Stereo soft X-ray microscopy and elemental mapping of haematite and clay suspensions

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Summary

The combination of high-resolution chemically sensitive soft X-ray microscopy with stereo imaging and processing techniques presented here forms a novel tool for the investigation of aqueous colloidal systems. Information about the spatial distribution within the sample is provided with small calculation effort processing just a pair of stereo micrographs. Thus, the extension towards investigation of dynamical behaviour is possible on the part of the experiment as well as of the processing.

The potential of this technique is demonstrated with applications in aqueous soil and clay samples. Within these samples, haematite particles are identified taking advantage of the elemental contrast at the Fe-L edge around \( E = 707 \) eV. In combination with stereo microscopy, information about spatial arrangements are revealed and correlated to electrostatic interactions of the different mixtures, addressing to an actual question of soil scientists. The technique allows in-situ sample manipulation, which is demonstrated by a test specimen where particles were added during imaging.

Introduction

Soft X-ray microscopy has become a mature analytical tool with various applications to nanoscale behaviour in materials, environmental and biological sciences. A compendium of the achievements can be found for example in Susini et al. (2003) and Aoki et al. (2006). As well known for soft X-ray microscopy, recording the X-ray images within the so-called water window, that is, using photon energies between the K-absorption edge of oxygen (\( E \approx 530 \) eV or \( \lambda = 2.34 \) nm) and carbon (\( E \approx 280 \) eV or \( \lambda = 4.43 \) nm), is especially suitable for imaging samples in aqueous media (Wolter, 1952). The optimum optical thickness is determined by the \( 1/e \) ratio of the transmitted radiation, which in the water window extends up to 10 \( \mu \)m around the oxygen K-edge (Kirz et al., 1995). However, to detect the iron-containing particles added to the clay or soil samples presented as test samples, element contrast around the iron L 3 absorption edge at 707 eV has been used and therefore imaging energies between 704 and 710 eV have been chosen. Due to the higher X-ray absorption in water at these energies, thinner samples thicknesses of approximately 2 \( \mu \)m are required.

Since spatial resolution scales with the wavelength, soft X-ray microscopy enables high-resolution transmission images of much higher spatial resolution compared to optical microscopy. Currently the best spatial resolution in soft X-ray microscopy has been achieved down to 15 nm resolution (Chao et al., 2005). This gives a unique way of studying biological or environmental samples in their natural aqueous state without staining or fixation. By changing the incident X-ray energy and taking one image above and one below an absorption edge, the distribution of the element corresponding to the absorption edge can be visualized. This can be done either by just comparing the images and identifying the stronger absorbing structures or by creating a map of the elemental distribution by dividing these two images.

The investigation of aqueous samples from the environment benefits particularly from the advantages of soft X-ray microscopy, concerning for example the relationship between soil colloids and iron particles. Iron is an abundant cation in soils and groundwater aquifers (Sparks, 1995; Stevenson & Cole, 1999; Tan, 2000). It can occur bivalent or trivalent, depending on the redox condition. Ferrous iron, Fe\(^{2+}\), is abundant under anaerobic conditions but unstable under aerobic conditions. At the groundwater surface or by mixing of anaerobic bank filtrate and aerobic water the groundwater may get in contact with oxygen. Iron will be oxidized to ferric iron, Fe\(^{3+}\), a source for precipitation. It gives rise to the formation of new colloidal particles (Hofmann et al., 2001).
An increasing formation of ferric hydrates causes turbidity in soil water and groundwater and is considered harmful. The use of water for engineering purposes containing too much iron may cause corrosion and the deposition of colloidal sized iron particles. The two most abundant colloidal iron oxides in soils are goethite, needle-like crystals and haematite, hexagonal platelets, followed by the poorly crystallized hydrous ferrihydrite (Sparks, 1995). The colloid fraction in soils, mainly clays, oxides and organic substances, show significant effects on many soil chemical processes due to their high specific-surface areas and especially in the micro pore system built with colloids. The surface of these colloids is charged, with stable layer charges, but pH dependent edge charges (Lagaly, 1993). The clays as well as humic substances show within a wide pH-range (3 ≤ pH ≤ 8) negative edge charges, whereas iron oxides are positively charged in the same range (Sparks, 1995). Therefore, iron oxides are able to bind other soil particles forming a micro texture. Small iron oxide particles might penetrate the formerly lose colloidal matrix and solidify it due to electrostatic interaction. A change in the texture of the microstructure of soils, which means to change the morphology of associations of soil colloids, influences for example runoff and erosion losses within soils. The relationship between iron oxide and the stability of soil colloid associations is, however, still debated in soil science, for example by Duiker et al. (2003) or Rhoton et al. (2003).

