



## Biological in situ remediation

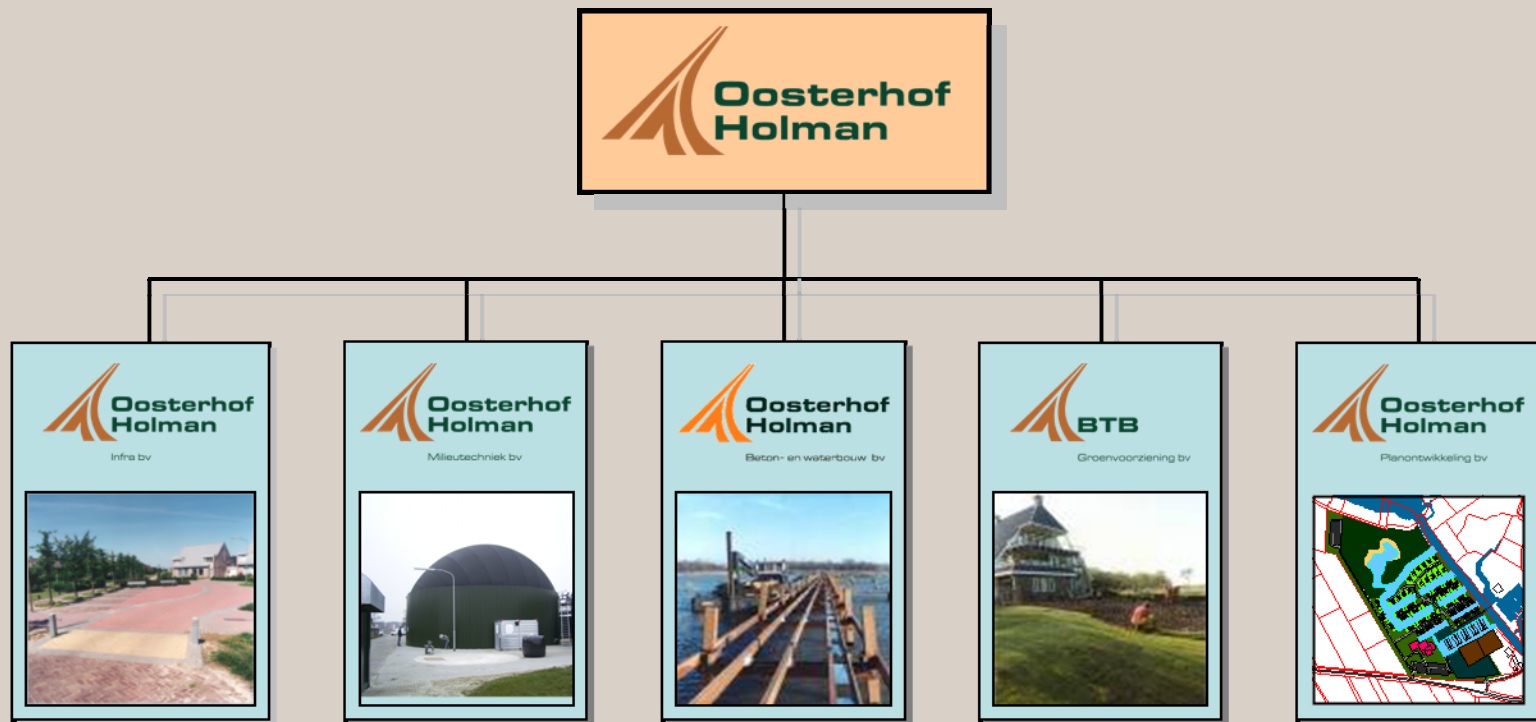
➔ Aleida de Vos van Steenwijk,  
RemTech 2011 (Banff, Canada)



## ➔ Presentation overview

- The companies
- A short introduction
- Case study: remediation in an urban area
- Innovation and development

