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Formation of hydroxyl radicals catalyzed by clay surfaces

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Abstract The formation of superoxide and hydroxyl radicals at the surface of smectite clays due to oxygen reduction is demonstrated by electron paramagnetic resonance spectroscopy. The yield of hydroxyl radicals is mainly a function of particle size of the clays and depends, to a lesser extent, on the clay lattice iron. Synthetic laponite clay with small platelet size (~20 nm) and without lattice iron is leading in the formation of hydroxyl radicals followed by montmorillonite (~200 nm). Fluorohectorite (~2000 nm) was inactive to hydroxyl radical formation by oxygen reduction.

Keywords Smectite clays · Hydroxyl radical · Superoxide · Spin trapping · EPR

Introduction

Smectite clays are layered aluminosilicate materials which possess characteristic swelling, intercalation, and ion exchange properties (Pinnavaia 1983). Smectites occur in nature in fine crystalline form with grains predominantly < 2 μm and contain lattice iron in both oxidation states, in octahedral or tetrahedral geometry, and at different compositional levels. The most familiar and common member of the smectite group is montmorillonite, having Fe(III) and Fe(II) in its octahedral layer. In addition, synthetic smectite clays with different platelet size and without iron substitution in their lattice are available. Two such synthetic clays are laponite and fluorohectorite, which represent two extremes in particle size within the smectite group. In connection with the redox role of lattice iron in minerals, it has been reported that lattice iron in smectite clays can catalyze the decomposition of hydrogen peroxide (Oyama and Anson 1988). If iron centers participate in a Fenton-type reaction, i.e., $\mathrm{Fe(II)} + \mathrm{H_2O_2} \rightarrow \mathrm{Fe(III)} + \mathrm{HO^-} + \mathrm{HO^{\bullet}}$, hydroxyl radicals would be expected to form. This assumption prompted us to study whether smectite clays are active in the formation of $\mathrm{HO^{\bullet}}$ radicals by oxygen reduction and to determine the key factors of such activity.

It is well known that hydroxyl ($\mathrm{HO^{\bullet}}$) and superoxide ($\mathrm{O_2^{-\bullet}}$) radicals from the decomposition of hydrogen peroxide (Weitzman and Graceffa 1984) or from oxygen reduction (Zalma et al. 1987) in asbestos fibers have been implicated in chromosome damage and sequential carcinogenesis (Gulumian et al. 1983; Weitzman and Weitberg 1985; Mossman et al. 1986). Important factors for the carcinogenic potential of fibers is their size and durability (Lee et al. 1981; Becklake 1982). In addition, as iron is a prominent constituent in fibrous silicate minerals (Pooley 1981), it has been suggested that iron cations are responsible for the generation of reactive oxygen radicals according to the well-known Haber–Weiss cycle (Fridovich 1976; Gulumian et al. 1983; Weitzman and Graceffa 1984; Fubini and Arean 1999; Fubini et al. 1999; Aust et al. 2000; Shen et al. 2000). Support for this mechanism is that antioxidant enzymes and iron chelators suppress the deleterious reactions and therefore render the fibers less biologically active (Fridovich 1976; Rosen and Klebanoff 1981).

In this work, three representative members of the smectite group, with different dimensions and iron contents, were used to study the effect of platelet size and lattice iron for the activation of oxygen.

Experimental section

The natural smectite was a dioctahedral montmorillonite (SWy-1) supplied by the Clay Minerals Society. The synthetic clays were laponite, and fluorohectorite, both trioctahedral hectorites obtained from Laporte Industries (first) Ltd. and Corning Inc. (second). All samples were fractionated to < 2 μm by gravity

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Table 1 Some physical properties and the structural formula of laponite, montmorillonite and fluorohectorite

Clay	Particle size (nm)	CEC[a] (m Eq 100 g^{-1})	BET surface area (m^2 g^{-1})	Charge density (e^{-1}unit cell^{-1})[b]	Structural formula
Laponite	20	48	300	0.4	$Na_{0.8}[Mg_{5.4}Li_{0.4}]Si_8O_{20}(OH)_4$
Montmorillonite	200	90	80	0.6	$Na_{0.62}[Al_{3.28}Fe(III)_{0.30}Fe(II)_{0.04}Mg_{0.38}](Si_{7.8}Al_{0.2})O_{20}(OH)_4$
Fluorohectorite	2000	122	3	1.2	$Na_{1.12}[Mg_{4.88}Li_{1.12}]Si_8O_{20}F_4$

[a] CEC is the cation exchange capacity

[b] Unit cell is the Si_8O_{20} unit

sedimentation, purified by standard methods, and finally saturated with sodium cations. Some physical properties and chemical formulas are summarized in Table 1 (Kaviratna et al. 1996).