The application of the here presented method of stereo soft X-ray microscopy reveals this relationship in three dimensions. Thus, not only the potential of the technique is demonstrated, but a deeper insight into a scientific topic is addressed as well.

Materials and methods

For the studies presented here, the high-resolution full-field soft X-ray transmission microscope of the Center for X-Ray Optics located at beamline 6.1.2 of the Advanced Light Source, Lawrence Berkeley Laboratory (Berkeley, CA, USA) has been used (Meyer-Ilse et al., 2000; Thieme et al., 2003; Fischer et al., 2006). The X-ray optical set-up of this transmission X-ray microscope consists of Fresnel optics for both the condenser zone plate and the micro zone plate as shown in Fig. 1. The condenser zone plate collects the synchrotron radiation emitted from a bending magnet and focuses it onto the object. In addition, it works in combination with the pinhole in front of the object as a linear monochromator (Niemann et al., 1974). It reduces the bandwidth of the radiation illuminating the object to about $E/\Delta E \approx 500$. A micro zone plate downstream the object acts as a high-resolution X-ray objective and forms an enlarged image of the object in the image plane. The image is recorded with a backside illuminated slow-scan CCD-camera. Exposure times are typically in the range of several seconds. For the experiments, a micro zone plate with an outermost zone width of $d_{\text{m}} = 25 \text{ nm}$ was used to take high-resolution images around the Fe-L edge at approximately $E = 707 \text{ eV}$ or $\lambda = 1.75 \text{ nm}$. According to

$$\sigma = 1.22dr_n$$

the lateral resolution is $\sigma \approx 30 \text{ nm}$. With

$$\delta = 4 \frac{dr^2}{\lambda}$$

the depth of focus in the experiments has been $\delta \approx 1.43 \mu\text{m}$ (Attwood, 1999). The magnification was about 2800. The CCD camera has been used either with $2048 \times 2048 \text{ pxl}^2$ or in a $1024 \times 1024 \text{ pxl}^2$ binned mode, resulting in a pixel size of either 4.5 nm or 9 nm in the object plane. Thus, the images have been sufficiently oversampled in regard to the optical resolution.

Due to the high absorption in air of X-rays with such a low energy, the optical path has to be in vacuum. To allow for samples in aqueous media, the sample stage is therefore located in a small air gap between the vacuum chamber containing the condenser and the vacuum chamber holding micro zone plate and CCD camera. To optimize between X-ray transmission and sample handling, the gap is typically about 200 nm wide. Both chambers are sealed with Si$_3$N$_4$ windows typically 100 nm thick.

To identify particles of a special element, images taken at energies below and above the element specific absorption edge can be either compared or used to create an elemental distribution map by dividing the image taken above the absorption edge energy by the one taken below the absorption edge energy:

$$I_{\text{map}} = \ln \frac{I_{\text{above}}}{I_{\text{below}}} = \ln \frac{I_0e^{-\mu d}}{I_0e^{-\mu d}}$$

Of particular interest is to know the spatial arrangement within a sample environment just by distances or lengths. Tomography reveals the three-dimensional structure and thus can be used to deliver such information. However, many pictures are needed and due to inevitable deterioration of radiation sensitive samples by radiation damage, cryo-fixation is mandatory. Instead, stereo imaging needs only two images to reveal distances between particles or lengths within a sample. For this, the aqueous environment can be sustained.
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It follows: under ambient temperatures. Especially for the investigation of the dynamics of colloidal structures it is necessary to keep the system aqueous. Furthermore, within an aqueous sample system, sample movements occur. The probability to loose the examined sample during a stereo experiment is much smaller than during the acquisition of a complete tomographic data set. Thus, to reveal structural changes within a colloidal sample is much smaller than during the acquisition of a complete tomographic system, sample movements occur. The probability to loose the examined sample during a stereo experiment is much smaller than during the acquisition of a complete tomographic data set. Thus, to reveal structural changes within a colloidal system, sample movements occur. The probability to lose the examined sample during a stereo experiment is much smaller than during the acquisition of a complete tomographic data set. 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Data Language) to mark or select prominent structures in a set of tilted X-ray images and to get information about the three-dimensional configuration, distances and lengths (Gleber et al., 2003). The alignment of the stereo images necessary for determination of the common rotation axis is now implemented in XSTEREO. It is much easier as for a complete tomographic data set due to the fact that stereo processing evaluates only relative distances. Matching points recognisable in both projections are marked manually. An additional feature of the programme allows for connecting these points with lines. The spatial coordinates of each pair of matching points are calculated and the points or lines are presented in a three-dimensional plot. By free hand drawing, structures like edges can also be marked in both images and are then reconstructed point by point. In the three-dimensional plot, features like curvatures are displayed. The distances of all chosen structures are given in user-defined units. The error can be estimated based on the resolution of the micrographs, as the markers of matching points can be set with the accuracy of one pixel.

