Effect of Fe-metabolizing bacteria and humic substances on magnetite nanoparticle reactivity towards arsenic and chromium

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ABSTRACT
Magnetite is a magnetic, Fe(II)-Fe(III)-mineral formed through abiogenic and biogenic pathways. It constitutes an attractive material for remediation due to its reactivity, large surface-area-to-volume ratio when present as nanoparticles, and magnetic recoverability. Magnetite can be repeatedly microbiologically oxidized or reduced, but it is unclear how this influences the reactivity of magnetite towards toxic metal or metalloid contaminants. In this study, magnetite (both abiogenic and biogenic) was exposed to microbial Fe(II) oxidation and Fe(III) reduction, before reacted with hexavalent chromium (Cr(VI)) or pentavalent arsenic (As(V)). Results showed microbial reduction of both magnetite types improved the removal rate of Cr(VI) from solution, though surprisingly microbial Fe(II)-oxidation also showed enhanced reactivity towards Cr(VI) compared to un-treated magnetite. Synchrotron based analysis confirmed the formation of Cr(III) at the surface of the magnetite. Reactivity with As was less dramatic and showed un-treated material was able to remove As(V) from solution faster than microbiologically Fe(III)-reduced and Fe(II)-oxidized magnetite. The presence of humic substances was also shown to lead to a decreased reactivity of biogenic and abiogenic magnetite towards As(V) and Cr(VI). Our results imply that Fe-metabolizing bacteria influence the immobilization of contaminants and should be considered when evaluating remediation schemes, especially where Fe-metabolizing bacteria are active.

1. Introduction
The widespread use of heavy metals such as chromium (Cr) and zinc (Zn) in industrial processes, or the release of uranium (U) as a waste product of nuclear energy production has left a legacy of pollution in many areas around the world (Riley et al., 1992; Jung et al., 2007; Pidchenko et al., 2017). This combined with the presence of the toxic metalloid arsenic (As) in groundwater aquifers used for drinking water and irrigation highlights the global importance of finding a solution to treating metal-contaminated land and water and reduce the threat
posed to human health. Chromium commonly occurs in two stable oxidation states, trivalent Cr(III) and hexavalent Cr(VI). Cr(VI) is a strong oxidizing agent which is toxic to plants, animals and humans and is known to be carcinogenic (Barnhart, 1997). World Health Organization (WHO) guidelines recommend a maximum safe drinking level concentration of 50 μg L⁻¹ (WHO, 2011). In comparison, Cr(III) is considered to be much less hazardous as it poorly dissolves at neutral pH. Arsenic contamination in Bangladesh has been described as the largest mass poisoning of a population in history due to the presence of naturally occurring toxic levels of inorganic arsenic in ground waters. It is estimated that between 35 million and 75 million of the Bangladeshi population of 125 million suffer a serious health risk from drinking arsenic contaminated water (Smith et al., 2000). The pollution of As is a worldwide problem with contamination of ground water present in Argentina, Chile, China, India, Mexico, Taiwan, Thailand, the United States as well as parts of Eastern Europe such as Hungary, Bulgaria and Romania.

