

NanoRem Bulletin

CL:AIRE's NanoRem bulletins describe practical aspects of research which have direct application to the characterisation, monitoring or remediation of contaminated soil or groundwater using nanoparticles. This bulletin describes the development and application of a range of analytical methods for *in situ* measurement and detection of nanoparticles in remediation.

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Development and Application of Analytical Methods for Monitoring Nanoparticles in Remediation

1. BACKGROUND

NanoRem (Taking Nanotechnological Remediation Processes from Lab Scale to End User Applications for the Restoration of a Clean Environment) was a research project, funded through the European Union Seventh Framework Programme. The NanoRem project focused on facilitating practical, safe, economic and exploitable nanotechnology for *in situ* remediation. This was undertaken in parallel with developing a comprehensive understanding of the environmental risk-benefit, market demand, overall sustainability, and stakeholder perceptions of the use of nanoparticles (NPs). The NanoRem Toolbox, available at www.nanorem.eu, provides outputs which address all these issues.

All remediation applications need to document the effectiveness of the technologies employed. For NP-based remediation, this includes providing information on the mobility and fate of the deployed NPs. Monitoring the behaviour of engineered NPs requires their detection in environmental media, and in particular their isolation from natural background colloidal material. This represents a potential challenge for the use of Fe-based NPs in remediation, because of relatively high levels of naturally occurring Fe. Hence, the development and application of analytical methods for *in situ* measurement and detection of NPs was a key objective of the NanoRem project.

This bulletin describes how this objective was met and includes measurements that actually monitor particles within the aquifer, and methods that combine *in situ* or at site sampling with subsequent at site or laboratory measurement. Within NanoRem, the method development work was organised around the following three main areas:

i. Development and optimisation of monitoring and tracing tools. Techniques based on the measurement of the ferro-magnetic properties (susceptibility) of Fe for monitoring Fe NPs in the field were optimised and new methods were developed for detection of Carbo-Iron® and Fe-zeolites. In addition, the feasibility and applicability of isotope and trace metal (rare earth element - REE) techniques for laboratory and field detection of Fe-based NPs were determined.



Figure 1. Joint sampling for monitoring method tests at the VEGAS facility, University of Stuttgart (Source: Deborah Oughton).

ii. Laboratory and field tests of the methods. A series of tests were conducted on a number of different techniques for field monitoring, including routine methods of NP characterisation as well as the magnetic susceptibility and REE methods developed specifically for Fe-based NPs during NanoRem. Work included evaluation of the methods' applicability for Fe-based NPs and *in situ* application, assessment of detection limits and potential for routine application. The results have been consolidated into the NanoRem monitoring toolbox.

iii. Development of protocols. Protocols were produced for on-site measurements and *in situ* characterisation of natural and engineered NPs. These include application of modern high performance analytics for samples collected in the field and analysed in the laboratory.



Taking **Nanotechnological Remediation** Processes from Lab Scale to End User Applications for the Restoration of a Clean Environment. This project has received funding from the European Union's Seventh Framework Programme for research, technological development and demonstration under grant agreement no. 309517.



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2. FROM LABORATORY TO FIELD

The ultimate aim of method development for remediation monitoring is to provide techniques that can be applied *in situ* (i.e. in the field). A good deal of experience is available from application in laboratory studies, and particularly those to study the mobility and fate of NP in various test media. However, laboratory experiments tend to use rather high concentrations of NP, in simple media, and can rely on relatively straightforward methods for NP measurement and characterisation. Measurement during field applications is more challenging, primarily due to more complex and heterogeneous media.

One of the strengths of the NanoRem project has been the opportunity to test a wide variety of methods, from measurement of simple chemical parameters to high-end sophisticated techniques, and to cover applications in simple laboratory experiments, large-scale tank experiments, and finally field applications. The tested field applications have focused on the injection of Fe-based NPs for groundwater remediation (see NanoRem Bulletins 7-11). This has enabled an evaluation of applicability of different methods for Fe-based NPs, as well as providing insight into specific challenges, advantages and factors influencing detection limits for field measurements.