## ➔ Oosterhof Holman Group 2011



## ➔ Locations



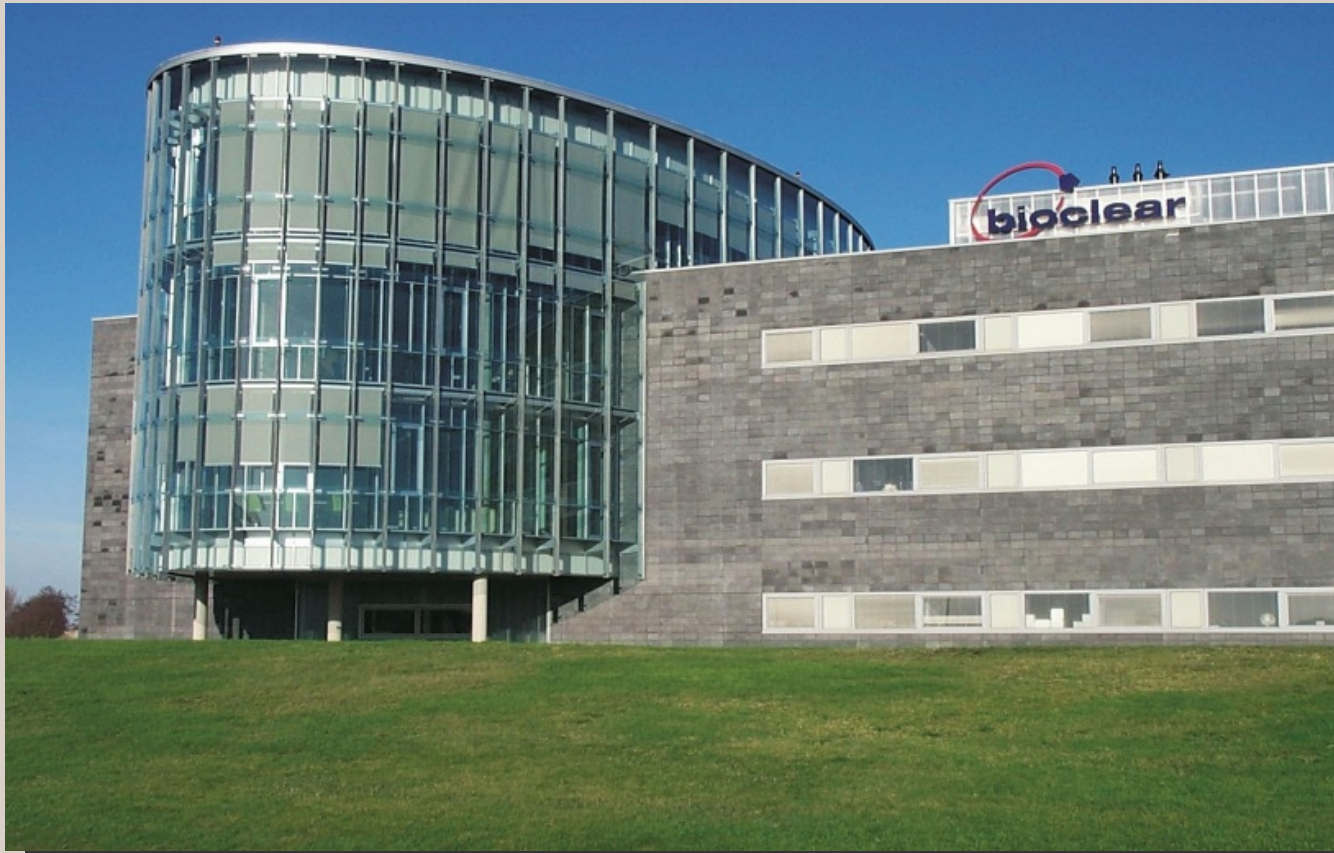
## ➔ Environmental Technology

- Soil and Groundwater Remediation
- Dredging
- Water Purification Plants
  - Conventional systems
  - Membrane systems
- Renewable Energy
  - Biogas
  - Wind
  - Biofuels
- Special Projects
  - “Sustainable Living”





## ➔ Bioclear bv



## ➔ Bioclear bv

- Consultancy and Innovation office for environment and sustainable production
- To make the world **clean**, **sustainable** and **safe** using **biological processes**
- 30 employees, multidisciplinary, teamwork
- A large knowledge network
- Offices in Groningen en Geertruidenberg (NL)



## ➔ Areas of expertise



Soil & Water



Hygiene &  
Safety



Biobased  
Society



Bioclear  
Laboratories



## ➔ Soil & Water



Soil & Water

- In situ bioremediation
  - Natural attenuation
  - Feasibility and design of bioremediation concepts
  - Performance and supervision of biological soil remediation
  - Sustainable use of soil / cycling
  - Buffering capacity of soil (ETS)
- Ecosystem services
- Ecological quality and risk assessments
- Knowledge and technology development
- Monitoring and control of biological processes

## ➔ Hygiene & Safety



### Hygiene & Safety

- Prevent unwanted processes (detection, monitoring and control)
  - biofouling
  - biocorrosion / MIC
  - Pathogens
- Monitoring and control biological processes
  - development custom made analyses
  - sampling strategies
  - interpretation
  - risk-assessments
  - preventative strategies

## ➔ Biobased society



Biobased  
Society

- Monitoring and control biological processes
- Valorisation of wastes and residues
  - biogas
  - green chemicals
  - fertilisers
  - market research
- Recycling of nutrients (NPK)
- Chain development
- Knowledge and technology development

## ➔ Bioclear laboratories



Bioclear  
Laboratories

- Routine analyses for monitoring and control
- Analyses for ecological and soil quality
- Analyses for product quality / safety
- Development of custom-made analyses
- Validation and implementation
- Courses and workshops
- Contract research





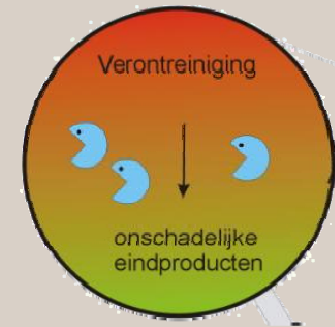
Short introduction  
*In situ bioremediation*



## ➔ Biological degradation of CVOC is possible?

### **In the nineties:**

Biodegradation is difficult and slow



### **From 1997:**

*Dehalococcoides* bacterium (Maymo-Gatell *et. al.*)