Several strategies currently exist for treating metal and metalloid contaminated water, in particular through adsorption to synthetic Fe (III) (oxyhydr)oxides minerals such as akaganeite and ferrhydrite (Kleiner et al., 2011). Alternatively, magnetite (Fe₃O₄), could also be applied in adsorption based reactive filter bed systems for treating such contamination. Magnetite is a naturally occurring mixed-valent Fe mineral, containing both Fe(II) and Fe(III), which has been shown to be highly reactive towards organic and inorganic pollutants including chromium and arsenic as well as azo dyes, U(VI), H₂SeO₃, or Hg(II) (Gorski et al., 2010; Huber et al., 2012; Swindle et al., 2015). Organic matter such as humic substances (HS) occur in aquatic and terrestrial ecosystems where they undergo important reactions, e.g. complexation (Lovely et al., 1987; Evans and Heller, 2003; Sundman et al., 2017). Several studies have shown that the reactivity of the mineral is dependent upon particle size (Vikesland et al., 2007; Swindle et al., 2014), stoichiometry (i.e. Fe(II)/Fe(III) ratio) (Gorski et al., 2010; Huber et al., 2012; Latta et al., 2012), and the presence of organic matter (Swindle et al., 2015). Organic matter influences the adsorption of metals, especially Cr(VI) (Nakayasu et al., 1999; Jiang et al., 2014; Joshi et al., 2018). While these studies address the influence of HS, such as how it can affect nanoparticle dispersion properties or mobility in an aquifer, they do not consider the effect of magnetite stoichiometry on the mineral reactivity. Stoichiometry has been shown to affect the redox conditions favoured Fe(II)-oxidizing or Fe(III)-reducing bacteria, respectively. Before exposure to respective bacteria, magnetite (~ 1 g L⁻¹) was added to modified basal medium, pH 7, NaHCO₃ buffer and allowed to equilibrate for 3 days before addition of bacteria. Acetate (1 mM) was added to all reduction experiment bottles from a stock solution (1 M sodium acetate; pH 7). Dense cell suspensions (approximately 10⁸ cells/ml) of either TIE-1 or G. sulfurreducens were added into the bottles, equivalent to 100% (v/v) inoculum, i.e. cell density in reaction vessels were equivalent to pure cultures, and left for 24 h. Fe(II) oxidation experiments were incubated in artificial light (True Light 15 W/5500 K) at 20 °C whereas Fe(III) reduction experiments were incubated in the dark at 30 °C. After reduction or oxidation, biomass was removed from A-mag and B-mag samples by washing magnetic minerals with anoxic ultrapure H₂O₂ (Milli-Q, Merck Millipore). This routine was repeated three times. Stock solutions of both A-mag and B-mag were made containing 20 g L⁻¹ magnetite in an anoxic buffered solution (NaHCO₃ 22 mM at pH 7).

Both A-mag and B-mag were microbiologically oxidized or reduced by exposure to the phototrophic Fe(II)-oxidizing bacteria, Rhodopseudomonas palustris strain TIE-1 or the Fe(III)-reducing bacteria, Geobacter sulfurreducens respectively. Before exposure to respective bacteria, magnetite (~ 1 g L⁻¹) was added to modified basal medium, pH 7, NaHCO₃ buffer and allowed to equilibrate for 3 days before addition of bacteria. Acetate (1 mM) was added to all reduction experiment bottles from a stock solution (1 M sodium acetate; pH 7). Dense cell suspensions (approximately 10⁸ cells/ml) of either TIE-1 or G. sulfurreducens were added into the bottles, equivalent to 100% (v/v) inoculum, i.e. cell density in reaction vessels were equivalent to pure cultures, and left for 24 h. Fe(II) oxidation experiments were incubated in artificial light (True Light 15 W/5500 K) at 20 °C whereas Fe(III) reduction experiments were incubated in the dark at 30 °C. After reduction or oxidation, biomass was removed from A-mag and B-mag samples by washing magnetic minerals with anoxic ultrapure H₂O₂ (Milli-Q, Merck Millipore) as described above.

2.2. Reactivity experiments

Reactivity experiments between magnetite with As(V) and Cr(VI) focused on quantifying the removal of the two different pollutants from solution. An anoxic As(V) stock solution (10 mM) was prepared by dissolving As oxide, Na₂As₂O₄ (Fisher Scientific UK), in anoxic ultrapure H₂O. An anoxic Cr(VI)-stock solution (10 mM) was made with K₂Cr₂O₇ dissolved in 0.5 M sodium carbonate solution. The pH of both solutions was adjusted to pH 7. All experiments were conducted in an anoxic glovebox filled with N₂ gas and the pH was kept constant using a 22 mM bicarbonate buffer at pH 7. Experiments were conducted in triplicate with Na₂CO₃ atmosphere and kept on a rolling shaker in the dark. At each time point, samples were taken for dissolved and solid phase Fe³⁺ and Fe⁵⁺, total As or Cr and for As(V) or Cr(VI) speciation. Aqueous Fe³⁺ and Fe⁵⁺ were analyzed via the Ferrozine assay (Stookey, 1970). For total As and Cr, samples were filtered (0.22 μm) and diluted with 2% HNO₃ prior to analysis by ICP-MS and MP-OES.
respectively. As redox speciation was determined using HPLC ICP-MS according to (Daus et al., 2002). Changes to Cr(VI) were followed using UV–vis spectrophotometer using acidified diphenyl carbazide in acetone as Cr(VI)- complexing ligand (Skougstad et al., 1978).

