



Research paper

Influence of layer charge origin and layer charge density of smectites on their aflatoxin adsorption



Ana Luisa Barrientos-Velázquez^a, Alicia Marroquin Cardona^b, Lian Liu^a, Timothy Phillips^c, Youjun Deng^{a,*}

^a Department of Soil & Crop Sciences, Texas A&M University, College Station, TX, United States

^b Toxicology Department, Faculty of Veterinary Medicine, Universidad Autonoma de Nuevo Leon, Monterrey, Nuevo Leon, Mexico

^c Veterinary Medicine and Biomedical Sciences, Texas A&M University, College Station, TX, United States

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ABSTRACT

The aflatoxin adsorption efficiency of a bentonite depends strongly on the physical, chemical, and mineralogical properties of the smectite, the dominant clay mineral in bentonites. The objective of this study was to determine the effects of the layer charge origin, octahedral cation composition, and layer charge density on the smectites' selectivity and adsorption capacity for aflatoxin.

Six smectite samples with different layer charge densities and octahedral cation compositions were evaluated: a montmorillonite (4TX) and a hectorite, which represent smectites with octahedral charge origin; and a beidellite, two saponites from Australia and Spain, and a nontronite, which represent smectites with tetrahedral charge origin. The clay fraction of the smectites was saturated with Na, Ca, and Ba, respectively. Aflatoxin adsorption isotherms were conducted for these smectites. The CEC of one high-charge montmorillonite (50K) and the nontronite were reduced by Li-saturation and heating.

Similar effect of the exchangeable cations was observed in all smectites, but significant differences in the aflatoxin adsorption capacity were observed among the smectites. The octahedrally charged smectites showed higher aflatoxin adsorption than the tetrahedrally charged smectites. The aflatoxin-smectite bonding mechanism was not affected by the layer charge origin or density. Optimal layer charge density was required to have sufficient non-polar sites on the smectite for aflatoxin molecules. High-charge-density smectites had low adsorption capacity and binding affinity for aflatoxin, but aflatoxin adsorption on these smectites can be improved by reducing the layer charge density of the clay mineral. Smectites with octahedral charge increased the interlayer accessibility of aflatoxin molecules. The octahedral cation composition had a negligible effect on the aflatoxin adsorption capacity of smectites.

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1. Introduction

A fatal disease that caused the death of about 100,000 turkeys in England in 1960 known as Turkey X disease was reported along with other mortality cases in dogs, guinea pigs, swine, and cattle (Heathcote and Hibbert, 1978). It was soon recognized that ingredients used in the animal feed such as corn, peanut, and other grains and nuts were contaminated with fungi *Aspergillus flavus*. A toxic organic compound was extracted from the contaminated peanut meal and was designated as aflatoxin B₁ as it fluoresced blue when irradiated with UV light. Fungi *Aspergillus flavus* and *Aspergillus parasiticus* produce aflatoxins as secondary metabolites. The first identified and the most

common aflatoxins are aflatoxin B₁ (Afb₁) and aflatoxin G₁ (Afg₁, fluoresces green under UV irradiation). The Afb₁ is the most toxic and carcinogenic. The chemical structures of aflatoxins are composed of difurocoumarins with some having a cyclopentenone end such as in Afb₁ and others having a lactone ring such as in Afg₁ (Bbosa et al., 2013).

Incorporation of bentonites into aflatoxin-contaminated feed is a low cost measure that has shown (based on animal trials) the ability in reducing the bioavailability of these toxic and potent carcinogenic compounds. Bentonites could adsorb up to 20% (w/w) of aflatoxin B₁ (Afb₁) in aqueous solution (Kannewischer et al., 2006), but animal trials with these high binding bentonites have not shown a complete protection from aflatoxin toxicity. The use of bentonites or other materials as aflatoxin (or mycotoxin) binders in animal feed is still restricted. In 2011, the use of aflatoxin binders in animal products in the state of Texas, USA was approved by the Office of the Texas State Chemist. A license is required to sell a product as an aflatoxin binder.

Although screening procedures to select effective aflatoxin binders have been proposed (Grant and Phillips, 1998; Dixon et al., 2008), a

* Corresponding author.

