



D1d E-USE(aq)

Technical performance & monitoring report

Danish pilot

Climate-KIC is supported by the
EIT, a body of the European Union





Summary Danish Pilot in Birkerød

The site Hammerbakken 10 in Birkerød, Denmark is highly polluted with chlorinated solvents, especially TCE is a dominant contaminant. In laboratory studies by Environmental Technology Wageningen University it was shown that biodegradation of chlorinated solvents could be stimulated at low redox conditions at elevated temperatures like in ATES systems. The hot spot contaminated site at Hammerbakken 10 was selected by the Capital Region of Denmark (CRD) to demonstrate the stimulation of biodegradation of chlorinated solvents at pilot scale. The pilot was realized by the end of December 2017 and monitored thereafter in 2018. The ATES recirculation systems showed to operate as expected, whereas biodegradation was not stimulated in the first 6 months, as the redox was too oxid to allow biodegradation of chlorinated solvents.

In June, electron donor was added to lower the redox. After July the first reductive dechlorination was monitored up to 30% dechlorination. However only DCE was formed and no significant other products as VC and finally ethene were formed.

In early November a DHC culture was added, which readily resulted in a high dechlorination degree of already about 78% in monitoring well 1 within 2 weeks.

Until March 2019 the wells will be further monitored for the reductive dechlorination of chlorinated solvent (mainly TCE).

From the pilot experiment in 2018 it can be concluded that the ATES system as such functioned well without clogging by the presence of measurable oxygen concentrations in the first half year of operation. The applied ATES system showed to be fit to stimulate biodegradation of chlorinated solvents at optimized conditions like addition of electron donor and addition of a specific dechlorinating culture.



1. Introduction

General

Climate Knowledge and Innovation Communities (Climate-KIC) is an initiative supported by the European Institute for Innovation and Technology (EIT) with the aim to overcome the challenges of bringing climate-focused innovation to market. One of the research programmes of Climate-KIC concerns sustainable production systems.

Aquifer Thermal Energy Storage (ATES) is a cost-efficient sustainable production system that involves storing energy resulting from cooling and heating in water-bearing soil layers and acquiring heat and/or cold from extracted groundwater whenever needed.

In recent years, approximately 2500 ATES systems have been successfully applied in the Dutch subsurface (with an estimated yearly turnover of 270M€) (CBS, 2012). Due to this success, it is expected that remunerative application will happen elsewhere in Europe, but to date the widespread application of ATES systems is hampered by socio-economic, legal and technical barriers.

Together with national and international partners, Deltares has conducted pilot studies in the Netherlands, Italy, Spain, Belgium and Denmark to prove the attractiveness and potential of large-scale ATES applications. One of these pilot studies concerns an ATES system at Hammerbakken 10 in Birkerød, Denmark where the combination of ATES and groundwater remediation is investigated in a highly VOCI polluted site.

a. Barriers to overcome and issues to address

Several barriers for the application of ATES that are addressed in Pellegrini et al, 2018 are also at stake in the Danish Pilot in Birkerød. As the experience in Denmark is limited with ATES systems, special attention is given to the cooperation between contracting companies (Table 1), consulting companies (Ramboll), the special niche companies (Bioclear earth, IF Technology) and knowledge institutes (Deltares, Wageningen University) (1). Compared to Pellegrini et al. (2018) this case is special as the groundwater is contaminated with high VOCI concentrations in groundwater in a specific Danish soil type. The Capital Region of Denmark (CRD) is concerned about the spreading of groundwater contamination, as nearly all drinking water in Denmark is prepared from groundwater (2). In other countries, like the Netherlands, several drinking water companies see ATES systems as a threat for the groundwater quality. Due to mixing of large groundwater volumes, they were afraid of the pollution of pristine groundwater. Such pristine groundwater is in the Netherlands the main source for the preparation of drinking water. Another barrier for the introduction of ATES can be the lack of adequate legislation especially in case of groundwater contamination (3).



In this pilot attention is given to all these three barriers: 1. The barrier upon the lack of experience is addressed by team-meetings with all the different partners during the design phase of the pilot; 2. Upfront of the design of the pilot, lab-studies were performed at WUR-ETE, which resulted in publications in international peer reviewed journals and a PhD thesis in 2015. In this research, the proof of principle is demonstrated that especially in the warm well of ATES systems the bioremediation of frequent reported contaminants by chlorinated solvents as PCE, TCE, DCE and VC can be accelerated by more than factor a 10 compared to Natural Attenuation. If this laboratory research could also be confirmed in real pilot studies this is expected to lead to the acceptance of the ATES-Bioremediation concept by water-boards and drinking water companies of the application of ATES systems.; 3. For this specific case no adaptations of legislation was necessary as the CRD is allowed to perform pilot studies. Further CRD visited the Netherlands where area based policies are developed that allow new developments in which a standstill in spreading of contaminants is realized or in which the environmental quality reaches a better status.

The emphasis in this pilot is especially directed to investigate the natural sciences based barriers, as this barrier could be a showstopper for the combination of ATES and Bioremediation.

b. Partners involved

Deltares and WUR-ETE from the Climate KIC project E-Use aq are involved in the Pilot Birkerød in Denmark together with CRD (Problem owner and main financier of the Pilot), Ramboll (Danish consultant, hired by Capital Region), IF Technology (Dutch engineering company, design ATES systems), and Bioclear earth (Dutch company, design and performing innovative bioremediation projects). Further regional Danish partners were involved as presented in Table 1.

Table 1. : List of partners and roles performed during this project

Partner	Role
Capital Region of Denmark (CRD)	The orderer of the pilot, responsible for groundwater quality
Ramboll	Consulting, leading the realization of the pilot
Deltares	Monitoring, physical, chemical and DNA analyses in groundwater samples, data interpretation
Bioclear earth	Development of DHC culture, placement and monitoring of soil in-situ mesocosms
IF Technology	Design of ATES wells, flow calculations
Wageningen University, Environmental Technology	Develop generic knowledge, data interpretation and evaluation
Hans Frisesdahl A/S	conduct the design and installation of the system
Wicotec Kirkebjerg Teknikentreprenør	Installation of the heat exchanger
Eurofins Miljø	Analysis of groundwater



c. Organizational history

At the AquaConSoil conference in June 2015 in Copenhagen the results of the WUR-ETE PhD study by Zhuobiao Ni (Ni 2015) were presented. This resulted to a first contact by WUR-ETE (Tim Grotenhuis) and CRD (John Flyvbjerg). This was the starting point for a literature study that was carried out from December 2015 till May 2016 (Report: literature review Aquifer Thermal Energy Storage (ATES) and Enhanced Reductive Dechlorination (ERD), May 2016 by Ramboll, Appendix 1).

In September 2016, Tim Grotenhuis (ETE-WUR) spent his sabbatical leave at The Danish Technological University (DTU) in Copenhagen, where he gave presentations to DTU, Ramboll and Capital Region of Denmark about the combination ATES and Bioremediation. Further, he had meetings with scientists of DTU as well as with people at Capital Region of Denmark and Ramboll. In these meetings, the project E-Use aq was addressed as it focusses on solutions to overcome barriers in the application of ATES. CRD started after these meetings together with Ramboll the selection of a proper site for a pilot on ATES and Bioremediation in Denmark.