As spin trapping agent, the 5,5′-dimethyl-l-pyrroline-N-oxide compound (DMPO) from Aldrich was used. To produce adducts with hydroxyl radicals, a 1% suspension of the clay in aqueous buffer solution (potassium phosphate M/5 and NaOH M/5, pH = 7) were treated with 1 ml from a 0.1-M DMPO solution. Simultaneously, 1 ml of 1 M sodium formate solution was added as a probe anion ($HCOO^-$) to form the DMPO–$COO^{-\cdot}$ adduct[1] which is sufficiently stable and gives pronounced EPR signals (Zalma et al. 1987). The suspension was stirred for 30 min, centrifuged and filtered through 0.65-μm porosity filters. Aliquots from the three samples were taken and the detection of radical adducts was performed by EPR spectrometry. In addition, a reference sample without clay mineral was used for comparison.

The electron paramagnetic resonance (EPR) spectra were obtained using a Bruker ER 200D–SRC spectrometer equipped with an Oxford ESR 9 cryostat, a Bruker 035M NMR gaussmeter, and an Anritsu MF76A microwave frequency counter. Spectra were recorded at room temperature and were collected on a PC interfaced to the spectrometer.

Results and discussion

In experiments designed to trap the very reactive and elusive hydroxyl radicals, probably generated by smectite surfaces, we have exploited the free radical trapping action of DMPO compound in conjunction with formate anions. In the course of the reaction, any produced hydroxyl radicals donate electrons to formate anions to yield the formate radicals ($COO^{-\cdot}$), which react then with DMPO molecules to form the stable DMPO–$COO^{-\cdot}$ free radical adduct. The EPR spectra of three different smectite clays treated with DMPO in presence of formate anion are shown in Fig. 1. The spectrum of sodium-montmorillonite Fig. 1B. and that of sodium-laponite Fig. 1C exhibit the characteristic resonance lines of the formate radical adduct of DMPO. The g value from both signals at $g = 2.00555$ and the hyperfine splitting constants $a_N = 15.65$ G and $a_H = 16.75$ G compare well with values reported before (Harbour et al. 1974; Harbour and Bolton 1978; Finkelstein et al. 1980; Weitzman and Graceffa 1984). The synthetic fluorohectorite shows no remarkable EPR signal, Fig. 1D. Key feature of the spectra is the different intensities of the resonance lines. If structural iron sites were solely the effective catalytic centers for the reduction of oxygen, we would expect the ferruginous SWy-1 clay to show the highest intensity. In contrast, the iron-free synthetic laponite shows the highest intensity, reflecting the major size effect of the clay platelets. In addition, the iron-free synthetic laponite and fluorohectorite have different platelet size: laponite has a mean diameter of 20 nm and fluorohectorite of 2000 nm. This clearly implies that the dominant factor, affecting the intensities of the EPR lines and therefore the amount of the active oxygen species, is the size of the clay particulates. In order to confirm the above conclusion, we also tried the same method in two low-iron content smectite samples, sodium-hectorite (SHCa-1) and sodium-montmorillonite (STx-1) having size fractions after Stoke sedimentation $d > 2$ μm, since these smectites exhibit platelet sizes greater than that of SWy-1 and similar to that of fluorohectorite. No remarkable EPR signals were observed for either sample.

An important question with respect to the generation of active oxygen species is the nature of corresponding active sites at the clay layers. Laponite, for example, in distilled water forms stable colloidal dispersions in the form of nanoparticulate crystals of diameter 20–30 nm and thickness within the range of 1–4 nm (Thompson and Butterworth 1992). The surfaces of such nanoparticulates possess a large number of either homolytically (A) or heterolytically (B) broken bonds as depicted in Fig. 2.

Charged surface states derived in this matter are highly reactive (Fubini and Arean 1999) and can act as potential catalysts to activate oxygen molecules through chemisorption or polarization, as suggested by Thompson and Moll (1973). Upon chemisorption, O_2 accepts one electron from smectite broken edges to form the superoxide anion radical, $O_2^{-\cdot}$. Similar formation of superoxide radicals has been proposed for O_2 adsorbed on η-Al_2O_3 (Khoobiar et al. 1968). The hydroxyl radical formation through oxygen reduction to superoxide is strongly supported by the fact that the EPR signals from sodium-laponite were observed only when oxygen from the atmosphere was present but there was no EPR signal observed for the same sample when nitrogen gas was present. The following reactions

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[1] The formate anion was used to take into account the possible artifact in the formation of DMPO–$OH^{\cdot}$ by an oxidative pathway without invention of $OH^{\cdot}$, and also to reduce the importance of reaction, $OH^{\cdot} \xrightarrow{+e^-+H^+} H_2O$, i.e., destruction of $OH^{\cdot}$ in contrast with active sites of the solid (Zalma et al. 1987).