Cr(VI) removal data as a function of time were fitted in OriginPro 8.5.0 by a pseudo-first-order rate equation of the form:

\[ y = C_0 e^{-kt} + C_{ond} \]  

(1)

Where \( k \) corresponds to the rate constant, \( C_0 \) is the initial concentration and \( C_{ond} \) is the final concentration remaining in solution. The half-life \( \tau \) of the reaction was determined by:

\[ \tau = \frac{\ln 2}{k} \]  

(2)

2.3. Humic substance experiments

Pahokee Peat Humic Acid, from the International Humic Substances Society, was used as humic substance (HS). The HS-solution was prepared according to Jiang and Kappler (2008) but using a bicarbonate buffer instead of phosphate buffer. Briefly, HS (IHSS) was dissolved in 22 mM bicarbonate buffer and placed onto a rolling shaker for 2 h. HS was sterilized by filtration through a 0.22 μm filter to a sterile bottle. To reduce the HS, Pt-pellets were added to the bottle, with headspace changed to N2 and placed on a rolling shaker again. After 22 h, the headspace was changed to N2/CO2.The effect of HS on magnetite reactivity was evaluated in a set-up with 40 mg L\(^{-1}\) HS, 25μM As(V) or 200μM Cr(VI) and 2.4 mg L\(^{-1}\) A-mag or B-mag. The time-dependent As- or Cr- and Fe-speciation was analysed using the same methods as described above. Whenever possible, the HS was covered with aluminium foil to avoid light-induced reactions.

2.4. Mineralogical analysis

For synchrotron-radiation based X-ray absorption spectroscopy (XAS) and X-ray magnetic circular dichroism (XMCD) measurements, dried magnetite samples were loaded onto carbon tape, which was fixed to a sample holder. All sample preparation was carried out in an anoxic glovebag (100% N\(_2\)) to prevent abiogenic oxidation. The samples were loaded into the Portable Octupole Magnet System (POMS) system on beamline I10 at the Diamond Light Source (van der Laan and Figueroa, 2014) in a backflow of nitrogen and held in vacuum for the duration of the measurement. Samples were measured using circularly polarized X-rays across the photon energy range of the Cr L\(_{2,3}\) edge (560–600 eV) with spectra recorded using total-electron-yield (TEY) detection in energy steps of 0.2 eV with an effective probing depth of ~45 Å (Gota et al., 2000). XMCD data were obtained from the difference between X-ray absorption (XAS) spectra collected under opposite magnetic fields (± 0.5 T).

Samples were prepared for transmission electron microscopy (TEM) by dropping magnetite nanoparticles from anoxic stock solutions onto a carbon-coated 200 mesh copper grid. Excess water was removed by blotting. Micrographs were collected using a JEOL 2100 F microscope operating at 200 kV and equipped with a Schottky emission gun, a high-resolution UHR pole piece, and a Gatan US4000 CCD camera. Particle sizes were determined by measuring the longest length of approximately 250 particles with ImageJ software.

3. Results and discussion

Previous studies have shown that both abiogenic and biogenic magnetite nanoparticles synthesized following the same protocols as in the present study have an average diameter of 13.2 ± 2.4 and 13.6 ± 2.1 for A-mag and B-mag respectively (Fig. A1). The stoichiometry (i.e. Fe(II)/Fe(III) ratio) of A-mag and B-mag after exposure to Fe(II)-oxidizing *Rhodopseudomonas palustris* TIE-1 or Fe(III)-reducing bacteria *Geobacter sulfurreducens* is shown in Table 1. Using the ferrozine chemical extraction method we found that the A-mag was more oxidized by the Fe-oxidizer compared with B-mag. The Fe(II)/Fe(III) ratio varied between 0.37 and 0.54.

### Table 1

<table>
<thead>
<tr>
<th>Sample</th>
<th>Magnetite</th>
<th>Redox state</th>
<th>Fe(II)/Fe(III)</th>
</tr>
</thead>
<tbody>
<tr>
<td>A-mag(_N)</td>
<td>Abiogenic</td>
<td>Native</td>
<td>0.45 ± 0.04</td>
</tr>
<tr>
<td>A-mag(_O)</td>
<td>Abiogenic</td>
<td>Oxidized</td>
<td>0.51 ± 0.02</td>
</tr>
<tr>
<td>A-mag(_R)</td>
<td>Abiogenic</td>
<td>Reduced</td>
<td>0.51 ± 0.07</td>
</tr>
<tr>
<td>B-mag(_N)</td>
<td>Biogenic</td>
<td>Native</td>
<td>0.42 ± 0.02</td>
</tr>
<tr>
<td>B-mag(_R)</td>
<td>Biogenic</td>
<td>Reduced</td>
<td>0.54 ± 0.17</td>
</tr>
</tbody>
</table>