With the budget of E-Use aq a visit of the Danish parties to the Netherlands was organized on 20&21 March 2017 in which a session at the Ministry of Environmental affairs upon policy development related to contaminated groundwater and soil was included to discuss adequate policies for application combined with bioremediation (barrier 3). The pilot in Utrecht was visited as well as the new shopping mall under construction (Hoog Catharijne and Rabobank head office in Utrecht. At Rabobank, experiences were shared with their ATES system (designed by IF Technology). In a workshop, presentations by the municipality of Utrecht (financing the Pilot Utrecht) were given on the concept 'Biowashing machine' as well as on new areal-based policies in which contaminated sites are considered in a coherent way (related to barrier 3).

CRD announced at the end of the visit that they want to invest in a pilot in Denmark and invited Deltares, WUR-ETE, Bioclear earth and IF Technology for a visit to Copenhagen in May 2017.

In May 2017 the site at Hammerbakken 10, Birkerød, Denmark was selected by CRD, based upon information collected by Ramboll and in consultation with Deltares, WUR-ETE, and IF Technology and Bioclear earth. The characteristics of the site like type and concentrations of contaminants and soil type, are representatives for many sites in the Capital Region of Denmark.

On 11&12 May 2017 Deltares, IF Technology, Bioclear and WUR-ETE to discuss all the information that was collected about different locations in the CRD area visited Copenhagen.



This information led to the conclusion that the location in Birkerød (Hammerbakken 10) was the best for the pilot. The following in depth discussion focussed on: 1. Site data; 2. Goals; 3. Pilot test design - system design; 4. Pilot test design - pre investigation; 5. Monitoring plan; 6. Risk assessment; 7. Next steps. In addition, Hammerbakken 10 was visited and inspected for fitness for a pilot. As IF Technology and Bioclear earth have a lot of experience with implementation of ATES systems, either bioremediation it was decided that these companies were included in the design and realization of the pilots together with Rambøll (addressing the barrier of limited experience (barrier 1)). Further, the sound approach of the concept was guaranteed by the input of knowledge and evaluation and interpretation of monitoring data by Deltares and WUR-ETE. The costs for the pilot Birkerød of more than 330.000 Euro are funded by CRD.

Thereafter the pre-design was made and preliminary investigations at the site were performed, which were ready by the end of September 2017. Unfortunately, these results showed a relative high oxygen concentration in the site, although it was expected that that mixing the shallow oxic water with the more anaerobic deeper water would lower the redox to a more suitable level. The relatively high oxygen concentration is regarded as a major threat for the reductive dechlorination process that is needed for the biodegradation of the chlorinated solvents in the site. This problem can be overcome by the addition of electron donor, which unfortunately will lead to extra costs. Unless these findings it was decided to continue with the design for the pilot, as this site is regarded as a representative case for the area of the CRD. Therefore, IF Technology made main efforts in the design of the ATES system and facilities were included upon information by Bioclear earth to be able to take measures to stimulate and monitor bioremediation in the pilot.

Rambøll and CRD realized the installation and construction of the pilot Birkerød in December 2017.



Figure 1 Construction of pilot at Hammerbakken 10, Birkerød in December 2017. Upper left and lower picture show the installation of the cold well at the south side of the building. The upper right picture shows the 'Bactraps' that are installed in some of the wells to sample immobilizing bacteria that are present in the ground water.

In January 2018, the ATES system was started and monitoring began. Ramboll presented the first results in May 2018.

2. Pilot description

a. Site depiction and pilot design

This site is selected as it is highly contaminated with VOCl and is located in a soil layer that is typical for Denmark in the area of CRD. At the site, there is a demand for heat/cold, but it was decided not to connect, as the duration of the pilot test is a relatively short period of time.

Further, the use of heat at this site therefore can easily be provided by the owner of the site. Although there is no heat and cold demand for the ATES system, the access to the site could be readily arranged and as this site is representative for many other locations in urban areas which have an energy demand for heat and cold. As the hydrogeology and the type of contamination are representative for the area and the access to the location was readily organized, this site had the best performance for a pilot.

In the pilot design, calculations are included upon the potential addition of electron donor to be able to create a low redox potential, as this is a prerequisite for reductive dechlorination. Further, in the design the addition of a bacterial inoculation by a DHC culture was included. This addition of an active microbial culture is developed by Bioclear earth as the ATES-plus concept and aims at to stimulate biodegradation of chlorinated solvents when there is a lack of sufficient reductive dechlorinating biomass.

The final design is described in detail in Appendix 2

Background and aim of the pilot

ATES-systems are highly effective energy-storage systems, and provide energy with low CO₂-emissions.

There is an increasing interest in ATES systems and the potential in Denmark for ATES is evaluated to be at least 400 ATES plants.

In urban and industrial areas there can be a large need for cooling and heating of buildings and industrial processes. However, in such areas, contaminated sites are often encountered, and contamination with chlorinated solvents is one of the most frequently occurring contaminations. This contamination often hampers urban development. In addition, plumes of chlorinated solvents often occur in aquifers at the same depths at which an ATES-plant would abstract and inject its waters.

In the Netherlands, where ATES-systems are widely employed, and the number of existing ATES plants is estimated to be about 2500 plumes of chlorinated solvents is found to obstruct the development of ATES in urban areas. However, a new approach is to instead view the combination of ATES and remediation as an opportunity, in that there may be synergies and benefits derived from combining the two concepts.

Objectives

The purpose with the pilot test is:

- To document that it is possible to design a reliable combi concept with ATES and ERD
 - o General concept



- o Impact on biodegradation and general performance of the ATES system of temperature, adding nutrients, bacteria etc., should become measurable
- To document that we can enhance remediation at the site, but not necessarily to perform a full remediation of the site
 - o Heated water enhances the degradation
 - o Higher flowrate enhances the removal of the contamination by increased interaction of bacteria and VOCl
- To document that we can deliver energy (heat and/or cold) but not necessarily doing it
 - o Flowrate high enough for possible delivering of energy
 - Construct and exploit a reliable system and/or figure out how to deal with clogging risks
- To proof that the contamination is not spread in the groundwater on neighbouring locations
 - o Do not worsen the risk/presence/spread

A schematic overview of the pilot is presented in Figure 1. Warm water is injected in the northern injection well (red), thereafter the groundwater is extracted from the cold well, creating a recirculation ATES system in which the groundwater is kept in a circuit. Between the warm well and cold well 4 monitoring wells are installed at several depths to monitor all parameters as presented in Table 1.

b. System installation

The system is installed in December 2017. In Figure 2 an overview of the design is presented as a top view of the location. The system is operated by injection of warm water in the warm well and extraction from the cold well with maximum flows of 5 m³/h. During the operation, the flow rate is 3 m³/h. The detailed design is described in Appendix 2.