Research into optimum conditions for biodegradation

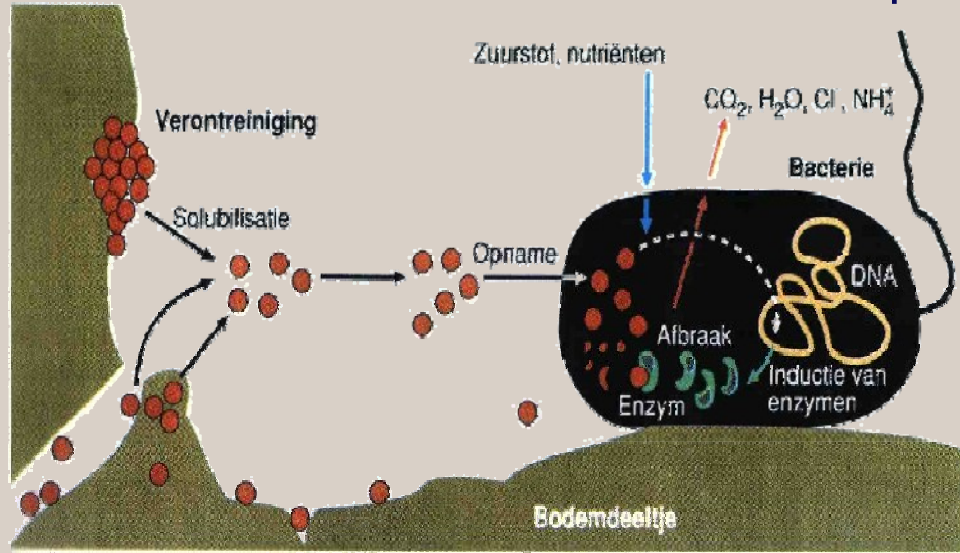
### **Now:**

Biological in situ remediation is proven technology

## ➔ Optimal conditions



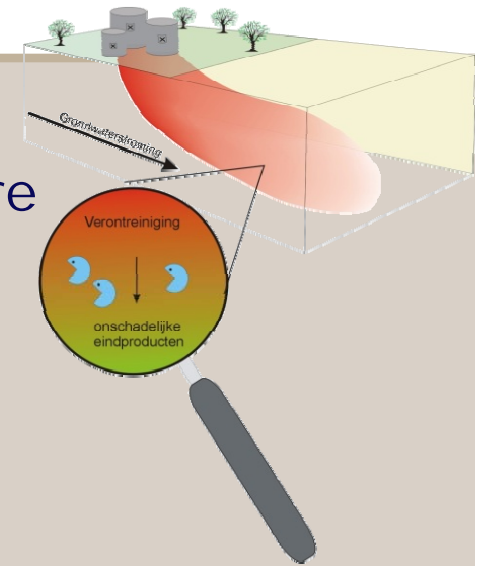
- Presence of *Dehalococcoides* bacteria
- Right conditions in soil
- Adequate nutrients and carbon source



## ➔ Natural or a helping hand?

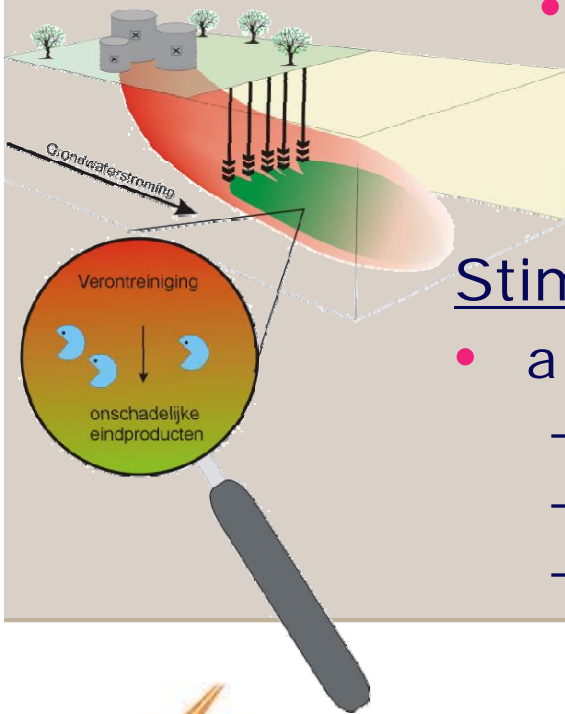
### Natural Attenuation (NA)

- naturally occurring conditions are suitable
- no active measures required
- this situation is rare!



### Stimulated degradation

- a limitation inhibits natural attenuation
  - create optimal conditions
  - add nutrients and/or carbon source
  - add bacteria → **bioaugmentation**