3. MONITORING AND CHARACTERISATION REQUIREMENTS AT DIFFERENT REMEDIATION PHASES

The applicability of the various methods depends on the phase of remediation and the question to be addressed, since the different phases have different analytical requirements and issues. The most important issues for monitoring purposes are:

- i. Field characterisation studies prior to NP remediation.
- ii. Monitoring the movement and distribution of NPs during injection. The main question at this point is whether the NP suspension reaches the required location, at the required concentration and state. During this phase the NP concentrations are relatively high, which makes detection more straightforward, but there is a need for rapid feedback at relatively high resolution.
- iii. Monitoring for transport of "fine" or "renegade" NPs out of the core application area during and after injection. Low NP concentrations give rise to challenges with detection against background levels of colloids, but monitoring can be carried out with a lower spatial resolution, and less urgency for rapid feedback.
- iv. Post-injection behaviour and information on the transformation and reactivity of the NPs. This is particularly relevant for the assessment of the need for reinjection.

The different phases are categorised as follows:

3.1 Pre-Injection Phase

The measurement techniques applied in this phase are mostly standard methods used in chemical or hydraulic engineering. Measurement systems such as sampling and injection wells, or *in situ* sensors (if needed) have to be installed, and disturbances of the system need to be accounted for, depending on the groundwater velocity and the time expected for stabilising of the system. The main task is to describe the temporal and spatial concentration profiles of contaminants, but sampling can also provide background and baseline data of relevance for NP tracking.

3.2 NP Injection Phase

The main focus during the injection phase is the behaviour of the NPs, namely the radius of influence (ROI), the travel distance and the homogeneity of the distribution around an injection point or well. Acute changes within minutes have to be detected. This phase has a duration of hours to days, and requires a high measurement frequency. Since NPs are injected as a suspension, the liquid and the solid phases may behave differently, and methods need to address both phases in order to provide information about the overall efficiency of the injection and potential deviations from the planned behaviour. For the liquid phase relatively simple methods are available such as temperature measurements (usually the temperature of the injected fluid differs from the groundwater temperature) or the addition of tracer substances (dyes or tracer ions) to the suspension. For the NPs, unfortunately, only a few truly *in situ* methods are available, and in most cases the NP detection will have to be based on sampling and on-site analysis.

3.3 System Recovering Phase

This phase is a relatively short, intermediate phase between injection and a return to the natural groundwater flow. The volume of injected fluid will cause considerable disturbance of the hydraulics in the aquifer and intensive analytical activities for both NPs and contaminants are usually not required during this phase. Thus a reduced monitoring programme is advisable where only some main chemical parameters are monitored in order to follow the overall changes and determine when natural groundwater flow conditions have re-established, taking into account the modifications to be expected by the injected NPs. Depending on the expected rate of groundwater movement, monitoring for the potential transport of NPs outside of the treatment area could be started.

3.4 Long-Term Steady-State Phase

An extensive monitoring programme should be undertaken in the long-term phase in order to verify the success of the remediation. In addition to monitoring of NPs, the programme should include analysis of the contaminants, reaction products, metabolites and general environmental parameters of the groundwater (e.g. pH, oxidation-reduction potential (ORP)). The main focus is to ensure the efficiency of the desired reaction and the point in time when the activity of the NP ceases. The criteria for the decision about a success of an application of NPs have to be fixed beforehand and a monitoring programme chosen accordingly. All monitoring results should be compared to the status defined during the pre-injection phase. The long-term phase should also include monitoring for potential transport of NPs out of the site. The programme should be designed so that decisions can be made about the need for reinjection, if a single injection does not reach the remediation goals.

3.5 Methods Tested

NanoRem has evaluated and developed a range of methods, covering application of general laboratory methods for characterisation, "truly" *in situ* measurements that actually monitor NPs within the aquifer, and methods that combine *in situ* or at site sampling with subsequent at site or laboratory measurement (Table 1). The results presented in this bulletin are focused predominantly on those *in situ* and at site methods that are applicable for monitoring of Fe NPs injected into groundwater during NanoRem field studies.