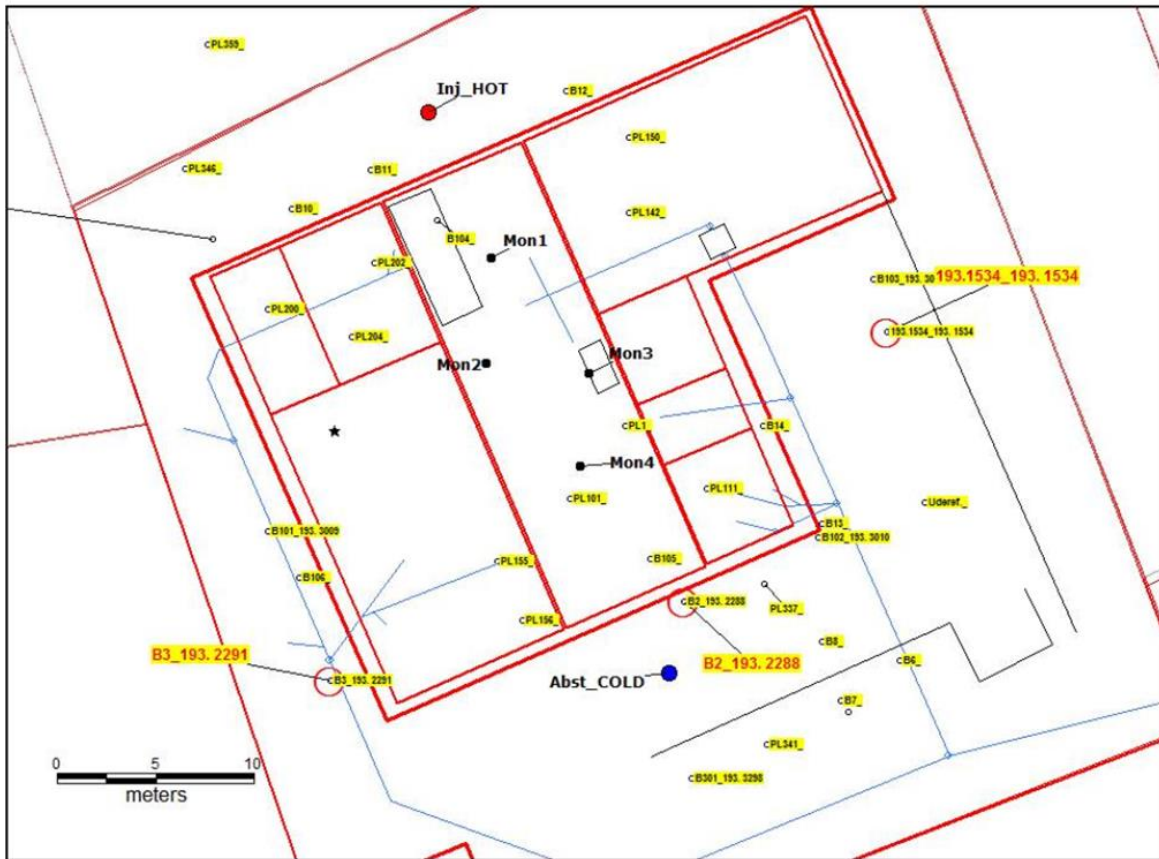


Figure 2. Top view of the pilot Hammerbakken 10. At the north site of the building the injection well with warm water (hot) is installed (red dot, at 21-27 m blg), whereas at the south side the cold well is positioned (blue dot, 21-27 m blg). Inside the building 4 monitoring wells are installed within each well filters at 21-23 and 22-27 m blg.



c. System operation with specific description of activities in 2018

In March 2018, Ramboll made an overview of activities for 2018 upon the Pilot Birkerød, as is described below.

In the beginning of 2018 the final installations of the system was conducted. The recirculation of heated groundwater started in the middle of March. The heated groundwater was recirculated for app. 2 months. In that period, two monitoring rounds were performed in April and May.

In May, the laboratory results were reported by Bioclear earth (Appendix 3).

The conclusion in the Bioclear earth report state:

Redox shift:

Redox shift occurred at laboratory scale from anoxic environment to methanogenic conditions in a few weeks of time. This indicates that there seem to be no limitations (e.g. a high iron content in the soil matrix or lack of bacteria that are needed for reduction) to make this transition. For field application, the rate of carbon source addition and spreading of the carbon source will be one of the limiting factors, combined with the calculated estimation of oxygen intrusion in the shallow groundwater layer at the Birkerød site.

Degradation test:

Based on the results of the degradation test it can be concluded that with addition of high concentrated DHC organisms (107 DHC/ml) there is a complete transfer of TCE into c-DCE within five weeks and a slight conversion of cis-DCE into VC within eight weeks after new addition of bacteria. This is without carbon source addition and under suboptimal redox conditions. This result is measured in one out of three batches. In the reference test no change of TCE occurs, indicating that the measured result in the test with added bacteria is due to biodegradation. The actual initial condition in the soil/water system by itself is inappropriate for complete dechlorination. Addition of bacteria one way or the other is needed, since the soil and groundwater does not contain any dechlorinating biomass by itself. Based on the present data it seems to be most appropriate to start with a first reduction of redox status in the field. The redox test showed this is possible quite effectively, but precipitates may be formed in the soil matrix. If the redox status is lowered into iron/sulphate reducing condition the addition of high concentrated biomass will be much more effective.



Further research was performed by Deltares and Ramboll.

The sampling of the biomass by Deltares showed that DHC bacteria were only present at non-significant numbers in January at the start of the Pilot as expected, as the redox potential was high at the start of the pilot.

Table 1. Laboratory data on genecopies in wells Birkerød sampled in January 2018 analysed by Deltares

code	Sample				gene copies / ml groundwater				
	well	depth	T	date	Total Bacteria	DHC	vcrA	bvcA	etnE
1	mon 1	22m	T=0	18-1-2018	7,52E+05	<4	<2	<3	6,37E+01
2	mon 1	26m	T=0	18-1-2018	1,84E+06	<4	<2	<3	2,03E+02
3	mon 2	22m	T=0	18-1-2018	2,20E+05	7,63E+01	4,06E+01	<3	7,00E+01
4	mon 2	26m	T=0	18-1-2018	1,03E+06	<4	<2	<3	1,97E+02
5	mon 3	22m	T=0	18-1-2018	1,31E+06	<4	<2	<3	1,55E+02
6	mon 3	26m	T=0	18-1-2018	8,03E+05	<4	<2	<3	8,13E+01
7	mon 4	22m	T=0	18-1-2018	1,90E+06	<4	<2	<3	7,12E+01
8	mon 4	26m	T=0	18-1-2018	6,03E+05	<4	5,96E+01	<3	4,95E+01
9	HOT	24m	T=0	18-1-2018	5,63E+05	<4	<2	<3	1,88E+02
10	COLD	24m	T=0	18-1-2018	1,69E+05	4,77E+01	<2	<3	1,46E+02

Ramboll monitored the ATES system in the monitoring wells 1, 2, 3 and 4 as well as in the warm well en cold well starting in January upon pH, conductivity, ammonium, NO_2^- , NO_3^- , SO_4^{2-} , H_2CO_3 , S^{2-} , non-volatile organic compounds (NVOC), iron(II, III, total), methane, ethane, ethene, chloroform, 1,1,1 trichloroethane, CCl_4 , TCE, PCE 1,2 dichloroethane, chloroethane, 1,1 dichloroethene, trans 1,2 dichloroethene, cis-1,2, dichloroethene, 1,1 dichloroethane, VC. The first raw monitoring data were send around in May 2018 and are included in the monitoring results until December 2018 by Ramboll (Appendix 4).

The results of the monitoring are discussed in chapter 3.

The activities for the pilot project in 2018 are shown in the time schedule below:



Table 2 Final time schedule 2018

Activity 2018	March	April	May	June	July	August	September
Recirculation of heated groundwater only							
Addition of carbon source							
Recirculation of heated groundwater and bacteria							
Manual supervision							
Monitoring						31	37
Injection of bacteria							
Meeting							
Report draft							

Activity 2018/2019	October	November	December	January	February	March	April
Recirculation of heated groundwater only							
Addition of carbon source		43					
Recirculation of heated groundwater and bacteria							
Manual supervision							
Monitoring		43	45	50	4	10	
Injection of bacteria		44					
Meeting							
Report draft							

The original time schedule is attached in Appendix 2 in a bigger format.

d. Monitoring with specific description of activities in 2018

In January 2018 the ATES system at Hammerbakken 10 was started.

