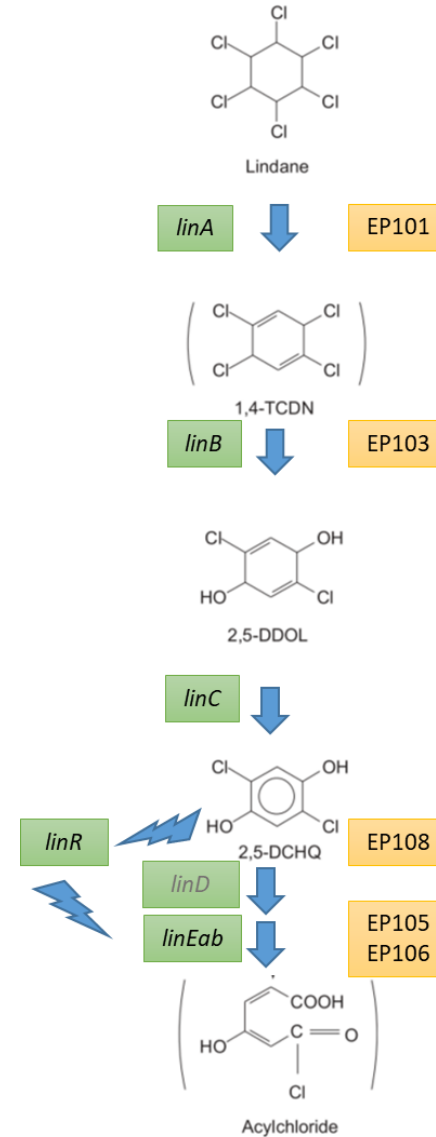


# Aerobic degradation: complete mineralization

- HCH isomers are degraded as a carbon source
  - Bacterial species involved in the degradation are **well known and easy to handle**
- genes are also well known



- The degradation pathway has been extensively studied



*Sphingobium*  
*Rhodococcus*  
*Microbacterium*  
...



# Lindane in the EU (2016)

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*“The advantage of biological methods is that they are:*

- Environmentally-sound;*
- Low-cost methods; and*
- They can be used in situ.*

*[...] Therefore, **further research, especially field studies, is needed** to better understand the factors affecting the bioremediation and to improve their efficiency.”*

*“Although efficient **microbial degradation of lindane and other HCH-isomers has been demonstrated in several laboratory studies [...].***

*There are only a few field studies on the microbial remediation of lindane contaminated soils and waters, and therefore more research is needed.”*





# Field demonstration under anaerobic conditions (2013)

## Stimulation of Hexachlorocyclohexane (HCH) Biodegradation in a Full Scale In Situ Bioscreen

Alette A. M. Langenhoff,<sup>\*,†,‡</sup> Sijf J. M. Staps,<sup>†,||</sup> Charles Pijls,<sup>§</sup> and Huub H. M. Rijnaarts<sup>†,‡</sup>

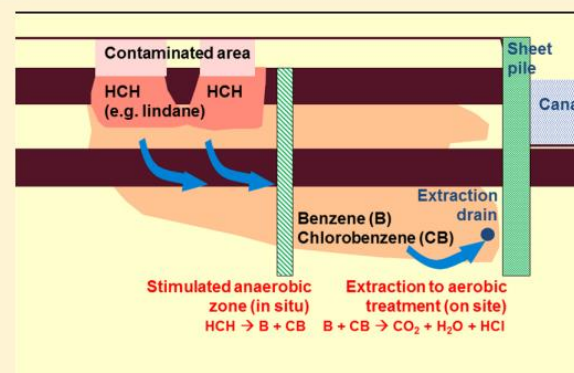
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**ABSTRACT:** The feasibility of a bioscreen for the in situ biodegradation of HCH and its intermediates is demonstrated at a contaminated site in The Netherlands, via the discontinuous addition of methanol as electron donor. An infiltration system was installed and operated at the site over a length of 150 m and a depth of 8 m, to create an anaerobic infiltration zone in which HCH is converted. The construction of the infiltration system was combined with the redevelopment of the site. During passage through the bioscreen, the concentration of HCH in the groundwater decreased from 600 µg/L to the detection limit of the individual HCH isomers (0.01 µg/L) after one year of operation. The concentration of the intermediate biodegradation products benzene and chlorobenzene increased and achieved steady state values of respectively 800 and 2700 µg/L.