3.1. Reactivity of magnetite to Chromate (Cr(VI))

The results of the reaction between microbially modified A-mag and B-mag nanoparticles (1 g L\(^{-1}\)) with Cr(VI) (200 μM) are shown in Fig. 1. The change in aqueous Cr(VI) concentration in the presence of native, oxidized or reduced A-mag or B-mag were followed using the 1,5-diphenyl carbazide (DPC) assay (Fig. 1) (Byrne et al., 2011). In all experiments and for all conditions tested, the initial concentration of Cr (VI) was much lower (<100 μM) than the initially added concentration (200 μM). This suggests that approximately 50% of the Cr(VI) was removed almost instantaneously.

Cr(VI) removal by magnetite is based on a redox reaction where three ferrous iron ions are oxidized for every Cr(VI) reduced (Peterson et al., 1996; White and Peterson, 1996; Kendelewicz et al., 2000). Therefore, we anticipated the increases in the Fe(II)/Fe(III) ratio of magnetite after exposure to Fe(III)-reducing bacteria (Table 1), that the A-mag\(_N\) and B-mag\(_N\) would both show a higher reducing capacity towards Cr(VI) (due to enhanced levels of surface Fe(II)) than the non-modified native magnetite (A-mag\(_O\) and B-mag\(_O\)) and that the magnetite exposed to Fe(II)-oxidizing bacteria (A-mag\(_R\) and B-mag\(_R\)) would exhibit the lowest reactivity of all (due to depleted levels of surface Fe (II)). In reality, whilst the hypothesis that microbially reduced magnetite would be the most reactive held true, the microbially oxidized magnetite was surprisingly more efficient at removing Cr(VI) from solution than the native magnetite (Fig. 1). Table 2 shows the differences in rate constants after fitting with a pseudo-first-order rate Eq. 1. For both abiogenic and biogenic native magnetite, the rate constant is lower than either the reduced or oxidized magnetite. This can be interprited more easily by the half-life of the reaction, i.e. the time required for the concentration to reach half of the initial value, which is much longer for native magnetite than the treated magnetite. This indicates that changing the stoichiometry either through reduction or oxidation had a clear effect on the speed at which Cr(VI) could be removed from solution. Furthermore, the extent of Cr(VI) removal, shown by \( C_{ond} \) is lower for native magnetite than oxidized or reduced magnetite. The rate constant is highest for the reduced magnetite as expected, indicating that it is the most reactive material with respect to Cr (VI). However, reduced magnetite also shows a much lower starting concentration (\( C_0 \)) than the other samples (i.e. 10.7 and 37 μM for A-mag\(_R\) and B-mag\(_R\) respectively) which was due to the fact that most of the reaction appeared to take place before the first sample could be taken (approximately 1–2 min after addition of Cr(VI)).

The half-life of the oxidized magnetite is close to half the values of native magnetite for both abiogenic and biogenic magnetite. This result was unexpected as we anticipated that oxidation of the magnetite would yield a lower surface Fe(II)/Fe(III) ratio, hence lower reactivity...
towards Cr(VI). We hypothesized that the enhanced reactivity of oxidized magnetite could be either a result of reactive organic compounds which remain fixed to the surface of the bio-oxidized magnetite from the Fe(II)-oxidizing bacteria, or due to the fact that the redox potential at the mineral surface has been modified. For instance, it has been shown that some species such as Cr(III) can form complexes with organics ranging from relatively simple molecules such as pyruvate, to more complex natural organic matter (NOM) (Luo et al., 2010; Gustafsson et al., 2014). Alternatively electrons can be donated to magnetite by organic matter such as humic substances (Sundman et al., 2017). Furthermore, porous or semi-intact cells have been shown to act as biosorbents to different metals including Cr(III) (Mullen et al., 1989; Churchill et al., 1995). Despite these potential mechanisms, the exact reason for enhanced removal capacity in microbially oxidized magnetite remains unknown. However, the results demonstrate the remarkable enhancement in remediation capacity for microbially modified magnetite compared to native material. This is especially true for the microbially reduced magnetite (~1 g L⁻¹), which was able to remove almost all Cr(VI) (starting concentration 200 μM) to less than 15 μM in under one hour. In comparison, native magnetite which has not been treated by any bacteria had a remaining concentration of Cr(VI) of 100 μM after the same time period and had still not removed all Cr(VI) after 2 weeks.