Samples were taken from the ATES system by Rambøll and sent to Bioclear earth for pre-investigation tests.

The following tests have been executed:

- Test 1: An anaerobic redox shift test. Aim is to determine whether and how fast reduction of the soil/groundwater can be accomplished by adding carbon source. By this test we can find out if this is easy and feasible in an efficient way (if needed for the process).
- Test 2: A degradation test, consisting of one test with addition of dechlorinating bacteria and one test under same conditions without dechlorinating bacteria (reference). Aim is to determine whether addition of an abundance of dechlorinating bacteria (*Dehalococcoides* spp. or DHC) will lead to degradation of the trichloroethylene present at the Birkerød site.

3. Results

The monitoring results are described in detail in Appendix 4 and will be described below for the most relevant results.

a. Results test 1: anaerobic redox shift test

First results show that mixing by the ATEs systems performs as expected and therefore the heat distribution performs according to the design (Figure 5).

However, for the biodegradation within the pilot the concentrations of oxygen and therefore the redox condition as such, were too high to stimulate the bioremediation of the chlorinated solvents.

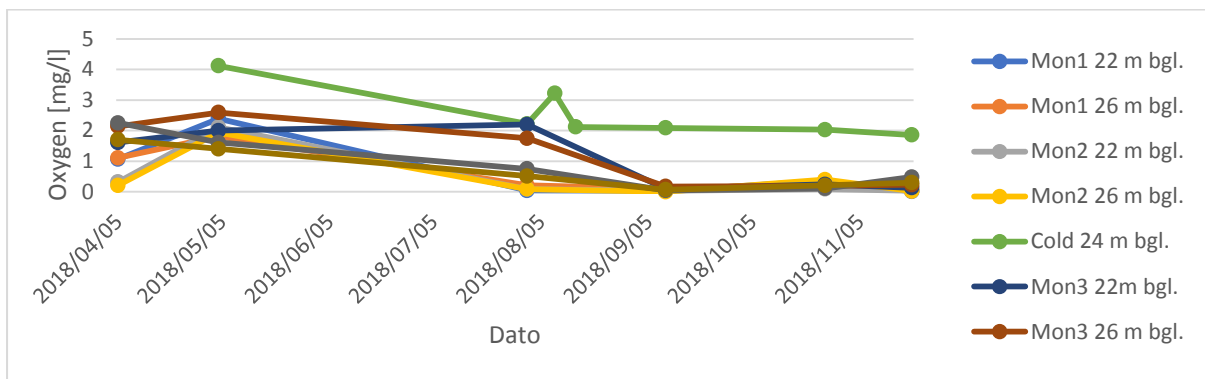


Figure 3. Oxygen concentration of groundwater in monitoring wells and cold well.

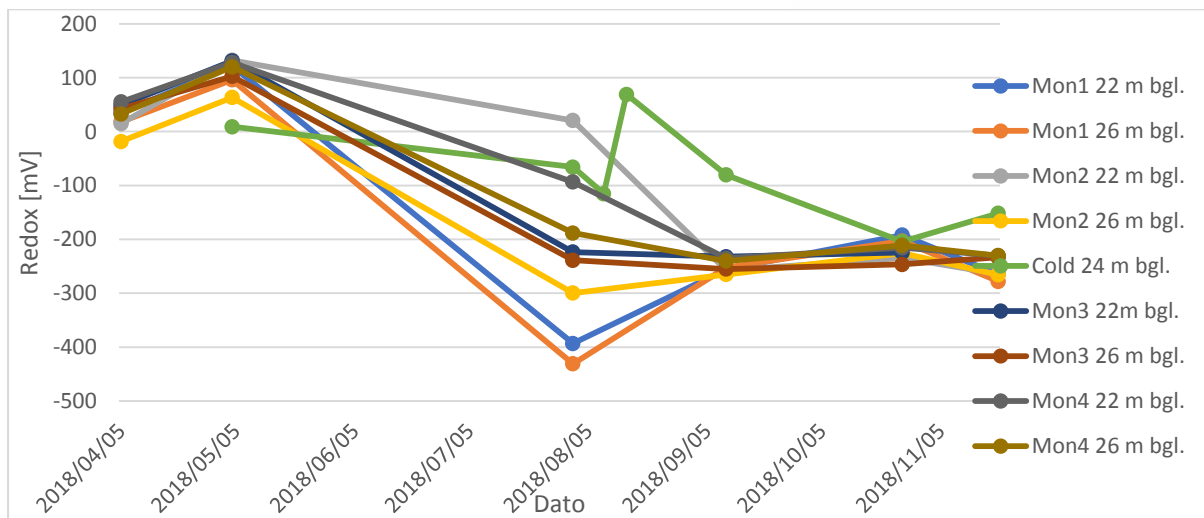


Figure 4. Redox potential (mV) in groundwater of monitoring wells and cold well with time.

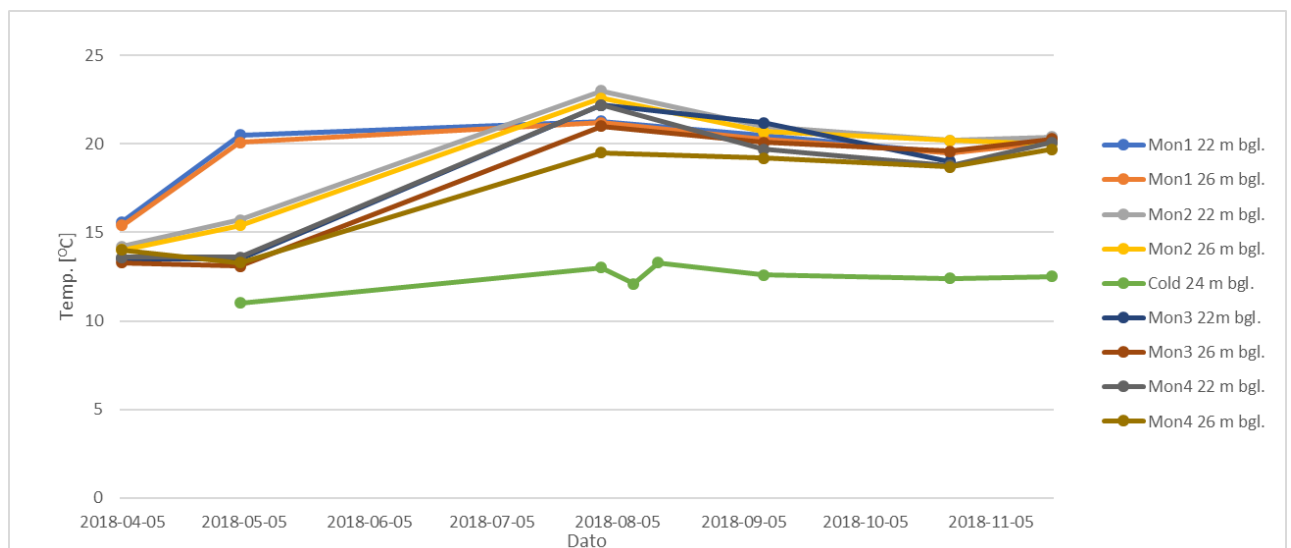


Figure 5. Temperature in monitoring wells and cold well with time.