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Table 1: Overview of NP monitoring methods tested in NanoRem.

Type of Method	Examples	Applications	Comments
Laboratory particle characterisation	Field flow fractionation (FFF), Inductively coupled plasma-mass spectrometry (ICP-MS), transmission electron microscopy (TEM), dynamic light scattering (DLS), synchrotron techniques, isotope tracing techniques	Many particle characteristics: size, structure, composition, aggregation, mineralogy.	Required to understand fundamental particle behaviour in laboratory and field experiments
<i>In situ</i> measurement and characterisation	Ferro-magnetic methods; redox measurement; H ₂ production	Particle concentration, particle reactivity	High data resolution over time and space is possible
On site applications: sampling combined with on site or laboratory measurement techniques	Turbidity, Fe spectrometry, ultrafiltration; stable isotope and REE ratios; Mössbauer, Temperature programmed oxidation (TPO)	Particle size and concentration, Fe concentration	Turbidity, spectrometry and ultrafiltration can be carried out on site. Mossbauer, TPO, Isotope and REE ratios are laboratory measurements that can provide more detailed information on field behaviour, and/or particle reactivity

4. WHICH METHODS SHOW THE GREATEST POTENTIAL?

The various methods developed and tested in NanoRem are complementary and depend on the remediation phase, the NP utilised and the question to be asked. Since the monitoring measurements made in the pre-injection phase are relatively straightforward, this bulletin focuses on the injection and post-injection phases. Of course, baseline measurements for NP monitoring will have to be made prior to injection, but will be guided by the method chosen for post-injection monitoring.

4.1 Monitoring of NP Dispersion During Injection Phase

Results from NanoRem field measurements during the injection of nanoscale zero-valent iron (nZVI) (NANOFEAR 25S, NANOFEAR STAR), Nano-Goethite and milled Fe (FerMEG12) show that the detection of NP suspension loads is relatively straightforward, and can be easily carried out at the site. The methods include a combination of on-site sampling and analysis of suspensions (turbidity, conductivity, redox, temperature and Fe content), or *in situ* methods such as magnetic susceptibility and H₂ measurements. The detection limits, from sub mg/L for total Fe to sub g/L for magnetic susceptibility, are sufficient to follow the dispersion of injection liquids and NPs during injection. Given the relatively low toxicity of Fe-based NPs to organisms, with very few effects seen below 100 mg/L (see NanoRem Newsletter #2), these detection limits should be sufficient to assess potential ecological impacts, both within and outside the injection area. Of the various methods tested, magnetic susceptibility, turbidity and total Fe measurements are most appropriate for monitoring during injection.

Magnetic Susceptibility

Magnetic susceptibility is one of the very few *in situ* methods that can be used to detect Fe NPs, and has the advantage of allowing for continuous monitoring. It can be combined with other sampling and monitoring arrays. The sensor arrays developed by the University of Stuttgart can be installed in the subsurface and use the magnetic properties of Fe to detect changes in magnetic properties in the vicinity of the susceptibility probe. The probe itself consists of two intertwined inductors, wherein an alternating electromagnetic field produced through the outer (primary) inductor induces a voltage in the inner (secondary) inductor that is proportional to the magnetic susceptibility of the environment around the probe (Figure 2).

The method was initially developed for measurement of nZVI NPs, and at concentrations of about 50 mg/L (for the laboratory device) and of 500 mg/L (for the field device). Laboratory tests showed that it can be used for other particles (e.g. Carbo-Iron®), albeit with higher detection limits. During NanoRem field tests, several susceptibility sensors were installed in arrays in the subsurface at the Spolchemie I site, Czech Republic (nZVI – NANOFEAR 25S and NANOFEAR STAR) and the Solvay site, Switzerland (using milled nZVI NPs called FerMEG12) field sites (Figure 3), together with a temperature sensor and sampling ports.