## Case Study

*TCE concept in centre of  
The Hague, the Netherlands*

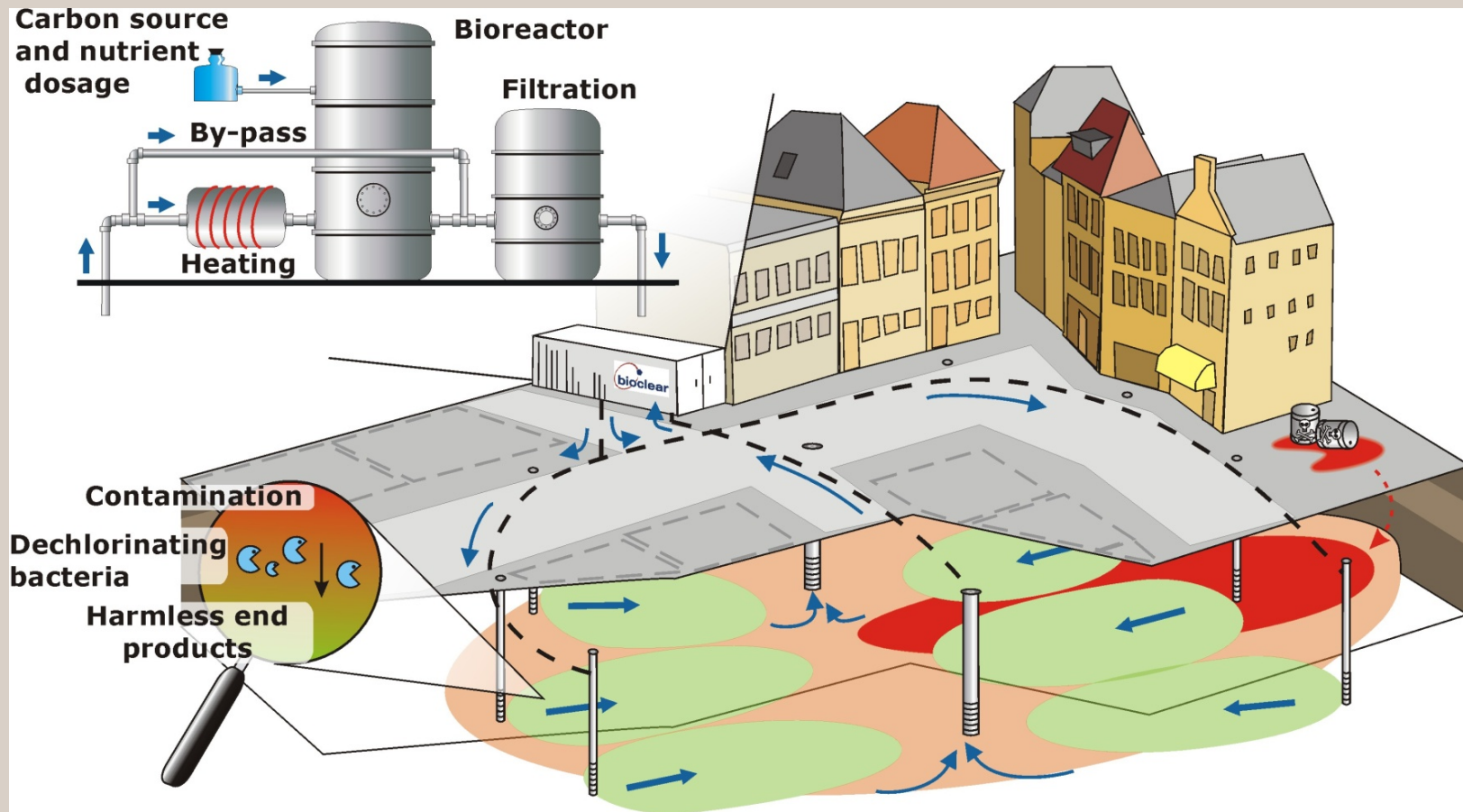


## ➔ Background information location

- Urban area (houses and shops)
- Renovated and protected buildings
- Contamination with chlorinated solvents (PCE and TCE)
- Likely cause: sewer leak containing waste from dry cleaners



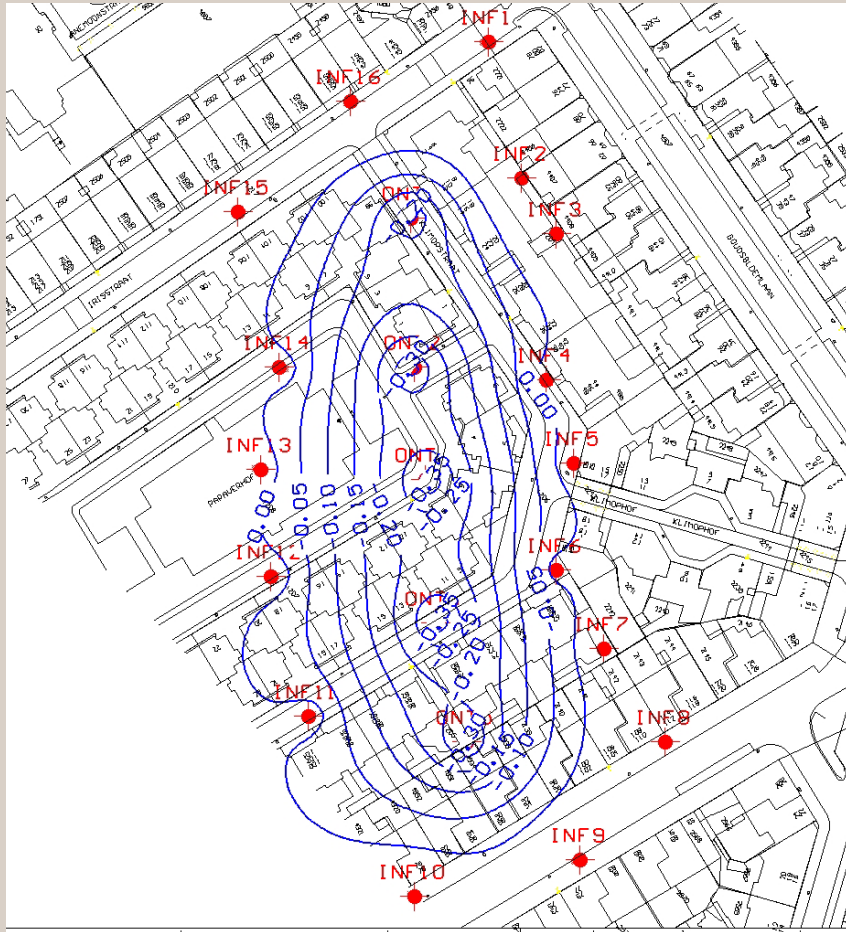
## ➔ TCE concept (bio-augmentation)