In summary, the first monitoring results showed that the natural redox at the site was too high for microbial reductive dechlorination. The laboratory tests that were performed in these months showed that the redox of the original soil was too high to allow reductive dechlorination. However, by adding electron donor either addition of biomass in laboratory tests showed a redox shift could be realized. After such lowering of the redox, the reductive dechlorination should be possible.

In e-mail exchanges and a Skype meeting by CRD, Rambøll, and Bioclear earth it was decided to inject bacteria and carbon source via the injection well (Appendix 5). First injections were performed by injection of electron donor in 6 injections from June 25 until July 13 in the warm well.



In the monitoring round of August 5 2018 the oxygen concentration was lowered below 1 mg O₂/l in most monitoring wells, however in monitoring well 3 and in the cold well the oxygen concentrations were still above 1.5 mg O₂ mg/l (Figure 3). Fortunately the redox values, that also refer to the degree of oxygenation of the aquifer shows for all wells a lowering of the redox potential after May 5, showing that addition of electron donor had a reducing effect (Figure 4).

In the monitoring round of September 5 2018, a further reduction and lowering of oxygen concentration were observed. This resulted in oxygen concentrations below detection level in figure 3, except for the cold well. Further, the redox potential dropped for all wells to -250 mV.

The different behaviour of the cold well can be explained, as this well is the latest well that will be affected by the changes in the injection in the warm well (and extraction of infiltrated water, downstream of the injection in the warm well). Therefore around the warm well conditions are more optimal for reductive dechlorination than around the cold well.

Monitoring on October 5 2018 showed a stabilization of both oxygen concentrations and redox potential. Even for the cold well a redox potential of -250 mV was reached, unless the presence of 2 mg O₂/l.

Conclusion test 1:

Within a period of 3 months, the redox could be lowered from values above +150 mV to -250mV by addition of electron donor to create conditions fit for reductive dechlorination in the field.

b. Results Test 2: Biodegradation test

b1 Contaminants concentration development

In theory, after the realization of a low redox potential the reductive dechlorination can be performed. As the low redox was only reached on September 5 2018, the biodegradation in principle can start when enough specific biomass is present that is able to perform reductive dechlorination. Further, in theory injection of specific bacteria as *Dehalocoocoides* (DHC) may lead to the complete conversion of chlorinated solvents and their intermediates like DCE and vinylchloride (VC) to the harmless final non-chlorinated product ethene.

In this report, the results related to reductive dechlorination of specific compounds and dechlorination degree is available until November 20 2018. Thereafter monitoring will be continued for CRD until March 2019.

After installation of the ATES system in December 2017, pumping and monitoring was started in January 2018. During operation it was decided to add electron donor in June 2018 and thereafter it was decided to inject DHC culture (6 m³) was injected to stimulate the biodegradation of the chlorinated solvents. As the TCE concentration was highest of the present compounds PCE (four chlorine atoms) and TCE (three chlorine atoms), the performance of the dechlorination is expressed by addressing TCE as parent compound in calculation of the dechlorination percentage.

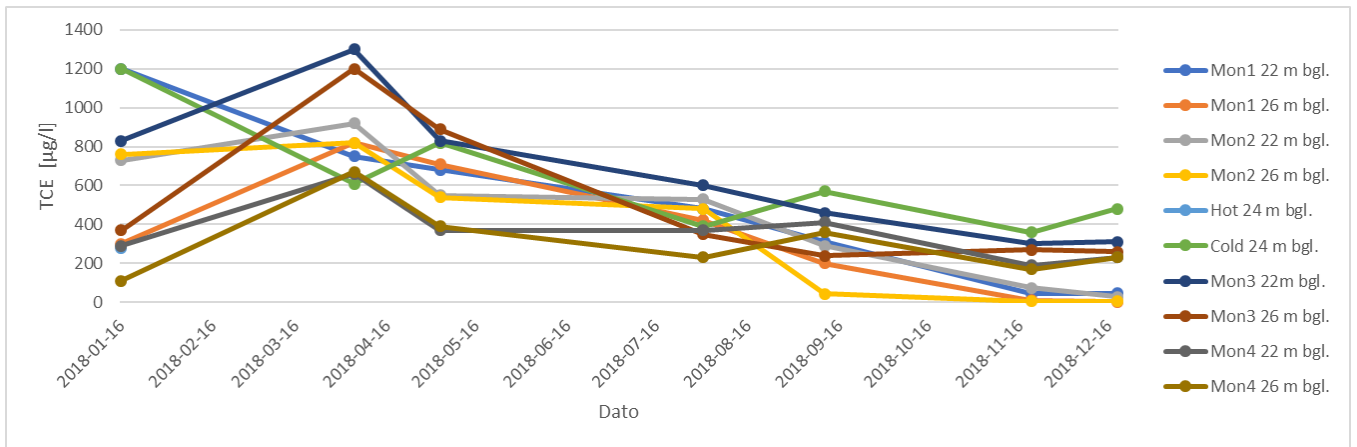


Figure 6. Trichloroethylene (TCE), parent compound in dechlorination degree, concentration with time

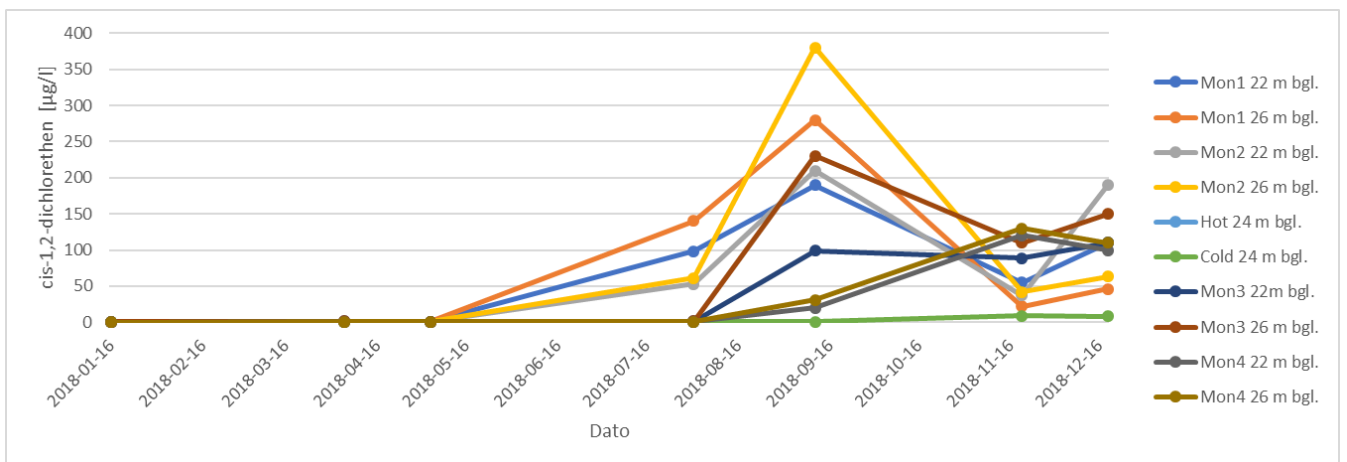


Figure 7. Cis 1,2 dichloroethene, intermediate product in dechlorination degree, concentration with time