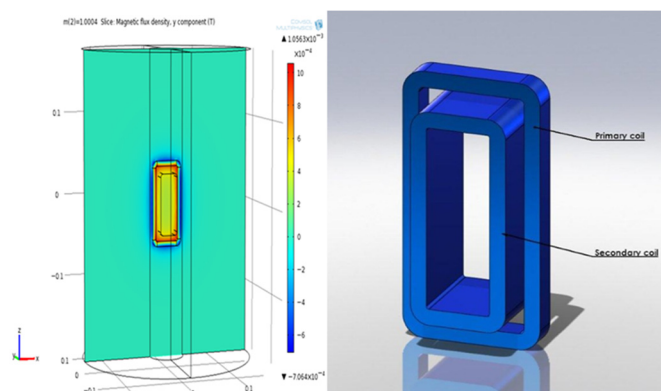


Figure 2. Sketch showing susceptibility probe, electromagnetic field (left), position and shape of the inductors (right), based on theoretical calculations

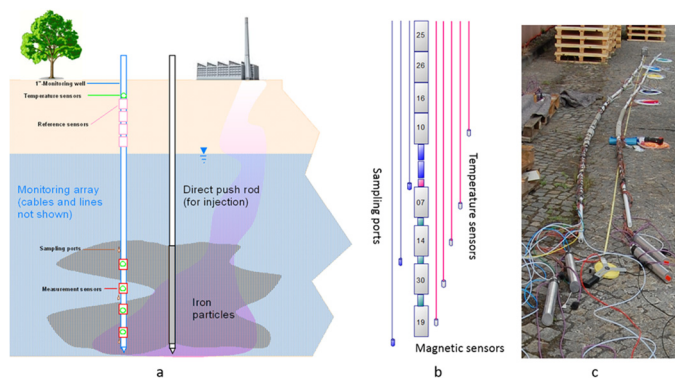


Figure 3. Magnetic susceptibility arrays: (a) use in nanoremediation, (b) components, (c) prior to on site installation.

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The pilot studies showed the arrays were successful in detecting the Fe NPs during injection at both sites. Figure 4 shows the susceptibility signals and the corresponding temperatures at 6.5, 8 and 9.5 m below ground level during injection of NANO FER STAR at the Spolchemie site. Although the monitors at 8 m showed little change during injection, both for magnetic susceptibility and temperature, clear signals were seen at the other two depths and corresponded with the direct push injections at the two closest wells (DP-4 and DP-5).

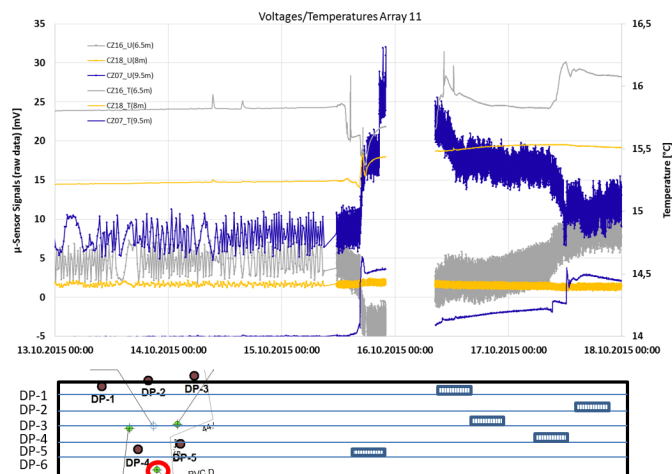


Figure 4. Monitoring results for susceptibility measurements (U) and Temperature (T) from array number 11 (marked with a red circle) at the Spolchemie site. The noisier data signals represent susceptibility. The five injection points are marked with DP and the injection times at these points are marked with the blue rectangles to the right of the map. The missing data in the night from Oct 16 to 17 was caused by a power failure.

Despite the fact that detection limits are slightly higher and instrumentation costs for the magnetic array sensors are greater than those for on-site sampling and measurement, (ca. 1000 EUR per array and 1000 EUR for the electronics), it is one of the truly *in situ* methods and has the advantage of giving continuous logging data.