## ➔ Remediation approach

- Active phase
  - Create right conditions in soil
  - Sufficient carbon source for reductive dechlorination
  - Sufficient and active dechlorinating population
- Passive phase
  - In situ reductive dechlorination until remediation aim is achieved
- Monitoring phase
  - Has a stabile end situation been achieved...?





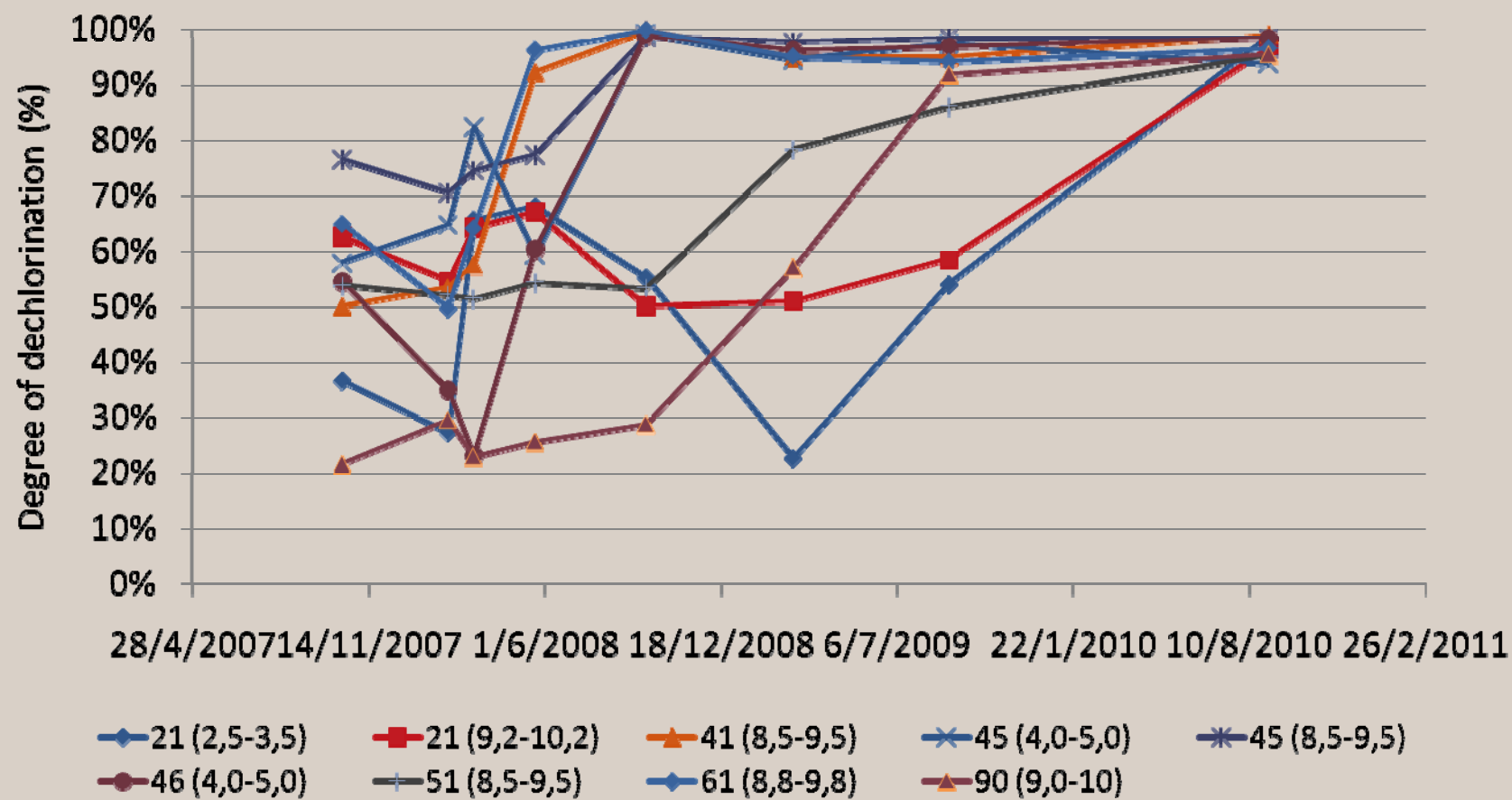
- 80.000 m<sup>3</sup> > I-value
- Up to 1.000 µg/l
- 10 m-sl
- Medium-grained sand
- 5 Extraction wells
- 16 Infiltration wells
- Closed water balance
- Water table change < 42 cm
- 10 m<sup>3</sup>/hour ext/inf

## ➔ Remediation Phases

- Active Phase: Dec. 2007 – Sept. 2008
- Passive Phase: 2 Years,  
goal: PCE=20 µg/l; TCE=262 µg/l; DCE=10 µg/l; VC=2,5 µg/l