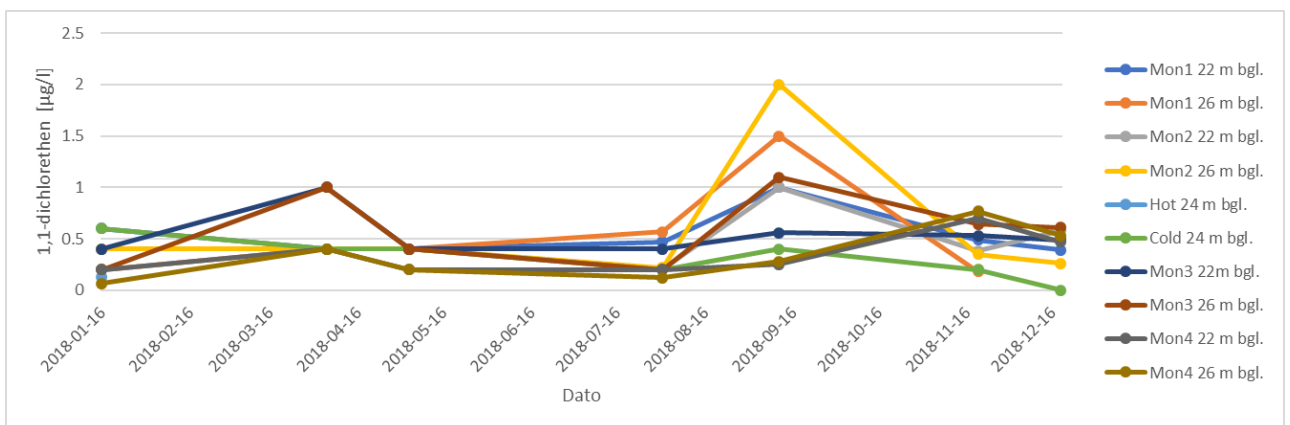


Figure 8. 1,1 dichloroethene, intermediate product in dechlorination degree, concentration with time

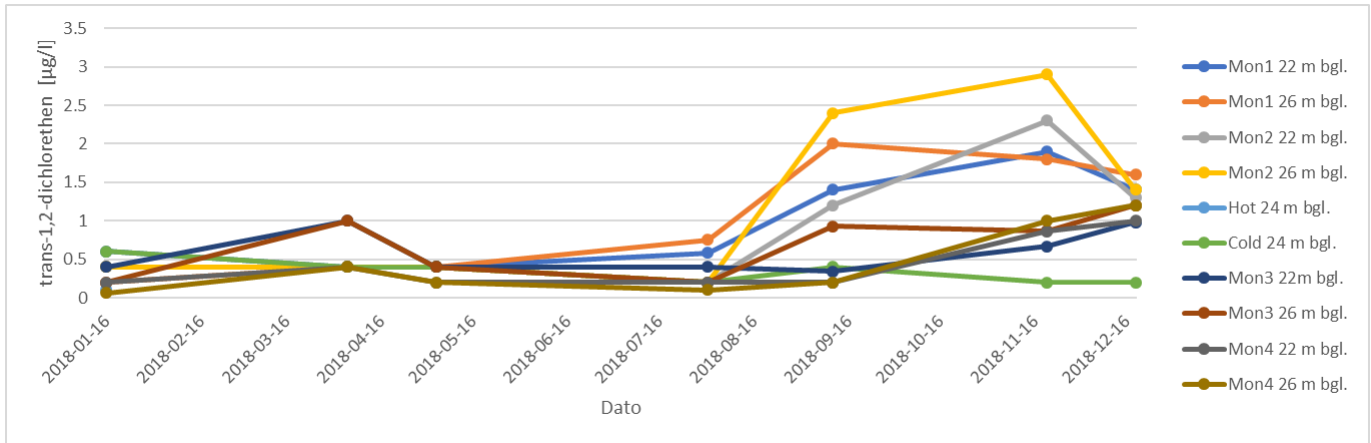


Figure 9. Trans 1,2 dichloroethene, intermediate product in dechlorination degree, concentration with time

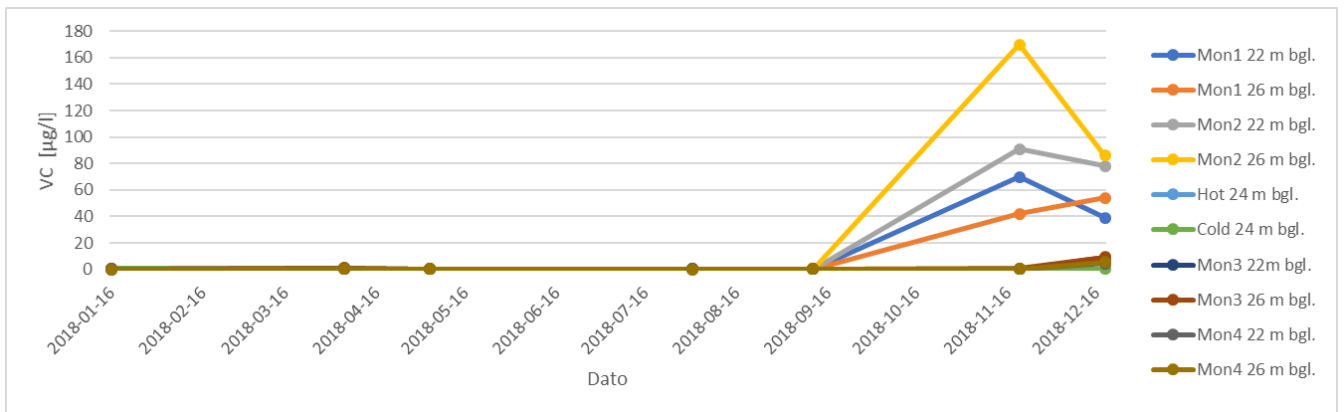


Figure 10. Vinylchloride, intermediate product in dechlorination degree, concentration with time

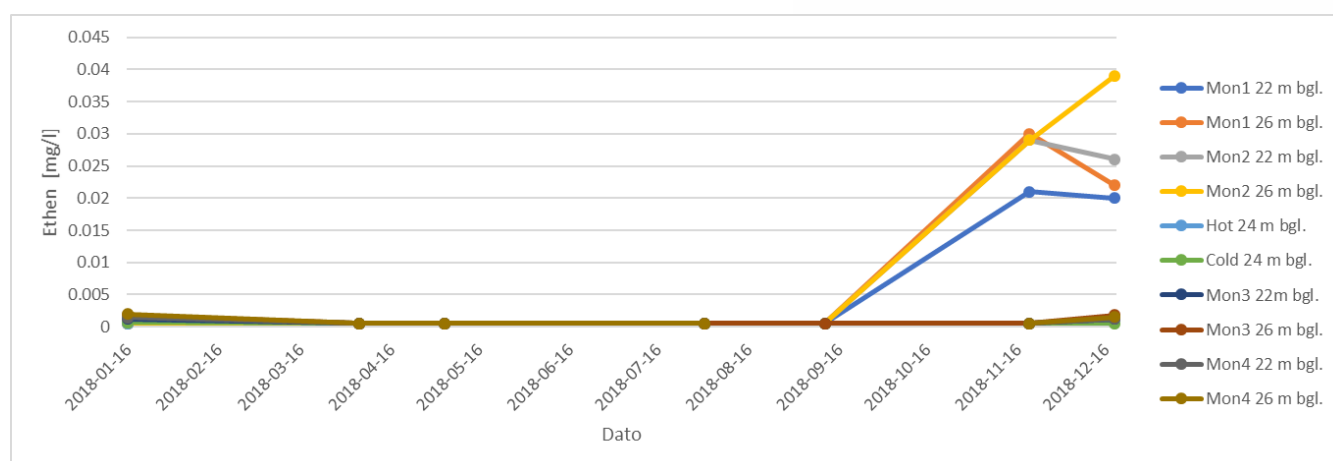


Figure 11. Ethene, end product in dechlorination (no chlorine atoms), concentration with time

In Figure 6 it can be observed that the parent compound TCE slowly decreases in concentration over time until June, when electron donor was added. This is in line with the observation in Figure 3 and 4, as in these figures the oxygen concentration as well as the redox potential showed a decrease after addition of the electron donor. Therefore, it can be concluded that addition of electron donor stimulated the biodegradation of TCE.