Turbidity and Fe concentrations

Changes in Fe concentration, pH, temperature, and conductivity can provide a relatively rapid assessment of the spatial and temporal status of the NP suspensions. Providing the chemical properties of injection suspensions are significantly different from those in the groundwater, a number of standard chemical techniques can be applied at site, and can give results within a few minutes of sampling. These include temperature, redox, pH, conductivity as well as turbidity and total Fe content, for which the instrumentation required for on-site measurements is portable and not expensive. Turbidity and spectrophotometry measurement of total Fe concentrations both provide a direct analysis of NP concentrations, and are relatively fast and cheap methods.

NanoRem carried out turbidity measurements on Nano-Goethite (supplied by HMGU and UDE) during tank and field injections using a Turbidimeter (2100N IS, ISO Method 7027). The required sample volume (undiluted) is 2 to 20 ml, the time of measurement about 1 to 2 minutes per sample, and the method can be applied over a concentration range of 0.5 mg/L to 1.0 g/L (depending on the water quality). Figure 5 shows turbidity measurements indicating the distribution of Nano-Goethite particles 20 hours after injection in tank experiments at the VEGAS facility, University of Stuttgart.

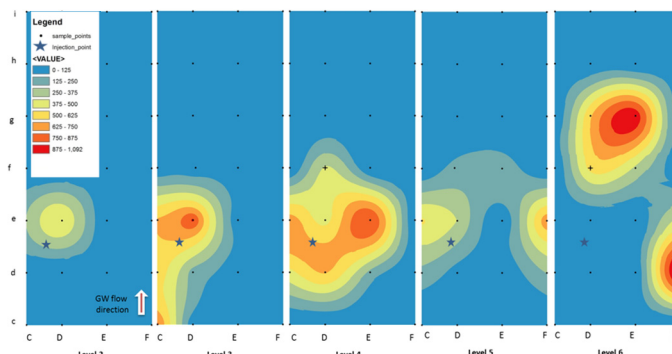


Figure 5. Turbidity data indicating particle distribution at different depths (Level 2-6) 20 hours after injection of Nano-Goethite at the VEGAS facility, University of Stuttgart. The red colour indicates the highest turbidity measurements.

Analysis of total Fe content provides a good overview of NP distribution, and was used at all NanoRem field sites. Whereas acid digestion followed by inductively coupled plasma optical emission spectrometry (ICP-OES) gives a quantitative measurement of Fe content, a rapid on site assessment can be provided with spectrophotometry. Portable spectrophotometers (e.g., Hach DR 2000) can provide measurement of Fe content in 25 ml samples within 15 mins. Information on both total Fe and Fe^{2+} , based on complexation with FerroZine[®] and 1,10-phenanthroline, respectively, can be obtained for concentration ranges of 0.2-200 mg/L (or greater with dilution). As for all methods, detection limits will depend on background concentrations, and the total Fe can be underestimated with large particle sizes due to incomplete dissolution. Tests during injections of nZVI (NANO FER 25S and NANO FER STAR) and Nano-Goethite at the Spolchemie field site showed that the method gave sufficient sensitivity to track NP distribution at the monitoring sites.

4.2 Post-Injection Monitoring

Monitoring during the post-injection phase needs to provide information on not only the concentrations of Fe, but also its speciation in order to understand the fate and reactivity of the injected NPs. For total Fe concentration, measurements on suspensions/liquids and soils/sediments can be carried out directly after acid digestion and measurement using standard chemical analysis (e.g., ICP-OES, or spectrophotometry). Alternatively, for low particle densities, pre-concentration by centrifugation or filtration can be applied to improve detection limits. Specific protocols for acid digestion need to be developed for the different NPs to ensure complete dissolution. The detection limits of all methods will be site specific, depending largely on background concentrations of metals and colloids, and, for Fe-based NPs, dissolved Fe concentrations. Field applications have demonstrated that Mössbauer (for nZVI) can give useful additional information on the time dependent changes in particle state and reactivity, in both water and solid phases. These can be supported by other methods for measurements of structure and oxidation state (e.g. X-ray photoelectron spectroscopy, transmission/scanning electron microscopies, X-ray powder diffraction, X-ray fluorescent spectroscopy).