## → Results



## ➔ Results

- After 1 year: 6 out of 9 wells < remediation target
- After 2 years: 8 out of 9 wells < remediation target
- Sept. 2010: Site closure
- In-situ treatment of TCE plume within 3 years
- 80.000 m<sup>3</sup> soil volume remediated
- Total actual costs € 441.000 (= 5,51 euro / m<sup>3</sup> )

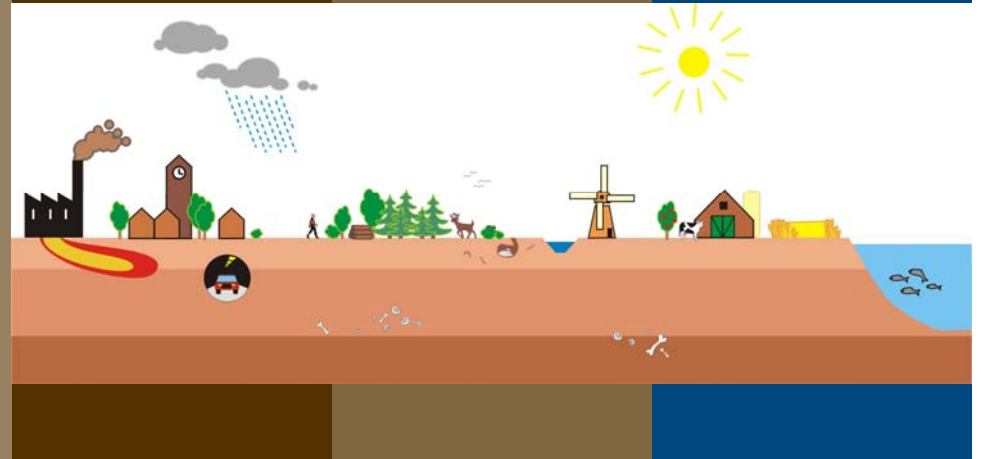






## Sustainability

- *TCE concept (as performed)*
- *Pump & Treat (hypothetical)*
- *ISCO using Ozone (hypothetical)*

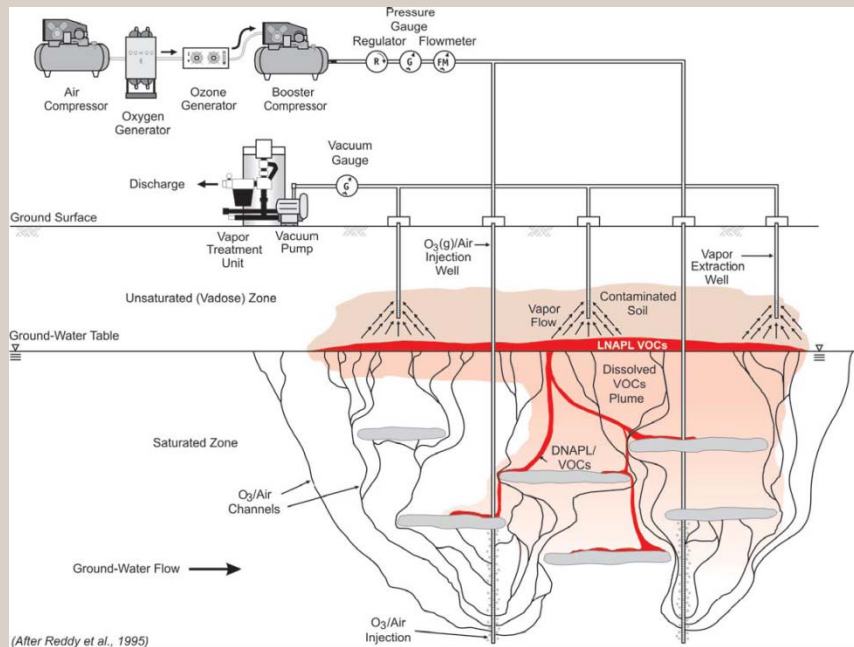


## ➔ Alternative 1. Pump & Treat



- 17 Infiltration wells
- 13 Extraction wells
- 10 m<sup>3</sup>/hour
- Closed water balance
- Stripping system 8 KW
- Duration 10 years
- Costs est. € 900.000

## ➔ Alternative 2. Ozone



- Ozone sparging
- Analogous to air-sparging
- NOD: 15 g O<sub>3</sub> / m<sup>3</sup> soil
- Sparging efficiency: 5 %
- 24.000 kg ozone
- PLI + SVE system 12 KW
- Duration 1 year
- Costs est. € 800.000

## ➔ Method for comparison: MCA

### SOCIAL EFFECTS

(smell, noise, hindrance, potential dangers, chance of calamities, chance of damage )

### ENVIRONMENTAL EFFECTS

(air, soil, groundwater, ecology, waste production, residual contaminations)

### RESOURCES AND MATERIALS

(use and reuse of water, groundwater, energy, fuels (transport), chemicals, materials)

### CLIMATE EFFECTS

(Carbon dioxide & methane emissions)

Compare remediation options on these 4 themes

## ➔ Summary MCA results

		Alternative 1: Pump&Treat	Bio-augmentation (TCE concept)	Alternative 2: Ozone
	weight	score	score	score
Social effects	1	6,1	7,1	6,0
Environmental effects	1	8,7	9,0	9,0
Resources	1	8,0	9,5	8,0
Climate	1	6,0	9,0	6,0
Total		<b>7,2</b>	<b>8,7</b>	<b>7,3</b>