Further, after addition of electron donor especially the intermediate DCE (2 chlorine atoms) is increased (Figure 7, 8 and 9). However, no significant increase is observed after June 2018 in the intermediate VC (one chlorine atom) and the final product ethene (all chlorine atoms removed) (Figure 10 and 11).

In September and October, the results above were discussed within the consortium and it was decided to add 6m³ of a DHC culture, which was grown in a special bioreactor by Bioclear earth. This bacterial culture is well known for its full dechlorination potential, meaning that PCE and TCE can be fully degraded to the final harmless product ethene.

Just before addition of DHC bacteria, more electron donor was added, as the concentration of electron donor was decreasing. In the first week of November the DHC culture was injected via monitoring well 1 and monitoring well 2. In the last data-points of Figures 6 to 11 represent the monitoring data of November 20 and December 19, after adding the DHC culture.

The monitoring results of November 20 show very promising and consistent results, as TCE (Figure 6) shows especially a sharp decrease in monitoring well 1 and 2. Also, a lowering in TCE was observed in the other monitoring wells, whereas the TCE degradation in the cold showed to be less reactive.

DCE showed a strong increase after the addition of electron donor in August 2018 for all three isomers. The increase of the DCE intermediate corresponds with the absence of formation of VC and Ethene in August 2018 in Figure 10 and 11.

From the figures 6 to 11, it can already be concluded that some biodegradation is ongoing after addition of electron donor in June 2018, which especially lead to the formation of all variants of DCE. Further, it seems at that time that DCE is not further degraded to VC and ethene.

The effect of the addition of the DHC culture was clearly visible for the sharp increases in VC (Figure 10) and for the final non-chlorinated product ethene (Figure 11) on 16 November 2018, within two weeks after addition of the DHC culture. The sharp increase of these compounds can be seen as a direct effect of the addition of the DHC culture.

In November 2018 significant changes in the figures 6 to 11 were observed after the addition of the DHC culture, showing a complete conversion of chlorinated solvents to the harmless product ethene could occur.

The biodegradation can be visualized in a more specific form by the determination of the dechlorination degree.

In Figure 12, the dechlorination degree is presented, by selecting TCE as the parent compound by the calculation in each specific well. The formula applied for the dechlorination degree is:

% *dechlorination* =

$$\frac{(1,1 \text{ DCE} + \text{trans } 1,2 \text{ DCE} + \text{cis } 1,2 \text{ DCE} + 2 * \text{VC} + 3 * \text{Ethene})}{(\text{TCE} + 1,1 \text{ DCE} + \text{trans } 1,2 \text{ DCE} + \text{cis } 1,2 \text{ DCE} + \text{VD} + \text{Ethene})} * 100 \%$$

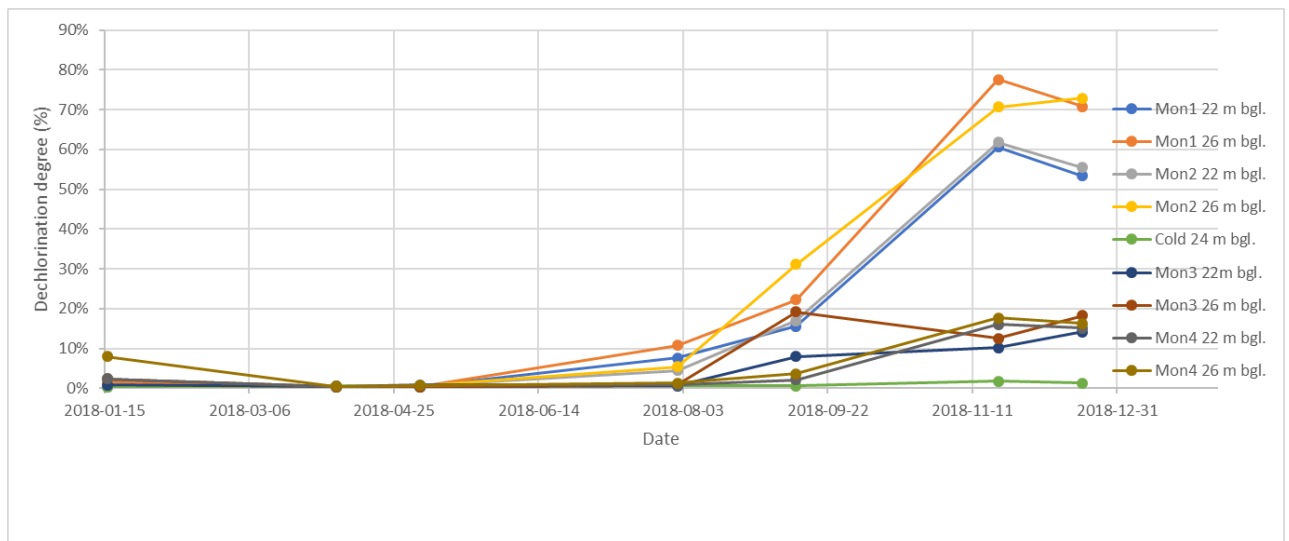


Figure 12. Percentage dechlorination of TCE with time



From Figure 12, it can be observed that reductive dechlorination started after addition of electron donor in July 2018. After the addition of the DHC culture in the first week of November, a sharp increase of the dechlorination degree was observed especially in monitoring wells 1 and 2 at both 22 and 26 mbgl and in the warm well.

The biodegradation rate in the monitoring wells 1 and 2 and in the warm well are remarkably high, especially compared to a large variety of in situ bioremediation projects that are most often performed by natural biological processes and show much lower biodegradation rates, resulting in degradation times of years either decades.

The monitoring will be continued until March 2019. Then it will become clear, whether the biodegradation in the monitoring wells 3 and 4 and the cold well can also be stimulated in the ATES and bioremediation system.

B2 Microbial molecular analysis results


Groundwater

After the addition of the 6 m³ DHC culture in the first week of November, Deltares performed a DNA analysis of several groundwater samples in the 4 monitoring wells at various depths as well as in the cold well on November 21 and December 20 2018 (Appendix 6). By DNA analysis gene copy numbers were obtained for total bacteria, DHC bacteria as well as for enzymes involved in biodegradation of VC and formation of ethene (Table 2).

Results show that in the wells mon 1 and mon 2, in which the DHC culture was added, significant higher amounts of total bacteria, DHC and enzymes were found in the groundwater in these wells compared to the other wells (mon3, mon 4 and cold well). This distribution of DNA in groundwater was present on November 21 as well as December 20.