E-mail addresses: anabarrientos@tamu.edu (A.L. Barrientos-Velázquez), aliciamarroquin@hotmail.com (A. Marroquin Cardona), angela.liulian@yahoo.com (L. Liu), tphillips@cvm.tamu.edu (T. Phillips), yjd@tamu.edu (Y. Deng).

correlation between a single mineral property and aflatoxin adsorption is difficult to establish (Vekiru et al., *in press*; Magnoli et al., 2008; Mulder et al., 2008; Tenorio Arvide et al., 2008). Tenorio Arvide et al. (2008) observed that the structural composition of the octahedral sheet in smectites affected their aflatoxin adsorption capacity: the smectites with certain amount of structural Fe and Mg octahedral substitutions had higher aflatoxin adsorption capacities than the smectites with less octahedral substitutions for Al. Mulder et al. (2008) investigated the effect of physical properties of bentonites such as particle size and morphology on aflatoxin adsorption. A linear correlation ($R^2 = 0.73$) between the $<2 \mu\text{m}$ fraction clay content and maximum AfB_1 adsorption (Q_{max}) was observed, but not all of the bentonite samples tested could fit the correlation as they shown poor dispersion and high aflatoxin adsorption capacity. The morphological analysis indicated that the better adsorbents occurred as thin particles, while the moderate to poor adsorbents were observed as thicker particles. In a recent study, Vekiru et al. (*in press*) compared the *in vitro* and *in vivo* aflatoxin adsorption efficiency of seven dioctahedral smectite samples. The animal trials showed that the high *in vitro* binding samples reduced the aflatoxin toxicity. The differences in AfB_1 adsorption capacity were attributed to the structural configuration: the *cis*-vacant smectites samples showed higher AfB_1 binding than the *trans*-vacant sample. Several smectite properties affect the AfB_1 adsorption. The investigation of these properties and how they affect the adsorption of aflatoxins offers the scientific basis for the modification of the smectites to increase the their adsorption and selectivity for the toxin.

Smectite, the dominant clay mineral in bentonites, has an expandable layer structure. Adsorption of AfB_1 molecules has been shown to occur mainly in the interlayer of smectite (Phillips et al., 2002; Kannewischer et al., 2006). Based on the strong effect of the exchangeable cation on aflatoxin adsorption, it was proposed that the adsorption mechanism was an ion-dipole interaction between the carbonyl oxygen of the mycotoxin and the exchangeable cations under low humidity, and H-bonding between the carbonyl group and the hydrated ions (Deng et al., 2010). It has been demonstrated that the hydration energy of the exchange cations significantly influenced the amount of aflatoxin adsorbed (Deng et al., 2012). For instance, smectite saturated with divalent cations with low hydration energies, such as Ba^{2+} , showed higher affinity and adsorption capacities for aflatoxins.

High-charge density smectites had shown low adsorption and poor affinity for AfB_1 (Jaynes and Zartman, 2011; Deng et al., 2012). Jaynes and Zartman (2011) found that the aflatoxin adsorption efficiency of the smectites was inversely related to the layer charge density. Furthermore, the aflatoxin adsorption of the high charge density smectite was improved by decreasing the layer charge with the incorporation of Li^+ into the vacant octahedral site. Deng et al. (2012) also reported that smectites with high CEC (CEC = 130 cmol/kg) were not as effective as the smectites with lower CEC values (CEC 78–94 cmol/kg) in adsorbing AfB_1 . It was proposed that the lower charge density of smectites had more non-polar sites on the mineral's surface for AfB_1 molecules adsorption.

Montmorillonite is one of the most common smectites and has been used in many batch equilibrium experiments and animal trials. The other members of the smectite group minerals such as beidellite, hectorite, nontronite, and saponite have similar 2:1 expandable interlayer structure but different layer charge origins and octahedral cation compositions. The negative layer charge in both montmorillonite and hectorite originates from the substitutions in the octahedral sheet, while in saponite, beidellite, and nontronite the charge is located in the tetrahedral sheets. In montmorillonite, the Al^{3+} dominates the octahedral position and the charge deficiency is generated by the isomorphic substitutions of Mg^{2+} for Al^{3+} . On the other hand, Mg^{2+} prevails in hectorite with Li^+ being the replacement ion. Beidellite, nontronite and saponite have Al, Fe, and Mg as the dominant octahedral structural ions, respectively. In beidellite, nontronite, and saponite, the charge originates from the tetrahedral sheet by the isomorphic substitutions of Al^{3+} for Si^{4+} .