Mössbauer Spectroscopy

Within the NanoRem project, transmission ^{57}Fe Mössbauer spectroscopy has proved to be a useful tool for the characterisation of nZVI NPs and it represents a unique technique for probing the

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Fe^0/Fe_{tot} ratio in field samples, including the identification of nZVI (NANOFER 25S and NANOFER STAR) NPs in complex environmental and geological matrices. While the Mössbauer spectrometer is compact and portable, the main disadvantage lies in the relatively long counting times. These are typically about 1 day per sample of nZVI, but can be up to 1 week for environmental samples with a low Fe content (i.e., below 1% Fe atoms in the sample), during which time the measured sample could further oxidise. Therefore, samples need to be analysed in a protective atmosphere (e.g., a glove box under nitrogen, see Filip *et al.*, 2014) or pre-concentrated samples are frozen and measured at low temperatures (optimally at 150 K or simply at liquid nitrogen temperature, see Filip *et al.*, 2007). The method has been successfully tested during nZVI injections at Spolchemie I, and measurements taken for suspensions, sediments and soil samples directly proved both the formation of nZVI reaction products and extent of nZVI migration in groundwater conditions (Figure 6).

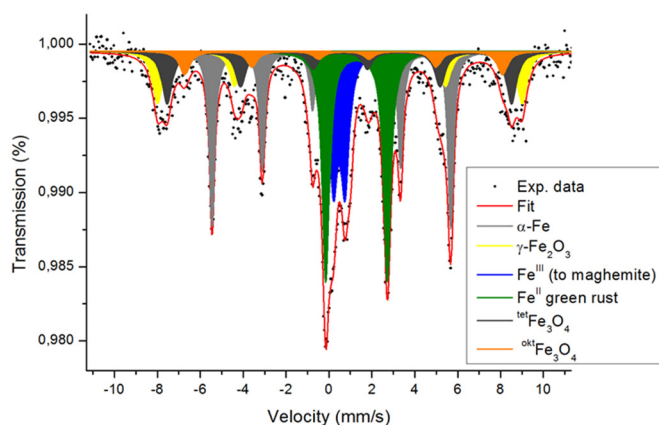


Figure 6. Mössbauer spectrum of sediment from well PV-129, Spolchemie I site, Czech Republic, collected June 2015, 9 months after application of NANOFER STAR. Black dots represent the measured values and the red line a fitted curve. The other colours are reference measurements of different oxidation/crystal states of Fe.

4.3 Methods for Carbo-Iron® and Trap-Ox Fe-Zeolites

Methods for tracing Carbo-Iron® and Trap-Ox Fe-zeolites are still at the laboratory development phase, although preliminary results are promising. A combination of temperature-programmed oxidation (TPO) with parallel CO_2 -analysis seems to be the best approach to distinguish Carbo-Iron® from other carbon containing sediments, and fluorescence labelling has proved to be successful for quantitative analysis of Trap-Ox Fe-zeolites concentration in water samples.

Carbo-Iron® - Temperature-programmed oxidation (TPO)

TPO utilises the fact that the immediate vicinity of the carbon has a fingerprint-like effect on the carbon-specific oxidation temperature. In the case of Carbo-Iron®, the embedment of Fe decreases the incineration temperature significantly in comparison to pure powdered activated carbon (AC) (Bleyl *et al.*, 2012). One can take advantage of this temperature shift to detect carbon-based NPs within a complex matrix containing a natural carbon background. Figure 7 shows the specific incineration patterns in air atmosphere for different batches of Carbo-Iron® colloids and other carbon containing materials. Detection limits of 0.1% wt (1 mg particles per g sediment) are sensitive enough to trace the particle fate within the application area (assuming injection concentrations of g/L).