In order to determine the speciation of Cr after removal from solution by magnetite, synchrotron-radiation based X-ray absorption spectroscopy (XAS) was performed on samples A-magO and A-magR. This technique is element specific and is highly sensitive to the oxidation state of an element. Fig. 2A shows the Cr L₂,₃ edge of both magnetite samples after reaction with Cr(VI). The spectrum corresponding to the magnetite after reaction shows a clear similarity to that of the Cr (III) reference compound rather than the Cr(VI) reference compound. This indicates that the magnetite was able to completely reduce the highly toxic and mobile Cr(VI) to Cr(III) which is much more stable and generally regarded as much less toxic. Additionally, X-ray magnetic circular dichroism (XMCD) was performed on the mineral after reaction with Cr(VI). XMCD is a magnetic probe that only yields a spectrum if the element under analysis is magnetically ordered. Magnetic ordering in magnetite can only occur if the element under analysis (in this case Cr) is present within the crystal lattice (Telling et al., 2009). The clear XMCD signal observable at the Cr L₂,₃ edge in Fig. 2B shows that the Cr is magnetically ordered and is therefore present in the mineral structure of the magnetite. A calculated spectrum for Cr(III) is shown for comparison (van der Laan and Thole, 1991).

These results suggest that magnetite nanoparticles exposed to Fe(II)-oxidizing or Fe(III)-reducing bacteria are able to readily remove Cr(VI) from solution and reduce it to the less toxic, less mobile Cr(III). Thus, the magnetite is able to efficiently remove and immobilize the Cr in a stable way in order to lower the risk of remobilization at a later time. This Cr(III) is incorporated directly into the mineral which would suggest it is likely more stable and less likely to leach out of the magnetite than if it was simply adsorbed to the surface of the mineral. Such a result has significant implications for the use of magnetite nanoparticles in the treatment of Cr pollution.

### Table 2

<table>
<thead>
<tr>
<th></th>
<th>k (1/s)</th>
<th>τ (h)</th>
<th>C₀ (μM)</th>
<th>C₉₅ (μM)</th>
<th>R²</th>
</tr>
</thead>
<tbody>
<tr>
<td>A-magO</td>
<td>6.64E-06</td>
<td>28.9</td>
<td>86.30</td>
<td>9.73</td>
<td>0.97</td>
</tr>
<tr>
<td>A-magR</td>
<td>2.32E-04</td>
<td>0.838</td>
<td>10.72</td>
<td>2.26</td>
<td>0.93</td>
</tr>
<tr>
<td>A-magO</td>
<td>1.56E-05</td>
<td>12.4</td>
<td>80.03</td>
<td>4.58</td>
<td>1.00</td>
</tr>
<tr>
<td>B-magO</td>
<td>5.63E-06</td>
<td>34.2</td>
<td>81.12</td>
<td>15.05</td>
<td>0.97</td>
</tr>
<tr>
<td>B-magR</td>
<td>1.60E-04</td>
<td>1.20</td>
<td>37.20</td>
<td>5.16</td>
<td>0.92</td>
</tr>
<tr>
<td>B-magO</td>
<td>1.09E-05</td>
<td>17.7</td>
<td>92.06</td>
<td>5.19</td>
<td>0.98</td>
</tr>
</tbody>
</table>

### 3.2. As(V) reactivity

Addition of 0.3 μmol/m² and 0.5 μmol/m² As(V) to A-mag and B-mag, respectively, showed similar adsorption despite slightly different loading caused by differences in their surface area (Fig. 3). This is in contradiction to a study by Kleinert et al. which suggested that cellular biomolecules at the surface of biogenic Fe(III)-minerals block sorption sites, thus limiting the availability of sorption sites for As to bind (Kleinert et al., 2011). Weng et al. reported that biogenic Fe(III) minerals contain organic compounds derived from the bacteria and therefore biomolecules such as carbohydrates, fragments of cells (i.e. carbohydrates, lipids, nucleic acids and proteins which typically constitute bacteria) may compete with As for sorption sites which is not shown in our results (Weng et al., 2009). As(V) concentration in solution decreased for all types of magnetite, though there was no clear increase in the concentration of As(III) (Fig. A.2). Since less than 1% of the initially 25 μM As(V) was reduced to As(III) in the presence of magnetite (Fig. A.2) no major redox reactions are taking place between As(V) on the surface of magnetite and all As removal results from sorption.