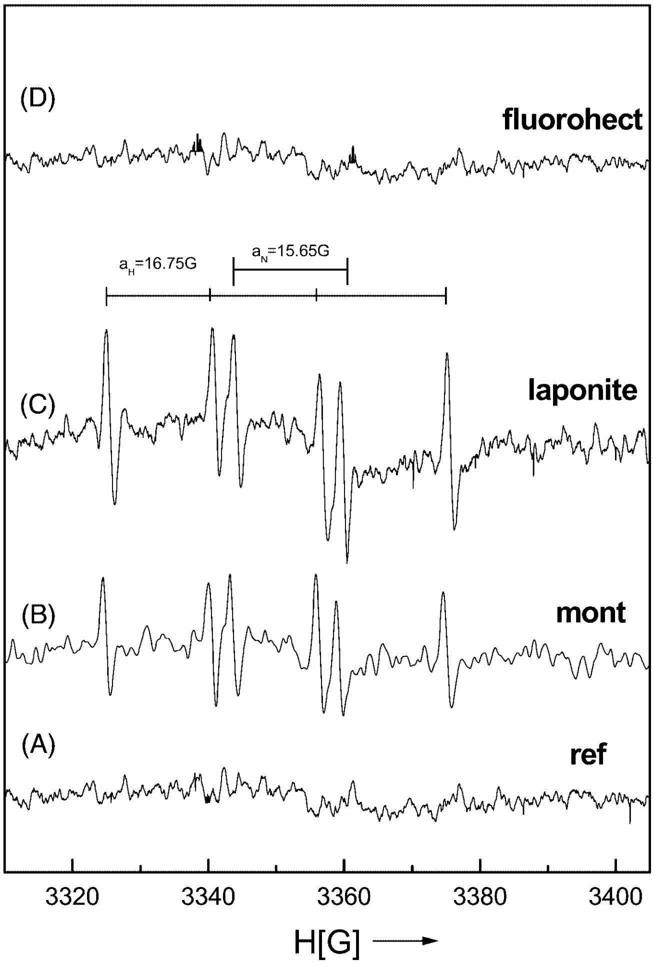


Fig. 1a–d EPR spectra of DMPO-radical adducts, after 30 min of incubation of minerals in buffer, with DMPO and formate solution. **a** Reference (without clay). **b** Sodium-montmorillonite. **c** Sodium-laponite. **d** Sodium-fluorohectorite. EPR conditions: modulation amplitude, MA = 1 Gpp, microwave power, MP = 10 mW and temperature, $T = 300$ K

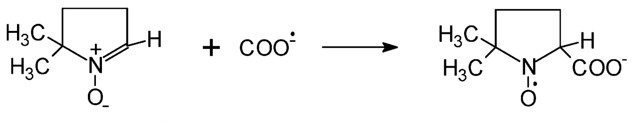
demonstrate the manner superoxide radicals ($O_2^{-\cdot}$) react to produce long-lived formate radicals (DMPO–$COO^{-\cdot}$) (Zalma et al. 1987):

$$O_2^{-\cdot} + e^- + 2H^+ \rightarrow H_2O_2 \tag{1}$$

$$H_2O_2 + e^- \rightarrow OH^\cdot + OH^- \tag{2}$$

$$HCOO^- + OH^\cdot \rightarrow COO^{-\cdot} + H_2O \tag{3}$$

(4)



As expected, the observed order of oxygen activation follows the respective order of mineral surface area (Table 1). Analogous correlations relating the oxidative power of smectites and other mineralogical samples with the surface area of the mineral have also been reported (Thompson and Moll 1973; Zalma et al. 1987).

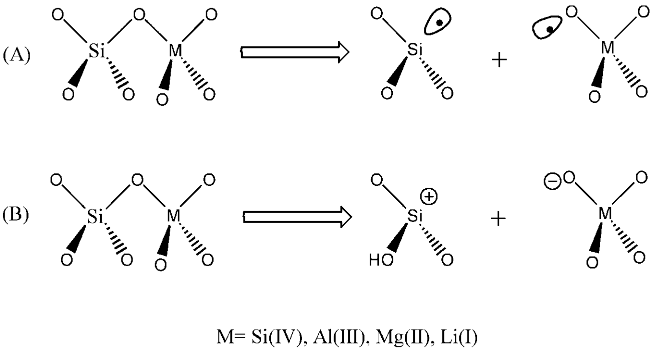


Fig. 2 Homolytically (**A**) or heterolytically (**B**) cleavage of Si–O–M bonds in laponite

Natural montmorillonite with platelet size of 200 nm can yield hydroxyl radicals not only by oxygen activation as laponite, but also via electron transfer from electron donor metal sites in the solid (Solomon 1968). Divalent iron, present in the octahedral sheet of SWy-1 montmorillonite, can donate electrons to oxygen, producing superoxide radicals:

$$\text{Structural} - Fe^{2+} + O_2 \rightarrow \text{structural} - Fe^{3+} + O_2^{-\cdot}\ . \tag{5}$$

However, in spite of this additional source of electrons, the intensity of the EPR signals of SWy-1 is lower than that of laponite. This result reflects the prime role of particle size of minerals in oxygen radical production. In summary, the present study demonstrates the role of minerals in fine dispersion to generate toxic hydroxyl and superoxide radicals from the reduction of molecular oxygen on their surfaces. The quantity of these reactive radicals depends primarily on particle size of the minerals and to a lesser extent on the clay lattice iron.

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