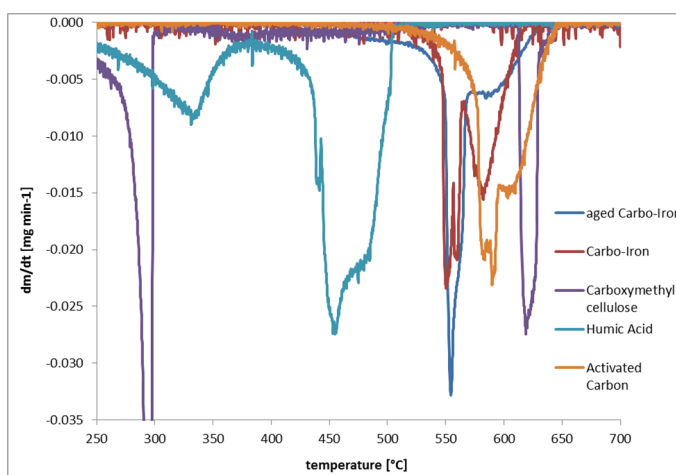


Figure 7. Temperature-programmed oxidation (TPO) of carbonaceous materials in a thermogravimetric balance (TGA-50 Shimadzu: air flow rate 50 mL/min; $m_{sample} = 6-16$ mg; $\Delta T: 10$ K/min; $T_{max} = 700^\circ C$). Aged Carbo-Iron® was obtained by oxidation of fresh Carbo-Iron® particles in aqueous media to generate an Fe-oxide/activated carbon composite, which represents the transformation product of injected Carbo-Iron® suspension. Carboxymethyl cellulose is the colloid stabiliser and coal-derived humic acid a model compound for natural carbon background.

The main challenges are sample preparation of natural heterogeneous aquifer sediment to achieve representative results for the sediment loading with carbon species and the detection of low-concentrated particle fractions ($\ll 0.1$ wt%) in complex matrices. To quantify Carbo-Iron® particles immobilised on sediment grains, predefined loadings of aged Carbo-Iron® on the NanoRem standard material Dorsilit® in a typical expected range of 0.1 wt% up to several wt% have been studied. The combination of TPO with gas analysis (formation of CO_2 and CO) is an additional promising tool to unequivocally trace NPs in natural matrices and increase the sensitivity, reliability and applicability for real sediment samples from field sites (limit of detection ~ 0.03 wt%). As an off-site method the TPO approach can be understood as a complementary tool, which contributes to existing on-site methods such as turbidity and spectrophotometry methods.

Trap-Ox Fe-zeolites - Fluorescence Labelling

Since Trap-ox Fe-zeolites are very close in their composition to natural sediments, particle tracing is a challenge. By intermixing 1% fluorescent-labelled zeolite particles to the Trap-ox Fe-zeolite injection suspension, detection limits of about 1 mg/L (10 $\mu g/L$ labelled particles) can be attained in water samples, which can provide information on NP fate post injection (Gillies *et al.*, 2016). For quantification of Trap-Ox Fe-zeolite content in sediments, analysis of probe compound adsorption can detect zeolite concentrations of 0.01 to 2 wt%, depending on the difference in K_d for the original sediment ($K_{d, sed}$) and the zeolite ($K_{d, zeolite}$). For Trap-Ox Fe-BEA35 on standard porous medium the detection limit of this method is 0.05 wt%, with four orders of magnitude difference in $K_{d, zeolite} = 1500$ L/kg and $K_{d, sed} = 0.17$ L/kg. As zeolites are stable during calcination at $550^\circ C$ in air, this pre-treatment can be used in order to remove adsorbed contaminants from the zeolites or higher natural organic matter loads in the sediment, which otherwise could interfere with probe adsorption analysis.