Fitting the As_{sor,aq} data with a first order exponential decay curve only yielded satisfactory results for the A-mag series, while all B-mag fits had low R² (less than 0.35) suggesting a poor correlation between data and fit (Table A.1). The rate constant was similar for all three treatments in the A-mag series, corresponding to half-life between 1.1, 1.75 and 2.1 h for A-magO, A-magR, A-magR respectively.

Aqueous Fe analysis showed that Fe_{aq} concentration for microbially reduced A-mag increased over time by 225%, with no significant increases in Fe_{aq} observed for oxidized or native A-mag (Fig. A.3). Reduced B-mag showed a release of Fe_{aq} of 250% over the same time period (Fig. A.3) while oxidized or native B-mag showed no significant changes. The increasing Fe_{aq} concentration over time in the presence of microbially reduced magnetite indicates that during As adsorption the contaminant might substitute for Fe, causing a release of Fe into solution. However, any exact redox reaction mechanism cannot be concluded by only considering changes in aqueous Fe concentrations as they do not consider dissolution and formation of dissolved colloids.
Exposure to Fe(II)-oxidizing and Fe(III)-reducing bacteria reduced the magnetite capacity to remove As(V). More specifically, unmodified native magnetite displays the fastest As(V)-removal rates and highest capacity to remove As(V). These results have clear implications in the ability for magnetite to be used as a remediation agent for different pollutants and indicates that modification by bacteria can on the one hand be used to enhance reactivity, whilst on the other hand restricts reactivity depending on the route of contaminant removal: the adsorption capacity, e.g. As adsorption, worsened, whereas the reduction and subsequent formation of a new phase, e.g., Cr incorporation in a new phase, benefitted from the microbial activity.

3.3. Effect of HS

Subsequent experiments were performed to investigate the influence of humic substances (HS) on the reactivity of magnetite towards both As(V) and Cr(VI). In these experiments, only native abiotic (A-mag\(_N\)) and native biogenic (B-mag\(_N\)) were tested. The effect was expected to be dependent on the concentration of added HS (Tombácz et al., 2013; Swindle et al., 2015) with low concentrations expected to neutralize the net positive charges of the magnetite nanoparticles at pH below the point of zero charge (PZC) and thereby favour aggregation and sedimentation of the particles. In contrast, higher concentrations of HS may change the charge of the particles which stabilizes the dispersed particles. Another important process is competition between NOM and negatively charged anions, e.g. as Redman, 2002 (Redman et al., 2002).

The influence of the presence of HS when both types of magnetite were reacted with Cr(VI) are shown in Fig. 4A. For both types of magnetite, the presence of HS appeared to lead to a decrease in the overall capacity of the magnetite to remove Cr(VI) from solution, likely due to competition between HS and Cr(VI) for reactive surface sites on the mineral (Redman et al., 2002; Swindle et al., 2015). These results are in line with those of Swindle et al., 2015 who investigated the effect of different types and concentrations of organics on Cr(VI) removal by abiogenic magnetite sample (Swindle et al., 2015). Here we show that the same effect is observed for both abiogenic and biogenic magnetite, i.e. A-mag and B-mag (Fig. 4). The rate constant was actually higher when HS were present than without (Table A.2), corresponding to a shorter reaction half-life and suggesting that the presence of HS had a stimulatory effect on the reaction rate of the magnetite with Cr(VI). Nevertheless, the final concentration of Cr(VI) removal was noticeably lower when HS was present than without. There are different potential reasons why HS lowers the removal rate of Cr(VI) from solution during
the reaction with magnetite including the effect of particle aggregation. Humic acid has been shown to modify the surface charge properties of magnetite and affect its aggregation properties (Illés and Tombácz, 2006), leading to either larger or smaller aggregates depending on the pH. At the pH we performed our experiments, the HS is expected to increase the aggregation of the nanoparticles, and thus decrease the overall surface area over which the Cr(VI) can react. Alternatively, coverage of the magnetite surface by HS could lead to a decrease in the amount of surface Fe(II) available for reaction with Cr(VI), and again lead to a decrease in reactivity of the magnetite in the presence of HS. Whilst the overall reason for a decrease in magnetite reactivity in these experiments is not clear, it is likely that the interference of natural organic matter should be considered for any application of magnetite in real world scenarios.