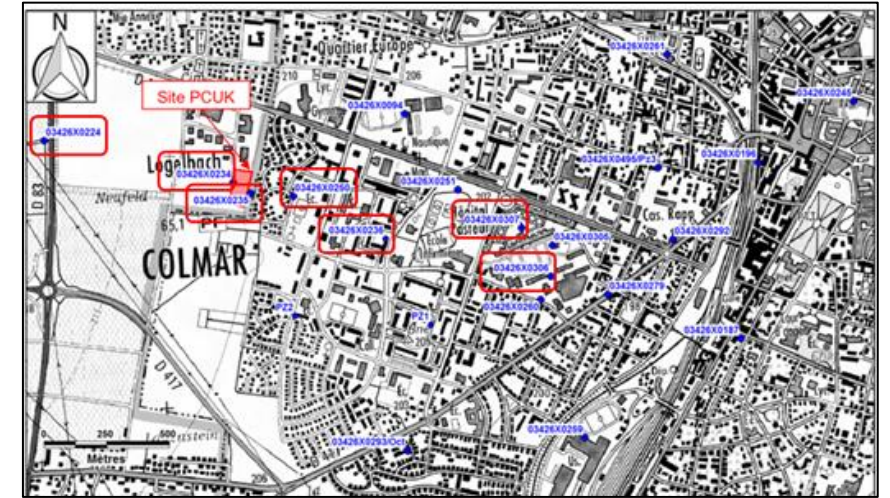
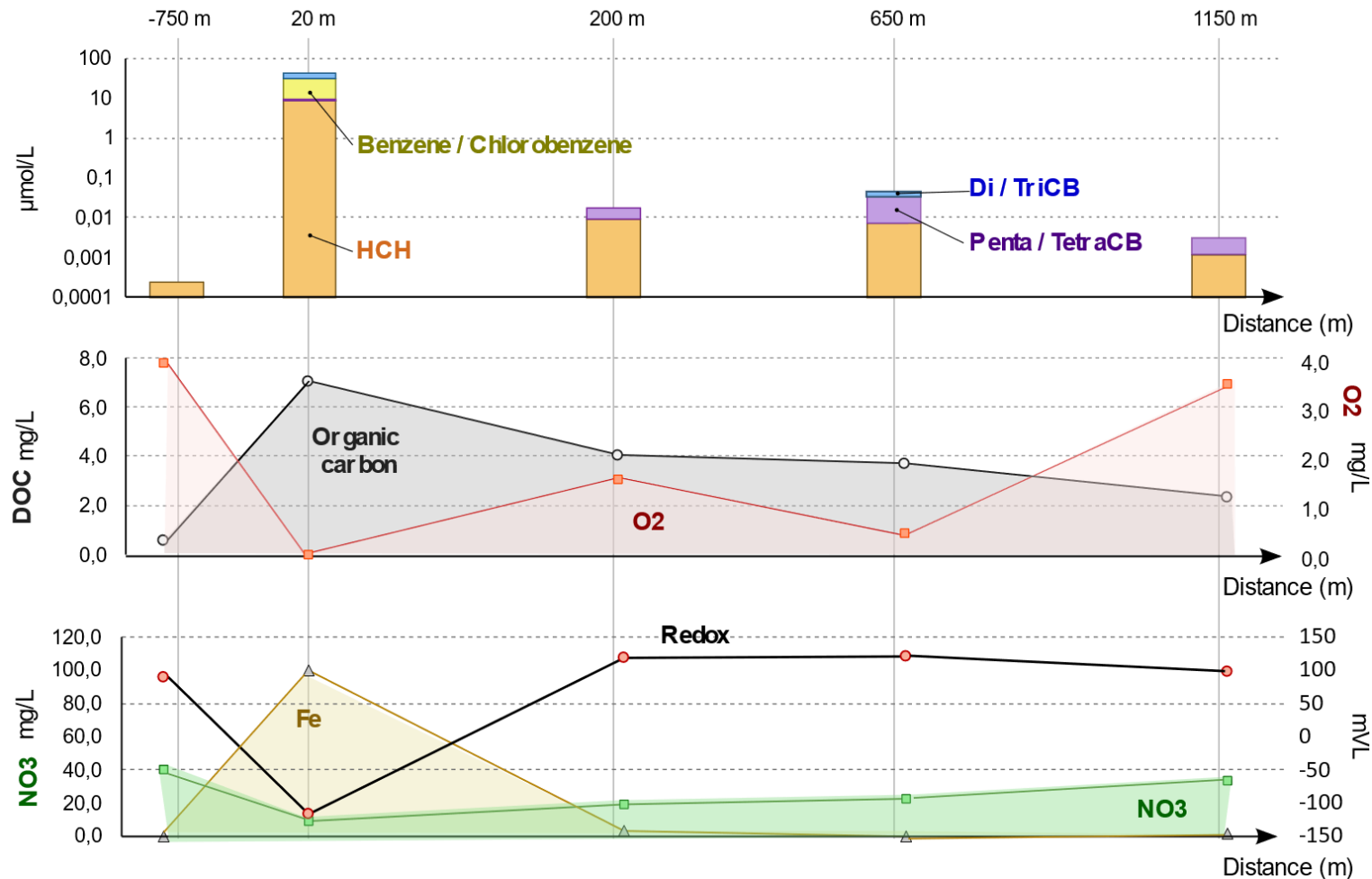
Benzene and chlorobenzene were treated aerobically on site in an existing wastewater treatment plant. By changing the infiltration regime, it is conclusively shown that HCH removal is the result of the biological degradation and stimulated by the addition of methanol as electron donor. To our knowledge, this is the first successful field demonstration of the stimulated transformation of HCH to intermediates in a full scale anaerobic in situ bioscreen, combined with an aerobic on site treatment to harmless end products.