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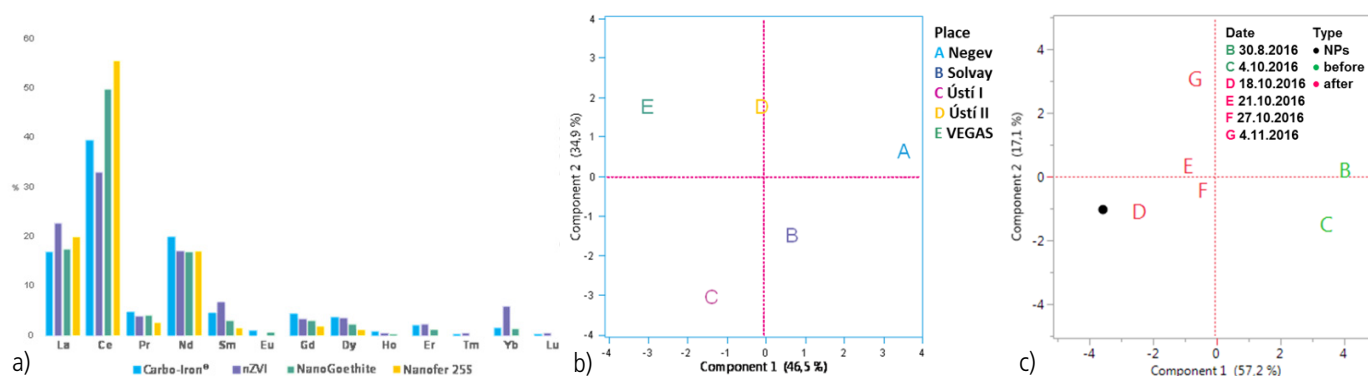


Figure 8. a) Lanthanide profiles of different NPs applied at NanoRem sites, b) Principal Components Analysis of lanthanide profiles of groundwater samples taken at these NanoRem sites before NPs application, and c) Principal Components Analysis of lanthanide profiles of groundwater samples taken at the Solvay site before (green points) and after injection of milled Fe (red points). A significant shift towards the NP profile can be seen immediately after injection (point D), returning towards the background profile later. No shift was seen at more distant wells - data not shown. Taken from Oughton *et al.* (2015, 2017).

4.4 Monitoring for Transport of NPs Out of the Treatment Area

Total Fe content and other chemical parameters can give a reliable picture of the behaviour of injected suspensions in the application area, but more sensitive methods are needed to control for the possible transport of NPs outside the treatment area, often termed “renegade” NPs. Demarcation of low concentrations of Fe-based NPs from background matrix requires greater sensitivity, but tests and developments of a variety of methods within NanoRem have been promising.

By ICP-MS analysis of lanthanides (rare earth elements, RREs) and other trace elements in particles and background groundwater site samples, a group of elements can be selected to “fingerprint” the injected NPs. By applying Multivariate Statistics tools such as Principal Components Analysis, it is then possible to discriminate injected NPs from the background with a much greater degree of sensitivity than by measuring Fe concentrations alone. Detection limits for these methods are extremely low (ng/L levels) in clean media (as tested in laboratory column experiments); but, as for all methods, the performance and applicability in the field is highly dependent on site-specific parameters. Nevertheless, field tests carried out at various NanoRem field injections show good separation of NPs from background components at most sites, with the potential for detection down to sub mg/L levels (Figure 8). Although the analytical costs are higher than for total Fe measurement (from 1000-3000 EUR per remediation site), by targeting selected monitoring wells, measurements can be carried out over a lower spatial and temporal frequency.

5. CONCLUSIONS/FUTURE WORK

The NanoRem project has demonstrated that there are a number of techniques that can be applied for monitoring Fe-based NPs during remediation, and that determination of concentrations at levels below those linked to ecotoxicological effects should be straightforward both within and outside the remediation area. Existing challenges include the discrimination of intact Fe particles from dissolved Fe, since increased Fe concentrations outside the treatment area do not necessarily mean movement of NP. However, fingerprinting techniques using trace element and lanthanides analysis look promising. Future work will consolidate all field experience to provide a quantitative assessment and design of standard operating protocols.

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For further information on NanoRem please visit www.nanorem.eu

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