As tiltable holders for experiments with aqueous samples thin-pulled glass capillaries can be used, which are already common for tomography (Lehr, 1997; Schneider et al., 2002; Larabell & Le Gros, 2004). For these studies, borosilicate glass capillaries (Hilgenberg) of 1-mm outer diameter and 0.1-mm wall thickness were used. For easier filling, the capillaries have a 0.1-mm thick glass filament attached to the inner wall, improving the capillary forces when filled with a liquid. Capillary tips down to diameters of 1 µm and lengths short enough to stay stable without movements in the microscope have been produced with a pipette puller (H. Saur). The thick base of the capillary is mounted into a steel tube, which is fixed to the actual object holder of the X-ray microscope (Fig. 4 left). A cog wheel attached to the end of the steel tube allows for rotation of the capillary along an axis perpendicular to the optical axis either manually or using a motor. The air gap is about 200 µm wide, ensuring the tilt of the capillary over the whole range of 180° without hitting a vacuum window even when the rotation axis is slightly mismatched. The X-ray transmission of a capillary with a diameter of 2 µm, which has subsequently a wall thickness of 0.2 µm, filled with water, can be calculated to be 7.5% at E = 700 eV.

The full tilt range cannot be covered with an extended sample holder, which is, however, advantageous for studying extended objects. Small tilting angles are sufficient for stereo imaging, so it is possible to place an aqueous sample into the X-ray microscope in a tiltable flat holder. Therefore, two Si₃N₄ membranes (Silson Ltd., Blisworth, UK) of 100-nm thickness each, have been used. The dimensions of the Si₃N₄ membranes are comparable to these of the Si₃N₄ membranes acting as vacuum windows, thus limiting the usable tilt range. With the sample in between, a pair of membranes is glued across a 1 mm hole onto a 100-µm thin aluminium shim, which is mounted to the microscope in a way similar to the capillary mounting (Fig. 4 right). The maximum tilt range between the two vacuum windows of the microscope is approximately 14°. This is sufficient to obtain images for stereo analysis.

To avoid drying of the samples during the experiments, holders of both types have to be sealed. The tips of the capillaries were carefully dipped into a drop of silicone glue after filling. For the studies of extended objects, a drop of the aqueous sample was placed onto one of the Si₃N₄ membranes and then covered with the other one. Due to capillary forces, the water layer between the two membranes adjusts to the micrometre range. Using a pair of Si₃N₄ membranes enclosing a water layer of 2 µm thickness yields a calculated X-ray transmission at E = 700 eV of 8.5%. The pair of membranes has been sealed with glue around the edge of the wafers and subsequently fixed onto the aluminium shim. It has been tested that the glue acts as a water seal keeping the sample in an aqueous state for several hours, thus providing enough time for the experiments presented here.

The samples for these experiments have been limited to two iron oxides and two clay minerals, all very abundant in the environment, and a well characterized soil. The iron oxides were haematite and goethite (Sigma Aldrich, St Louis, MI, USA. CAS number 20344–49-4), montmorillonite and kaolinite were chosen as clay samples, and phaeozem as a soil (Ahl et al., 1985). All samples have been used as 1% (w/w) dispersion in deionized water. As can be seen in the experiments, large clusters of colloids disaggregate to small clusters with an intact nano- and microstructure. These small clusters have dimensions in the size range accessible with X-ray microscopy. Haematite particles with approximately 150 nm diameter have been made according to Schwertmann & Cornell (2000) and supplied as well as montmorillonite and kaolinite by J. Niemeyer, Institute for Soil Science, University of Göttingen, Göttingen, Germany.