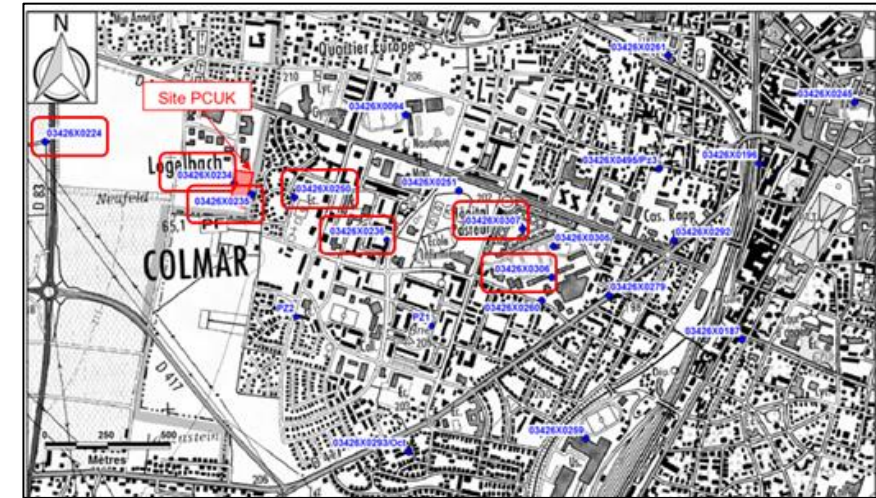
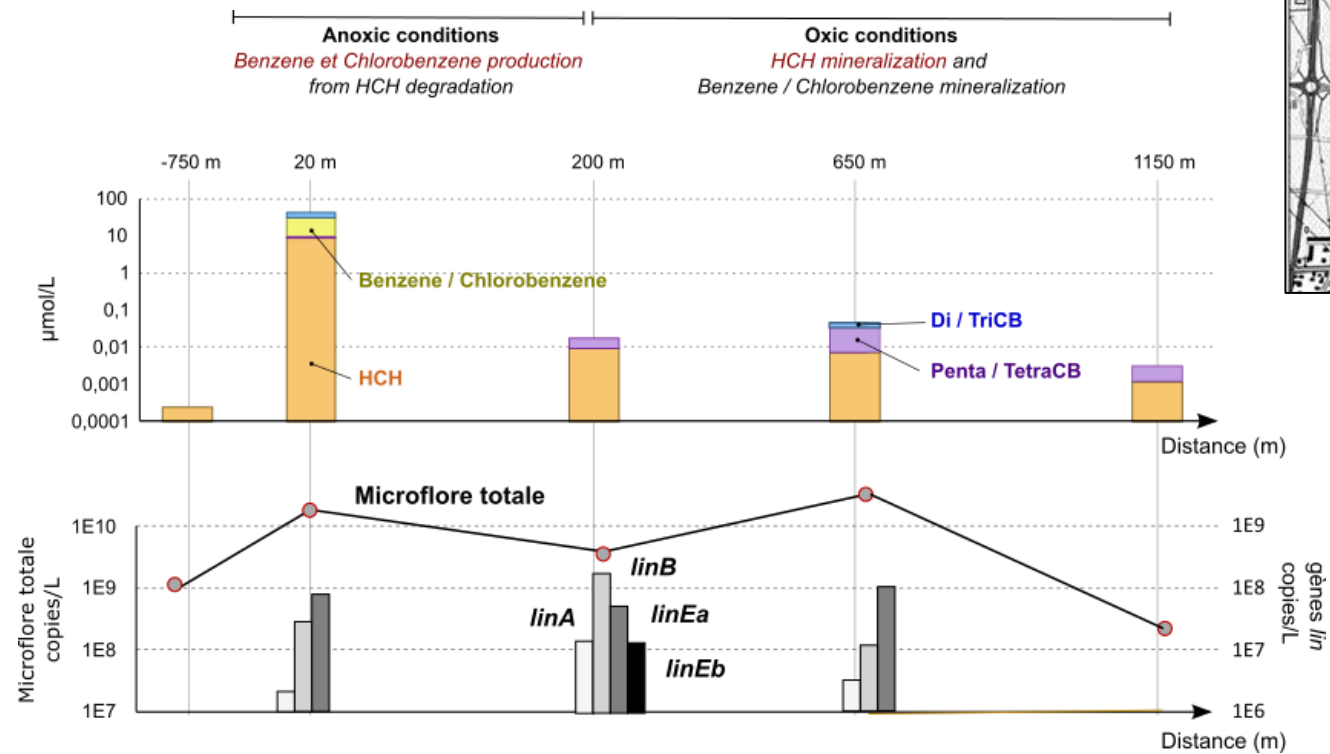
# PCUK site: Characterization of *in situ* biodegradation potential