Both A-mag and B-mag removed around 85% of the initially added As(V) within 7 days (Fig. 4B). The higher surface loading of As in B-mag (0.5 μmol m⁻²) than A-mag (0.3 μmol m⁻²) suggests that B-mag binds As more efficiently than A-mag. This decreased As sorption to magnetite with HS present was consistent with Yean et al. where they observed that As sorption to magnetic nanoparticles was less effective in the presence of NOM (Yean et al., 2005). In accordance with the experiments conducted in the absence of HS, in the presence of HS, As speciation was still dominated by As(V) at every time point (Fig. 4). In contrast to As(V), As(III) concentrations for both A-mag and B-mag fluctuated with no clear trend visible (Fig. A.4). Compared to As speciation without HS present, the As(V) concentrations are higher in the setup with HS, whereas the As(III) concentrations are in the same range.

The increasing concentrations of aqueous Fe²⁺ are similar to those observed in the set-up without HS (Fig. A.5), suggesting that HS does not result in any significant net redox reactions under the conditions used in this study. This is in contrast with Redman et al. who showed that every NOM sample they studied showed active redox behaviour towards As species, which indicates that NOM may greatly influence redox speciation (Redman et al., 2002).

4. Conclusions

The results presented here indicate that microbial reduction or oxidation of magnetite can influence mineral reactivity towards environmentally relevant pollutants including As and Cr. Here we have shown that for chromium, microbial Fe(III) reduction and Fe(II) oxidation can enhance the reactivity of magnetite towards Cr(VI). It was anticipated that the reduced magnetite nanoparticles would have a more reactive surface with higher reducing capacity (due to enhanced levels of surface Fe(II)) than native magnetite and that oxidized magnetite would exhibit the lowest reactivity of all magnetite samples with Cr(VI) (due to depleted levels of surface Fe(III)). In reality, whilst the hypothesis that bio-reduced magnetite would be the most reactive held true, the bio-oxidized magnetite was surprisingly more efficient at removing Cr(VI) from solution than the non-modified magnetite. It is thought that this could be either a result of reactive organic compounds which remain fixed to the surface of the bio-oxidized magnetite from the Fe(II)-oxidizing bacteria, or due to the fact that the redox potential at the mineral surface has been modified. For the As experiments, neither reduced nor oxidized magnetite showed any enhanced reactivity towards As(V). In fact, the biological modification of the mineral surface led to a decrease in the rate of As(V) removal from solution compared to native magnetite. This suggests that in the case of arsenic species, biological modification may in fact limit the sorption of As to the magnetite mineral surface for reasons which are currently unclear. This also raises questions about whether such behaviour might also be seen for other metalloids such as antimony. The results presented here provide potential routes through which magnetite could be improved for application as a remediation technique. However, it should be noted that some of the reactivity described towards Cr and As could be due to remaining biomass associated with the magnetite that could not be removed during preparation of the precursor materials. Fe (III) (oxyhydr)oxides minerals such as akaganéite or ferrihydrite are already being used as adsorbent materials for drinking water filtration in reactive filter bed systems (Kleinert et al., 2011). The A-mag and B-mag developed here, in either their native, reduced or oxidized forms could be applied in similar setups to treat contaminated water supplies. However, maintaining the long term performance of these materials, especially under oxic conditions will be especially challenging. Oxidation by microbes could potentially offer one way to passivate the surface of the magnetite against abiotic oxidation, while still maintaining reactivity and will be subject to further study.

Author contributions

All authors have given approval to the final version of the manuscript.

AS – Designed the experimental plan, did part of the experimental work and co-authored the manuscript
AV – Carried out experimental work, compiled and analyzed the data, contributed towards the manuscript
KA – Carried out experimental work, compiled and analyzed the data, contributed towards the manuscript
AIF – Helped to design the synchrotron experiments and analyze data
GvdL – Helped to design the synchrotron experiments and analyze data
BD – Contributed to the design of the sampling procedure for As speciation studies and carried out analytical measurements
AK - Designed the experimental plan and contributed towards the manuscript
JMB – Designed the experimental plan and co-authored the manuscript

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Declaration of Competing Interest

The authors declare no competing financial interest

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Appendix A. Supplementary data


References