Results

Figure 5 shows clusters of kaolinite platelets within one sample mixed with haematite between two Si₃N₄ membranes. The images in the top row were again taken at 704 eV and the
Fig. 5. Aqueous sample kaolinite platelets with haematite colloids between two Si$_3$N$_4$ membranes; images taken at $E = 704$ eV (top row) and $E = 710$ eV (bottom row). The scale bar in the top left corner indicates 500 nm.

ones in the bottom row at 710 eV, exposure times were 9.5 s with 2048 $\times$ 2048 pxl$^2$. When comparing both images, the ferruginous haematite particles can be identified clearly in the right image, so it is not necessary to create elemental distribution maps dividing the images. It is noticeable that the haematite particles, which are easily mobile within the sample holder system due to their small size relative to the water-layer thickness, are located at the edges of the clay platelets. This is an indication for an electrostatic interaction of the positively charged haematite particles with the negatively charged edges of the clay platelets.

To show that these kind of interactions happen not only in the model system clay – iron-oxide, phaeozem has been chosen as an example of a naturally occurring soil. Figure 6 shows a X-ray micrograph of an aqueous phaeozem sample where haematite has been added. As extended clusters of soil colloids were expected, the flat holder has been used for this experiment. The left image was taken at $E = 704$ eV below the iron K-absorption edge, the right one shows the same region but taken at the absorption maximum at 710 eV. Exposure times for both images were 7.2 s with 2048 $\times$ 2048 pxl$^2$. The energies have been chosen according to peak position of L-3 edge of haematite (Garvie & Buseck, 1998). The pair of images shows a part of a cluster with a large particle in the image centre, probably a clay particle. The haematite particles are located just at the edge of the large clay platelet, indicating electrostatic interaction as in the case of the kaolinite sample discussed previously.

In the left and centre image of Fig. 7, a montmorillonite sample mixed with haematite between two Si$_3$N$_4$ membranes and imaged at 700 eV and 707 eV can be seen. Exposure times were 0.45 s with 1024 $\times$ 1024 pxl$^2$. Although due to a slight mismatch in energy the contrast is not optimum, the haematite particles can be seen clearly in the centre image when comparing it with the left image. The discrimination between soil and haematite particles is supported and complemented by an elemental distribution map presented in the right image of Fig. 7. Here, haematite particles appear clearly as dark dots and are revealed also situated in position overlapping with position of the soil particles in the projection image. This sample was then tilted 14° around a horizontal axis and imaged again. Figure 8 shows details of the sample shown in Fig. 7, one selected from the centre image of Fig. 7, the other one selected from the image of the same region taken at $E = 707$ eV after tilting the sample. For stereo analysis, haematite particles were identified and marked as single dots in different colours, whereas nearby edges of clay platelets were marked as lines in magenta (Fig. 8), and then processed with XSTEREO. The created plot of the three-dimensional arrangements of the marked structures is presented in Fig. 9. Three different viewing angles have been chosen to clarify proximity relations. Note, that the units of this plot are pixels, with one pixel corresponding to 9 nm. Distances between the structures were determined. So, it became clear that the haematite particles, whose centres are marked as structures no. 2 (dark green) and 3 (blue), mark haematite particles attached to the edges of clay platelets, which are marked partially as no. 8 (dark magenta) and 9 (light magenta), respectively. The distances have been determined to 60 nm between haematite particle no. 2 and clay platelet edge no. 8, and 80 nm between haematite particle no. 9 and clay platelet edge no. 9. Taking the radii of the haematite particles and the lateral resolution of the X-ray microscope into account, this corresponds to a direct attachment. Haematite particle no. 1 (red) is not as close to the clay edge no. 7 (light magenta) as it seems in the left image of Fig. 8, but has a distance of 180 nm. The minimum distance between the centre of the haematite particle no. 4 (orange)
Fig. 7. Aqueous sample of montmorillonite particles with haematite colloids between two Si$_3$N$_4$ membranes; images taken at $E = 700$ eV (left) and $E = 707$ eV (middle). Right: Elemental distribution map where haematite particles appear as dark dots. The scale bar indicates 1 µm.

Fig. 8. Detail of the montmorillonite sample shown in Fig. 7 viewed with an angular difference between the images of 14° around a horizontal tilt axis, taken at $E = 707$ eV. For analysis, spherical particles are marked with dots and edges are marked with lines (please confirm text for details). The scale bar indicates 500 nm.