## Innovative developments

*Monitoring and control of microbial processes*



## ➔ Monitoring key microbial processes

*"A good understanding of the capacity for natural attenuation is the key to feasible and practical bioremediation"*

- Sampling
- Analysis

## ➔ Groundwater sampling

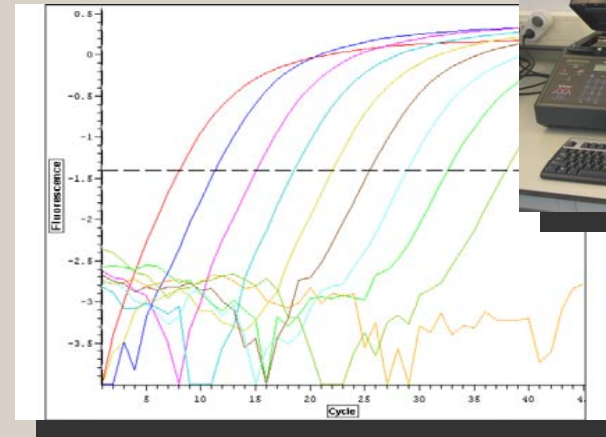
- “Standard method”
- Large volume sampling (dialysis method)
- BACTRAPs<sup>©</sup>



## ➔ Who's there and what are they doing?

### Quantification of micro-organisms and their activity

- Q-PCR:
- Flexibility
  - Accurate quantification
  - Activity measurements
  - Sampling
  - All sample types
  - Objective determination
  - Quality control

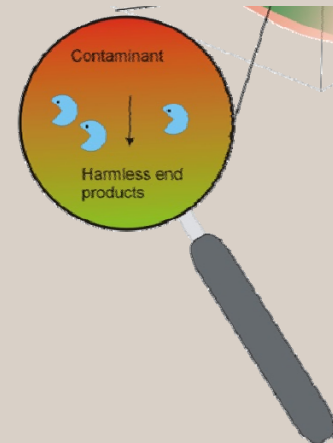


*"Most species cannot be cultured as their growth conditions are unknown or cannot be reproduced in the laboratory. The percentage of cultivable micro-organisms depends on the complexity of the sample and can vary from 90% to as little as 1%. The risk of false negatives therefore is significantly increased."*

## ➔ Degradation processes of chlorinated ethenes

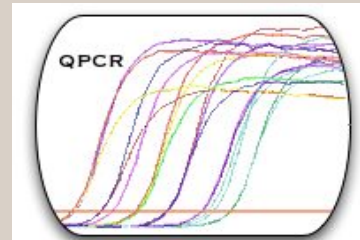
- Reductive dechlorination

- Methanogenic conditions
- Degradation of PCE, TCE, DCE & VC
- Bacteria involved: *Dehalococcoides* spp
- Key enzymes involved: VC-reductase



- Micro-aerophilic oxidation

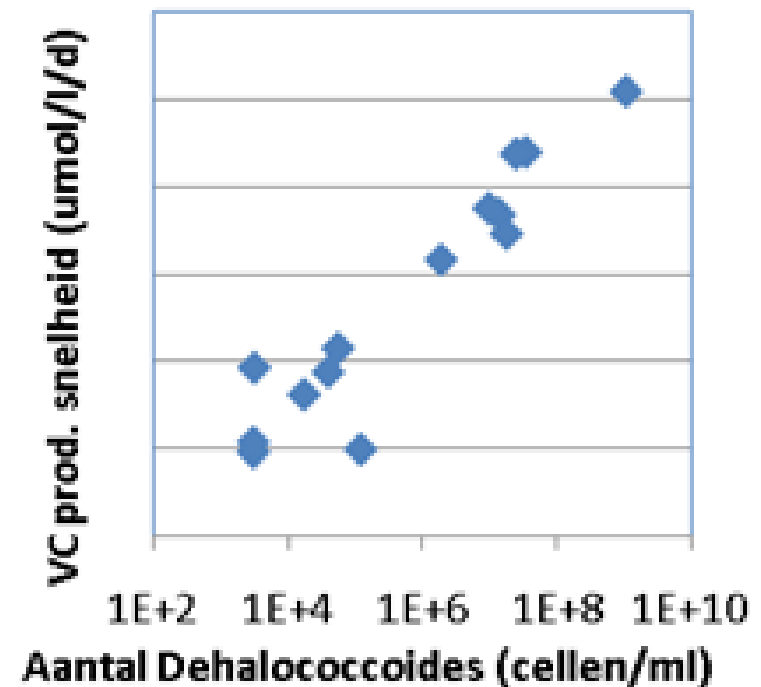
- For example Iron reducing conditions, low oxygen (as low as 0,03 mg/l)
- Degradation of DCE & VC
- Bacteria involved: *Polaromonas* (*cis*-DCE degradation)
- Key enzymes involved: *EtnC* & *EtnE* (VC degradation)





## ➔ Reductive dechlorination

Correlation between number of *Dehalococcoides* cells and degradation in groundwater per m<sup>3</sup> per day