**Anoxic conditions**  
Benzene et Chlorobenzene production  
from HCH degradation

**Oxic conditions**  
HCH mineralization and  
Benzene / Chlorobenzene mineralization



# PCUK site: Characterization of *in situ* biodegradation potential

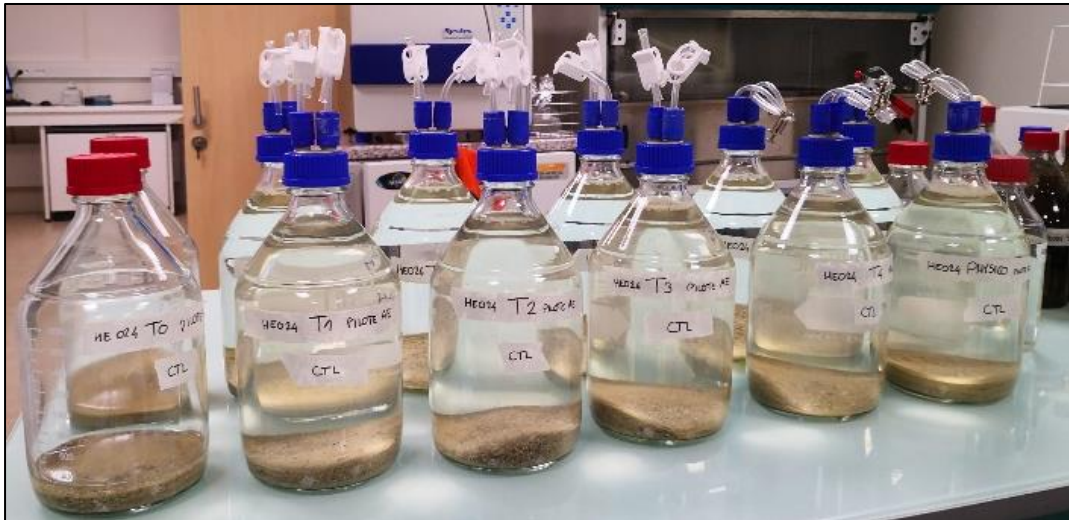




# Laboratory tests

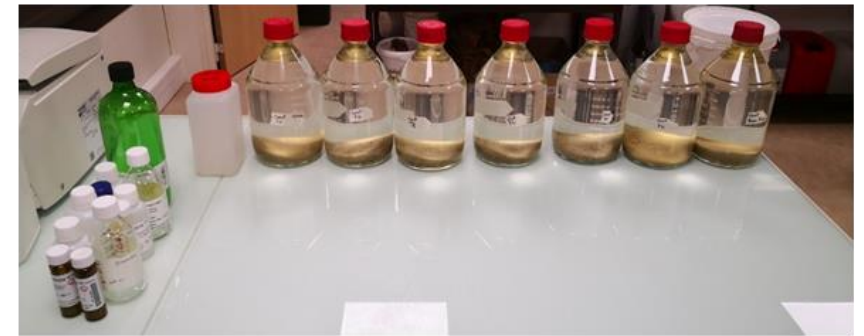
## Aerobic conditions:

- Stimulated:  $\text{H}_2\text{O}_2$  + nitrogen (DAP)
- Control



## Anaerobic conditions:

Contrôle



Stimulée



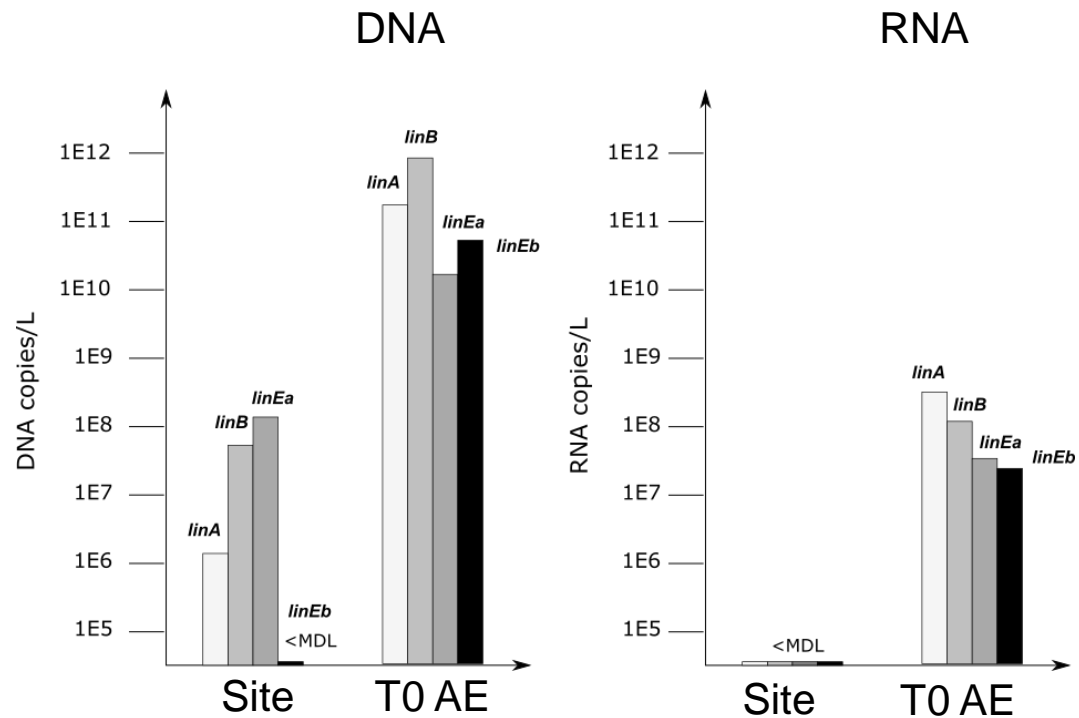
↓ + 0,3% de SC (vol/vol)  
(Lactate/mélasse/eau)