Also capillaries were filled and mounted to the X-ray microscope. Figure 10 shows two images of a capillary tip filled with kaolinite taken at $E = 707$ eV with a 14° tilt angle between them. Exposure times were 0.32 s with $1024 \times 1024$ px$^2$. The tip was sealed by dipping it into a drop of silicone glue to prevent drying. Unlike Fig. 6 where bigger kaolinite platelets lay parallel to the Si$_3$N$_4$ membranes, the structures inside the capillary seem to be more needle-like because only smaller platelets could reach the tip of 1.5 µm diameter. These small platelets show a good absorption contrast only when oriented parallel to the X-ray beam. The platelets oriented perpendicular to the light absorb the X-radiation so weakly that they are not visible. So, it became impossible in this case to identify the same edges in both images of the tilted pair.

Due to the lack of absorption contrast for small clay particles in a capillary tip sufficiently thin for transmission at the iron edge, goethite, another iron oxide abundant in the environment, has been chosen to show demonstrate the potential of this method to study dynamics. Small, elongated goethite particles access the thin capillary tip and provide a notable absorption contrast above the iron absorption edge. The upper image of Fig. 11 shows a cluster of particles of this goethite dispersion in a capillary of approximately 2.5 µm diameter, taken at 710 eV with an exposure time of 0.85 s with $1024 \times 1024$ px$^2$. In the subsequent images, micrographs are shown taken of the same region as above and under the same imaging conditions, but after haematite particles had been added to the capillary to induce changes. The tilt angle between these two images is 44°. When comparing the top image and the bottom image on the left side of Fig. 11, additional particles within the cluster are visible after the addition of haematite. As these particles are of different shape and smaller than goethite particles, they can be identified as haematite. Slight morphological changes of goethite clusters are also detectable, which are due to dynamics within the
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Fig. 9. Plot showing the points and edges marked in Fig. 8 presented under three different viewing angles. The unit of the axis is pixels. In the used images, 10 pixels relate to 90 nm.

Fig. 10. Small particles of a kaolinite dispersion in a capillary tip with approximately 1.5 µm diameter, taken at $E = 707$ eV. The tilt angle between the two images is $14^\circ$. The scale bar is 1 µm.

dispersion. The two images presented subsequently were taken 1 h after the addition of haematite.

In Fig. 12, details from the micrographs shown in subsequent Fig. 11 are presented, where some edges and centres of the goethite and haematite particles have been marked. A total of 8 structures have been marked and evaluated for their respective distances to depict the spatial arrangement of the cluster. The right images in Fig. 12 are taken under two different viewing angles from a three-dimensional plot of the marked structures, visualizing the distances between the clusters and the particles. The points no. 1 (red) and no. 2 (red) represent two corners of a goethite particle, so the connection (red line) between both points marks the edge. When comparing these images with the top image in Fig. 11 taken before the addition of haematite, and assuming that the association of the relatively large goethite particles is spatially stable, it becomes clear that only the small particles marked as no. 7 and no. 8 are haematite particles. To underline the potential of the presented technique, distances have been determined, for example the distance between the haematite particles no. 8 and the edge of the goethite particle mentioned above is approximately 250 nm.

Four systems have been studied, kaolinite, montmorillonite, goethite and the soil phaeozem. It is known that clay minerals show a negative surface charge in combination with positively charged edges at low pH values. When increasing this value, the edge charges change to negative as well. For kaolinite, the point of zero charge, that is the transition from positive to negative surface charges, lies around $pH \approx 2.7–4.1$ (Appel et al., 2003). The haematite dispersion used here was measured to $pH \approx 3–4$. The point of zero charge for haematite is around $pH \approx 8$ (Cornell & Schwertmann, 2003), resulting in positively charged particles added to the kaolinite dispersion. As the kaolinite has been dispersed with deionised water at neutral pH, and only little quantities of haematite have been added in order to not change the pH drastically, the pH of the mixture is approximately 5. Thus, the edge charge of the kaolinite is negative, resulting in an electrostatic attraction of the haematite particles to these edges. This can be seen in Fig. 5.

The point of zero charge of goethite lies around a pH value of 9 (Cornell & Schwertmann, 2003). As the dispersion of
of haematite particles around the edges of the soil particles visible in Fig. 6 supports this expectation.