## ➔ Micro-aerophilic oxidation

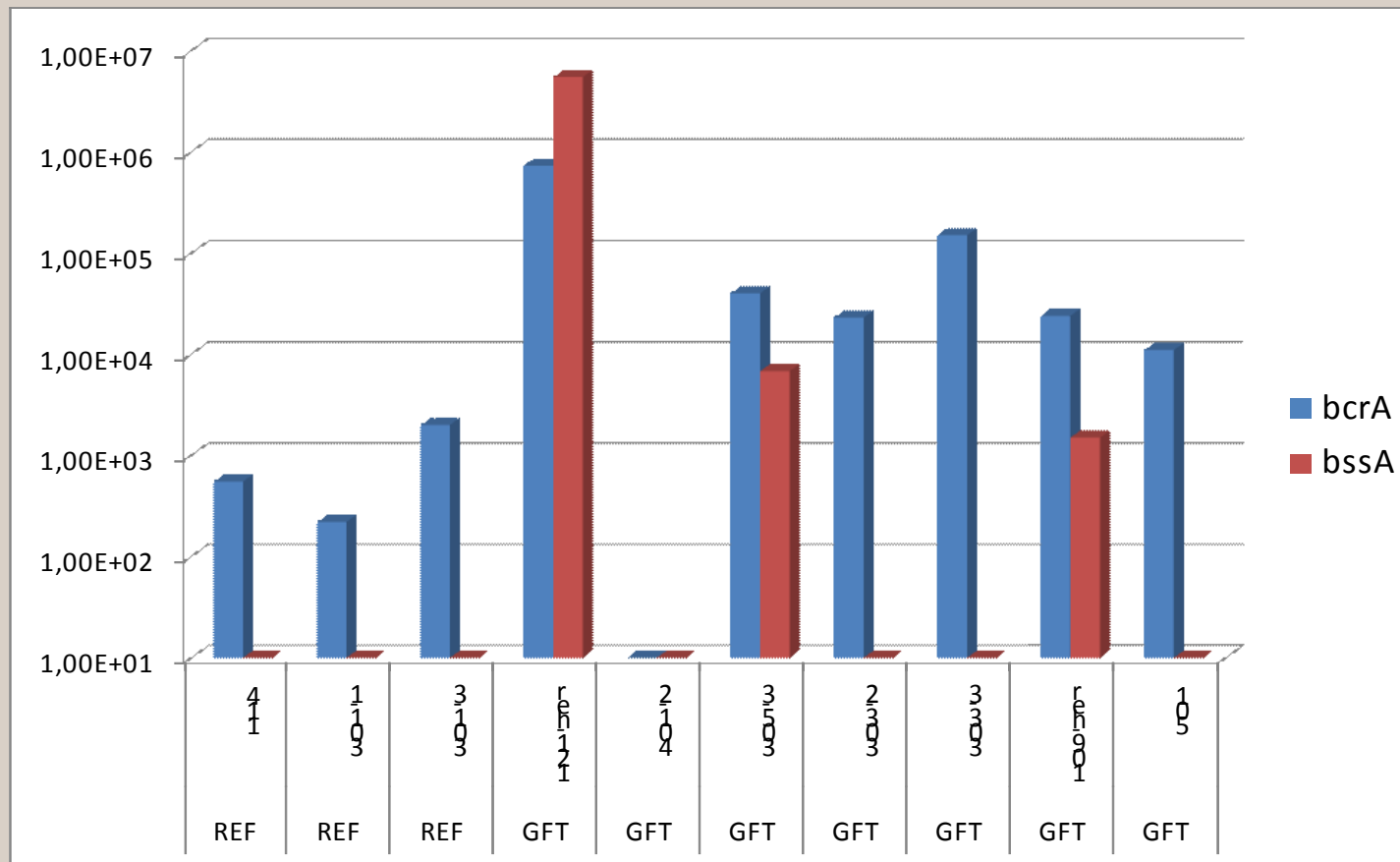
Monitoring well		Pb61-A	Pb53	Pb67	Pb86	Pb86	Pb106	Pb 106
Depth		26 m-mv	38 m-mv	26 m-mv	14 m-mv	24 m-mv	20 m-mv	35 m-mv
<i>Dehalococcoides</i>	cells/ml	<	<	<	<	<	3,9x10 <sup>4</sup>	2,1x10 <sup>5</sup>
<i>EtnC</i>	cells/ml	<	7,4x10 <sup>3</sup>	1,6x10 <sup>1</sup>	<	<	<	8,4x10 <sup>2</sup>
<i>EtnE</i>	cells/ml	<	9,7x10 <sup>2</sup>	<	<	<	<	1,0x10 <sup>3</sup>
<i>Polaromonas</i>	cells/ml	<	<	<	<	<	<	<

## ➔ Anaerobic degradation of aromatic compounds

### Degradation of BTEXN

- BcrA enzyme: breaks open ring structure aromates
- BssA enzyme: first steps of degradation of aromates. Adds a molecule (fumarate) to methylated aromatic compounds (toluene en de xylenes). Likely also involved in degradation of benzene.
- NcrC enzyme: only recently discribed in literature. Same function as BcrA enzyme, but then for naphtalene

## ➔ Anaerobic degradation of aromatic compounds



## ➔ Conclusions

- In situ bioremediation is proven technology
- Bio-augmentation highly sustainable: low energy, low CO<sub>2</sub>, low cost & very effective
- Continuing innovation and development to further improve in situ bioremediation





*Thank you for  
your attention*

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