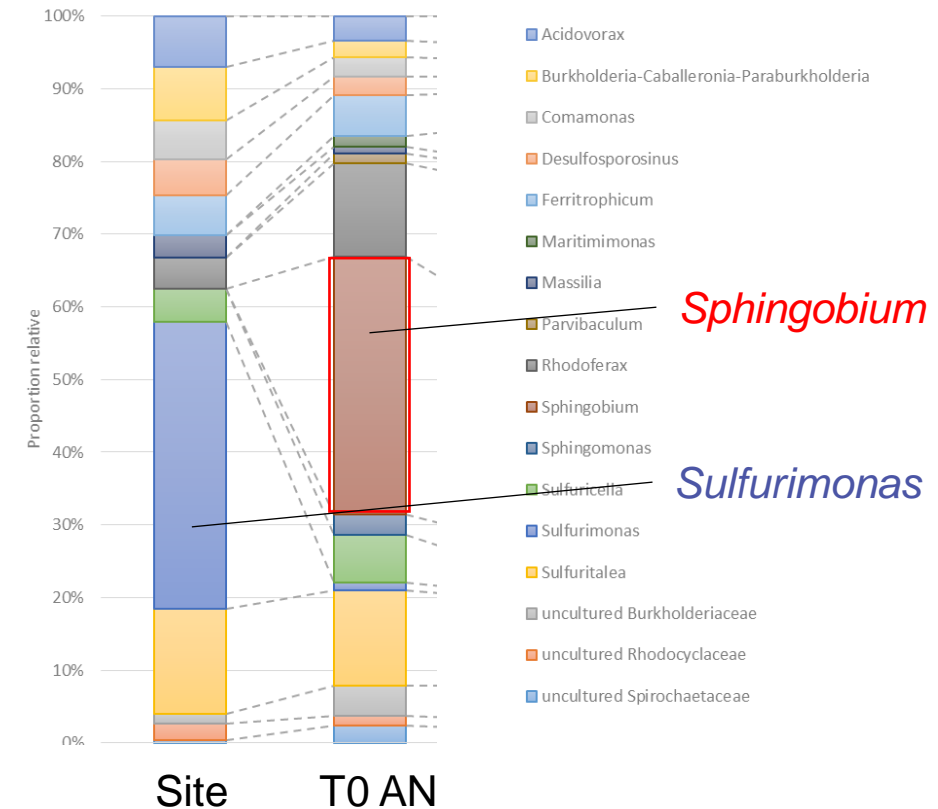


# Fast Growth of *Sphingobium* during the storage period

## Aerobic conditions

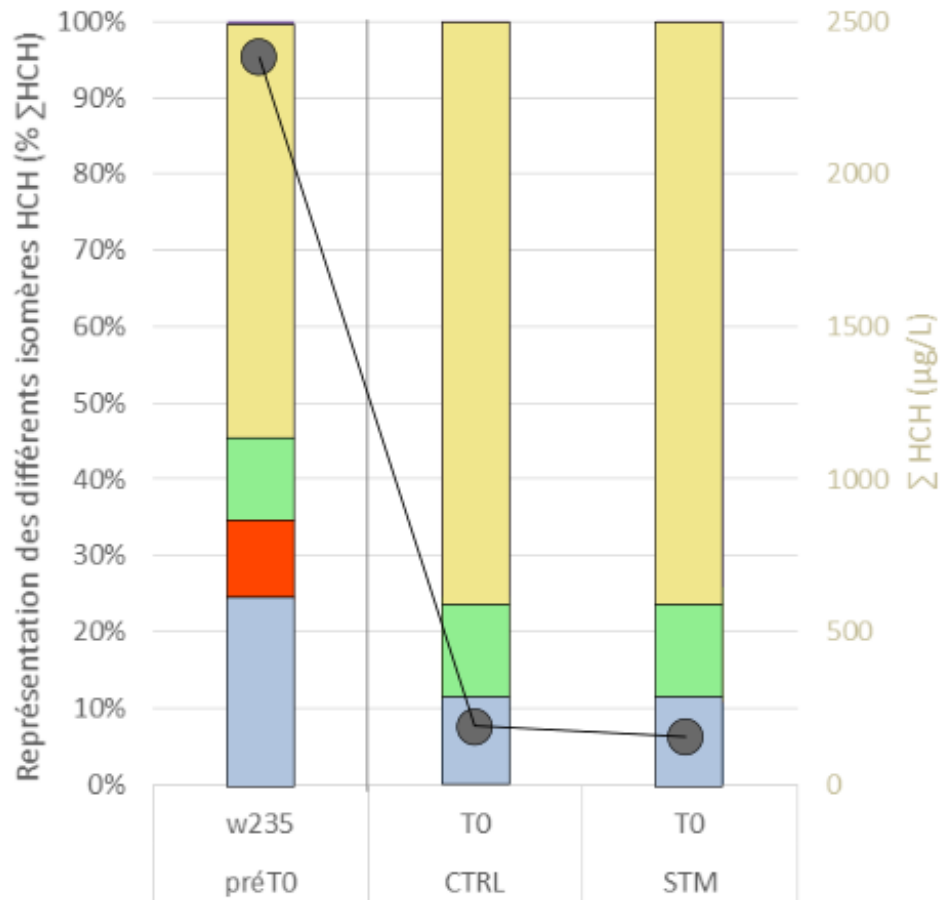


## Anaerobic conditions

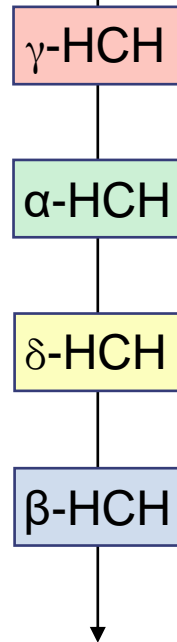