The montmorillonite dispersion has a pH of approximately 6, resulting in a pH of the mixture measured to pH $\approx 5$. Haematite is positively charged at this pH value, however, for montmorillonite, the point of zero charge varies widely between pH $\approx 4–6$ (Lagaly, 1993; Sposito, 1998), dependent among others on its origin. In the right image of Fig. 7 it becomes clear that the haematite particles arrange themselves at the edges of the montmorillonite cluster. Therefore, it can be assumed that the edge charge of the montmorillonite particles is slightly negative, leading to a point of zero charge below 5. When looking at the subcluster depicted in Fig. 8 and using XSTEREO, it becomes clear that two of the haematite particles seen there are very close to the clay platelets, and three particles are attached to one of the support membranes. As the resulting pH value is close to the point of zero charge, the electrostatic forces are very weak. This has been sufficient during sample preparation in a test tube to attract all haematite particles to the clay clusters, however, in the vicinity of the support membranes, probably charged as well, the weak interaction is not strong enough to prevent the haematite particles from switching places.

**Conclusion**

The results of the project presented here can be separated into three parts, instrumental, methodical and soils scientific.

Instrumentally, the presented results show the potential of the X-ray microscope XM-1 for spectromicroscopy experiments of aqueous soil samples at the Fe-L edge around 707 eV. The mechanical precision of this instrument is well matched to the demands of stereo microscopy. The insertion and the use of capillaries or tiltable flat holders was uncomplicated, both could be quickly accessed and tilted due to the air gap.

The usage of Si$_3$N$_4$ membranes and capillaries as holders for aqueous soil samples to be transmitted with X-rays around the iron L-edge could be compared. Generally, for stereo images both membranes and capillaries were suitable as holders for aqueous samples and stayed stable over several hours. Between two Si$_3$N$_4$ membranes, large soil particles could be imaged, which could not access the small capillary tip. The extension of the soil particles along the optical axis made sure that the membranes could not collapse, so there was throughout the experiment a water layer of several micrometre thickness, and the arrangement of the smaller structures like the haematite particles was still undisturbed. However, large particles were aligned with the membranes. This was not the case in the capillaries, which do not force the sample particles at all. The capillary forms a sample holder tiltable over a large angle range with the possibility to manipulate the examined sample during the experiment by addition of other aqueous samples.
fundamental drawback is, however, that capillaries are not accessible for large particles. However, when using energies below the K-absorption edge of oxygen, thus within the water-window, capillary diameters of up to 10 µm can be used, accessible for larger particles. Furthermore, the higher mobility of particles within the capillary tip results in an higher probability to lose particles out of the imaging area even during the acquisition of just a pair of stereo images. As focusing demands the acquisition of usually two or three images for each focused one, and no changes were visible when comparing these images, for the samples of the here presented micrographs changes due to radiation damage or movements on the detectable length scale can be excluded.

With these features, it was possible to perform stereo experiments and demonstrate the methodically feasible outcome. Stereo images of a selected sample region were taken at different energies, so that elemental and spatial information could be combined. Iron containing particles could be identified non-ambiguously, for example haematite particles within aqueous colloidal soil samples. Distances between single particles or platelets within the three-dimensional sample association could be determined using XSTEREO. Positions and structures of points and platelet edges could be calculated and visualised in a three-dimensional plot. The diffusion of haematite particles into an already existing cluster of goethite particles is an example for the ability of X-ray microscopy to study the dynamics within an aqueous colloidal system. When the morphology of the particles differs, the dynamics can be followed even without elemental contrast.

With respect to soil science, the electrostatic interaction of colloidal soil particles with either opposite or equal charges could be investigated directly in aqueous media. The surface charge of soil particles changes with the pH of the dispersion. When oppositely charged, it causes electrostatic attraction, for example between clay particles and iron containing particles. These electrostatic forces are weak, and the underlying potential decreases by 1/r where r is the distance (Grasso et al., 2002; Vormoor, 2002). Therefore, particles have to be in close vicinity to each other to sense the electric field. The result of the electrostatic attraction was detected by identifying haematite particles close to edges of clay platelets. Thus, the consequences of different edge charges at certain pH values could be confirmed experimentally by direct imaging.

The results presented here open the path for future experiments on the dynamical behaviour of such aqueous colloidal systems from the environment. The study of morphological changes in situ is possible with stereo soft X-ray microscopy.

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References


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