Table 2. Molecular analysis of groundwater in wells at Hammerbakken site

Sample					gene copies / ml groundwater				
code	well	depth	T	date	Total Bacteria	DHC	vcrA	bvcA	etnE
1	mon 1	22m	T=2	21-11-2018	2,62E+07	3,68E+03	5,04E+04	1,17E+04	2,27E+02
2	mon 2	22m	T=2	21-11-2018	6,58E+07	3,17E+04	2,18E+05	3,26E+04	4,72E+02
3	mon 3	22m	T=2	21-11-2018	4,91E+02	<4	1,24E+01	<3	<13
4	mon 4	22m	T=2	21-11-2018	6,10E+02	<4	1,43E+01	<3	<13
5	mon 1	26m	T=2	21-11-2018	2,57E+03	<4	3,30E+01	2,10E+01	<13
6	mon 2	26m	T=2	21-11-2018	1,71E+08	2,50E+05	7,27E+05	1,24E+05	6,75E+02
7	mon 3	26m	T=2	21-11-2018	5,61E+02	<4	5,49E+01	<3	<13
8	mon 4	26m	T=2	21-11-2018	4,49E+02	<4	8,96E+01	<3	<13
9	cold		T=2	21-11-2018	1,38E+05	<4	<6	<3	<13
10	mon 1	22m	T=3	20-12-2018	2,78E+07	1,38E+03	4,32E+04	6,84E+03	3,30E+02
11	mon 2	22m	T=3	20-12-2018	1,76E+07	6,09E+02	2,77E+04	4,64E+03	7,79E+01
12	mon 3	22m	T=3	20-12-2018	4,03E+06	<4	2,26E+01	<3	6,35E+01
13	mon 4	22m	T=3	20-12-2018	4,97E+06	<4	6,14E+01	<3	3,48E+01
14	mon 1	26m	T=3	20-12-2018	8,57E+07	2,79E+04	3,23E+05	3,19E+04	4,73E+02
15	mon 2	26m	T=3	20-12-2018	5,25E+07	2,51E+03	9,10E+04	9,67E+03	6,88E+02
16	mon 3	26m	T=3	20-12-2018	9,16E+06	<4	2,46E+02	4,65E+01	1,02E+02
17	mon 4	26m	T=3	20-12-2018	8,80E+06	<4	1,00E+02	<3	3,95E+01
18	cold		T=3	20-12-2018	1,46E+07	<4	1,58E+02	<3	6,11E+02

 The submitted sample material was very cloudy and dark colored, this may affect the results



Mesocosms

Just after the inoculation with DHC, mesocosms (tubes with a solid matrix in which bacteria can adsorb) were placed in several monitoring wells. These mesocosms were harvested on December 19 2018 and analysed for DHC (cells/gram) and the enzyme *vcrA* (copy number/g) (Appendix 7).

Table 3. Molecular analysis of mesocosms at Hammerbakken site

Sample	Well	Sample specific detection limit	Dehalococcoides spp.	Vinyl chloride reductase (<i>vcrA</i>)
			Cells /gram	DNA-copies /gram
1	mon 1 22/7	5.6×10^3	7.5×10^7	5.8×10^6
2	mon 1 26/3	3.4×10^3	1.8×10^8	1.2×10^7
3	mon 2 22/32	3.0×10^3	2.8×10^6	2.0×10^5
4	mon 2 26/29	5.0×10^3	6.6×10^6	4.2×10^5
5	mon 4 22/22	7.6×10^3	5.9×10^4	n.d.
6	mon 4 26/19	2.2×10^3	4.5×10^4	4.7×10^3
The range of the analysis results lies between $0.5 \cdot N$ and $2 \cdot N$ (N=number of detected cells or DNA-copies).				

Results from Table 3 show that the mesocosms in monitoring well 1 have 1000 to 10.000 more DHC spp. compared to the mesocosms in monitoring well 4. In Monitoring well 2 the mesocosms have about 100 more DHC spp. compared to monitoring well 4.

For the enzyme *vcrA*, which is involved in the conversion of VC, also 1000 to 10000 more copies were present in mon 1 and 100 more in mon 2 compared to mon 4.

These results show that inoculation with DHC lead to a significant increase of DHC spp. and *vcrA* compared to the non-inoculated wells.

Conclusion test 2:

Within 2 months after addition of electron donor and reaching a redox potential of -250mV the biodegradation of chlorinated solvents started resulting in a dechlorination percentage of up to 30%. The effect of the increased temperature cannot be evaluated yet, as it seems that the electron donor and the DHC culture did not reach the cold well yet.

The biodegradation could be further stimulated by addition of the DHC culture with specific bacteria that can fully degrade the chlorinated solvents to the harmless product ethene.

Within the monitoring wells 1 and 2 the dechlorination degree was increased from about 30% until about 71-78% within 2 weeks, which is remarkably fast.



Microbial molecular analyses confirm the increase of specific dehalogenating bacteria after inoculation with 6 m³ DHC culture within 2 and 6 weeks after inoculation.

The upcoming monitoring rounds until March 2019 will show whether such high dechlorination degree could also be reached with time in the other monitoring wells as well as in the cold well.

Future microbial molecular analysis will show whether the distribution of DHC and vcrA enzymes will stay stable in the inoculated monitoring wells either the spreading of these parameters in the whole ATEs system.



4. Evaluation of system performance

Until the end of 2018, no clogging problems were observed in the ATES pilot and it seems that the performance of the ATES system was according to the design. However, analysis on the working of the ATES system over a longer period of time will increase the long term reliability of the design of this ATES system with biostimulation.

The performance of the biodegradation within the ATES systems needs a follow up monitoring to be able to evaluate the stimulation by addition of specific biomass in the pilot. The Capital Region of Denmark is aiming at to continue the monitoring in any case until March 2019.



5. Conclusions related to barriers and issues addressed

Conclusions regarding the 3 barriers addressed:

1. The barrier upon the lack of experience:
The cooperation between the Danish and Dutch teams showed that knowledge could be transferred to the Danish team. Future developments will show whether this knowledge transfer will further stimulate the application of ATES in Denmark.
2. Acceptation of ATES by drinking water companies
This pilot showed the proof of concept in which at field scale the concentrations of chlorinated solvents could be lowered by the implementation of a combined ATES & Bioremediation approach. This will be the start for discussions with water companies to convince them that the combination of ATES & Bioremediation is a solution rather than a threat to drinking water preparation from contaminated groundwater.
3. Adaptations of legislation
For this specific case no adaptations of legislation was necessary as the CRD is allowed to perform pilot studies. However for the full application in other contaminated locations a discussion needs to be started, not only in the region of the Capital of Denmark, but also in other regions to convince legislators that ATES & Bioremediation can stimulate the transfer to sustainable energy together with improvement of environmental quality of groundwater.

6. References

Ni Z (2015): Bioremediation of chlorinated ethenes in aquifer thermal energy storage PhD Thesis, Wageningen University

Pellegrini M., N. Hoekstra, M. Bloemendal, A. Andreu Gallego, J. Rodriguez Comins, T. Grotenhuis, M. Mazzoni, S. Picone, A.J. Murrell, H.J. Steeman (2018): Novel developments in aquifer thermal energy storage through combinations with other sustainable technologies, submitted.



Appendix 1

Literature study

AQUIFER THERMAL ENERGY STORAGE (ATES)

AND

ENHANCED REDUCTIVE DECHLORINATION (ERD)

Available on request



Appendix 2

Detailed Design

Hammerbakken 10, Birkerød

Available on request



Appendix 3

Pre-investigation tests for pilot ATES+ERD for Capital Region of Denmark (Bioclear earth)

Available on request



Appendix 4

Monitoring results

Hammerbakken 10

Birkerød

in 2018

January – December

Available on request



Appendix 5

Proposal Supplementary Activities June 2018

Available on request



Appendix 6

Microbial molecular analysis groundwater

Available on request



Appendix 7

Microbial molecular analysis mesocosms

Available on request