# Laboratory tests: aerobic conditions (4 months)

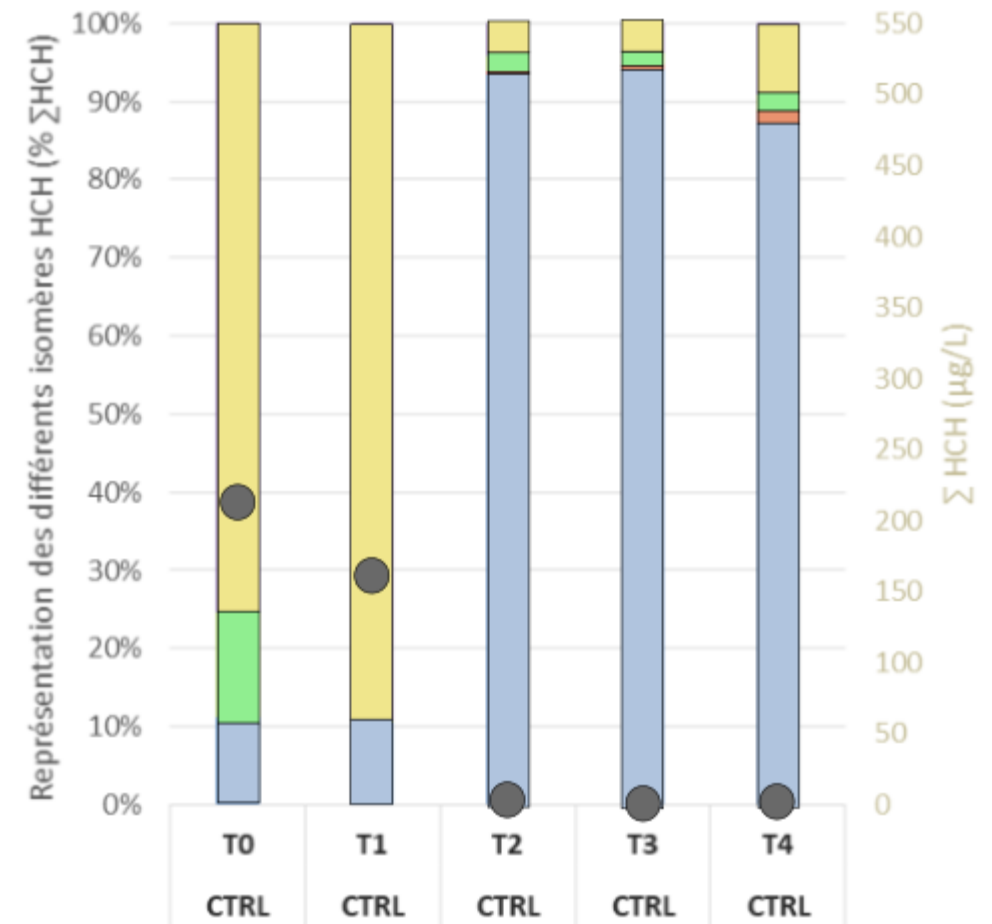
Fast degradation of  $\alpha$ -HCH and  $\gamma$ -HCH during transport of samples



HCH degradation

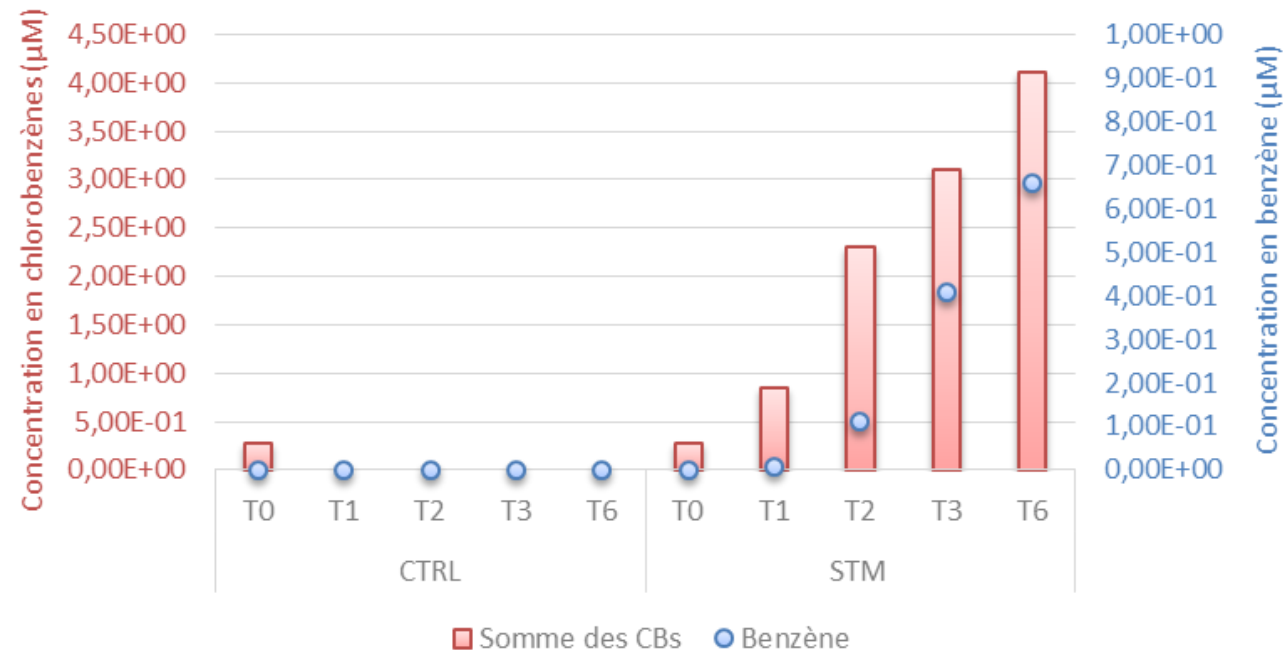


Fast degradation of  $\delta$ -HCH during the tests



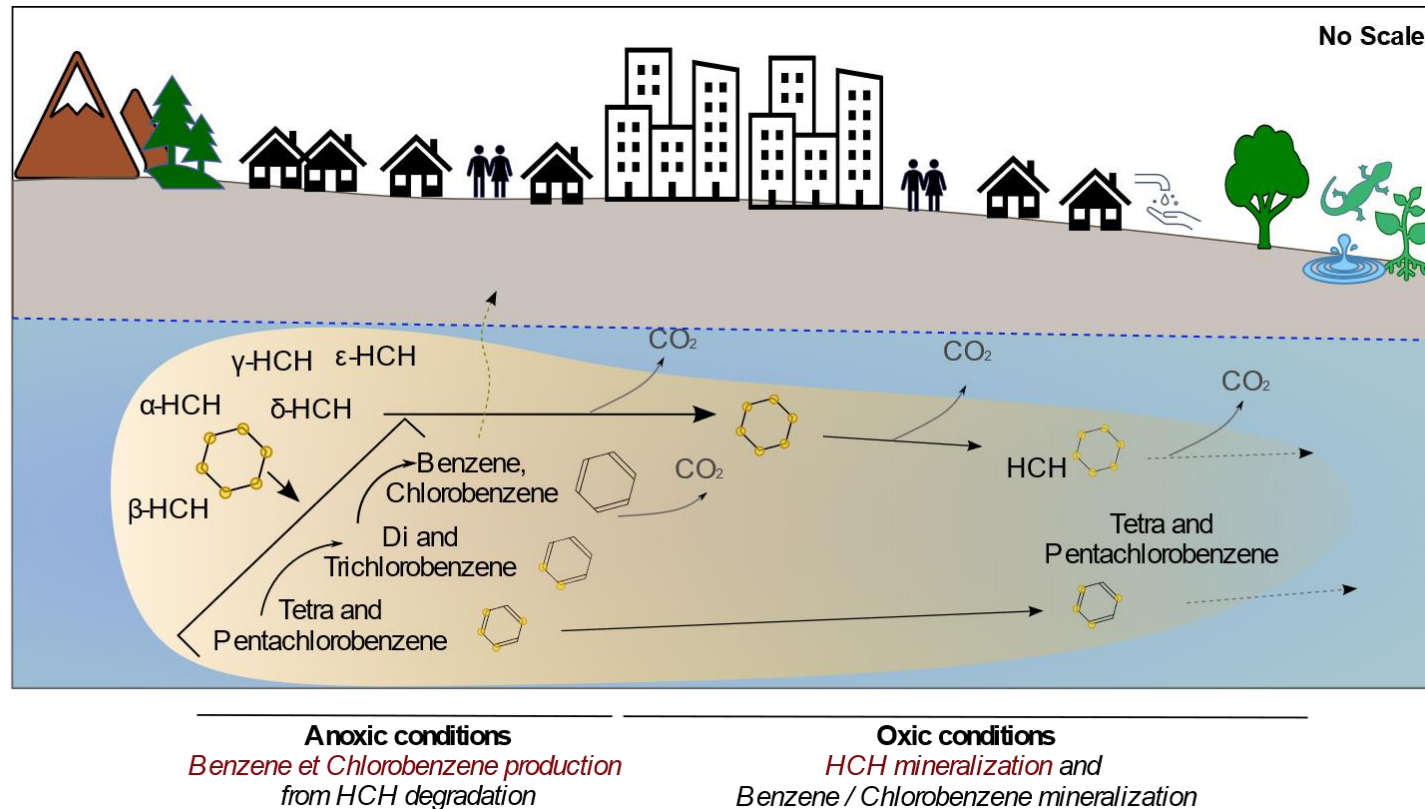
# Laboratory tests: anaerobic conditions (6 months)

- No decrease of HCH concentrations observed;
- **Production of toxic metabolites** under stimulated conditions: benzene, chlorophenols, chlorobenzene....;
- Difficulties to understand the mechanisms for degradation.



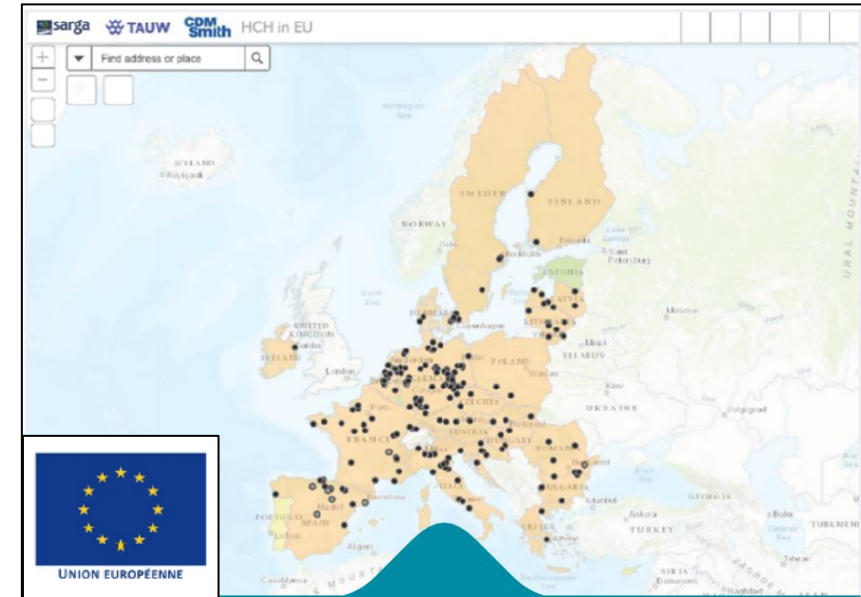
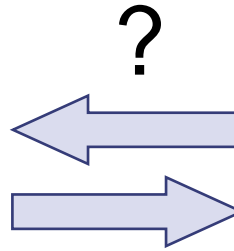
# Conclusions of laboratory experiments

- **Aerobic degradation can be easily stimulated;**
- Anaerobic degradation is partially understood, and can generate toxic compounds;
- Presence of **other non-negligible isomers** in groundwater ( $\epsilon$ -HCH ~ 20%).





# Assessment of the biodegradation potential for the sustainable management of HCH sites



- French government has decided that hazardous waste be excavated (0 to 15m bgl)  
- cost estimated between 22 and 38MEUR;
- Cost for Groundwater treatment by bioremediation is estimated at ~500,000 EUR

- What treatment / solutions for the other sites
- HCH as a recalcitrant pollutant ?
- Potential for biodegradation should be assessed



# Aknowledgements



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