Limited studies have reported the use of beidellite, saponite, hectorite, and nontronite as aflatoxin binders. Beidellite and saponite, however had been used as adsorbents of no-ionic organic compounds such as atrazine and showed high adsorption efficiency (Aggarwal et al., 2006).

The effect of octahedral structural cations and the layer charge origin in smectite on aflatoxin adsorption has not been systematically addressed. Hence, the objective of this study was to determine the effects of the layer charge origin, octahedral cation composition and layer charge density of the smectites on their selectivity and adsorption capacity for aflatoxin.

2. Materials and methods

2.1. Mineral and chemical characterization of smectites

Six smectite samples with different octahedral cation compositions and layer charge origins were selected. Beidellite and hectorite samples were obtained from the Clay Minerals Society repository (SBCa-1 and SHCa-1, respectively). Nontronite was a sample taken near Spokane, Washington. The montmorillonite sample (4TX) was collected from Gonzales, TX. Two saponite samples from Toledo, Spain (SS), and the other from Western Australia (SA) were used. A high charge montmorillonite from Oklahoma (5OK), reported in Deng et al. (2012), was used for the charge reduction experiment. The samples were fractionated to collect the $<2 \mu\text{m}$ clay particles (Deng et al., 2009).

Approximately 60 mg of each smectite sample were saturated with Mg^{2+} and K^+ to facilitate the identification of phyllosilicates. The saturated clay mineral dispersions were air dried on glass discs. The X-ray diffraction (XRD) patterns of Mg-saturated clay minerals were recorded at room humidity and after glycerol solvation. The K-saturated samples were recorded at room temperature, and after 330 °C and 550 °C heating. XRD patterns were recorded from 3 to 32 degrees two-theta using a Bruker D8 ADVANCE diffractometer with $\text{CuK}\alpha$ radiation, 30 rpm spin rate, and 0.017 step size. A1-D position sensitive detector LynxEye was used during XRD analyses.

The octahedral cation composition of each smectite was determined using Fourier transform infrared (FTIR) spectroscopy. Ten milligrams of dry clay fraction of each smectite was mixed with 300 mg of KBr using a Wig-L-Bug mixer. The FTIR spectra were recorded using diffuse reflectance infrared (DRIFT) accessory on a Spectrum 100 Perkin Elmer FTIR spectrometer with an average of 32 scans and a resolution of 2 cm^{-1} .

Cation exchange capacity (CEC) was determined by saturating 100 mg of the clay fraction with Ca^{2+} then replacing the adsorbed Ca with Mg^{2+} . Atomic adsorption was used to quantify the Ca^{2+} concentration.

2.2. Layer charge reduction of high charge density smectites

To test the effect of the layer charge density on the aflatoxin adsorption, one high-charge montmorillonite from Oklahoma (5OK with 0.49 charge/half unit cell) and the nontronite were subjected to a lithium saturation/heating treatment. A reduction in the layer charge of smectites by incorporating Li^+ into vacant octahedral sites after heating the sample at high temperatures is known as the Hofmann and Klemen effect (Lim and Jackson, 1986; Jaynes and Bigham, 1987). The clay fractions of both samples were treated with mixed LiCl/NaCl solutions that had different Li/Na molar ratios. Fifty mg of smectite sample was mixed with the corresponding Li:Na solution three times and with DI water twice. The samples were placed in aluminum dishes, dried at 100 °C, and then heated at 250 °C for 12 h. A1:1 water:ethanol solution was used to redisperse the samples. One control sample was prepared by using the 15% Li:Na solution without heating. After the charge reduction, the nontronite and montmorillonite 5OK samples were redispersed and saturated with 0.5 M BaCl_2 .

After saturating with Ba^{2+} solution the smectite samples were washed twice with 1:1 methanol:water. Twelve milligrams of the Ba-

saturated smectites were added to 50 mL centrifuge tubes, and digested using 2 mL aqua regia and 3 mL hydrofluoric acid (HF). Standard Li⁺ solutions were prepared using the same digestion solution. The samples and standards were placed on an orbital shaker at 200 mot/min overnight. Saturated boric acid was added to each tube up to the 25 mL mark to promote the formation of soluble fluoroborates of cations. The concentration of lithium in solution was quantified using flame emission on a Perkin Elmer AAnalyst 400 atomic adsorption spectrometer. The CEC values of the Li-treated smectites were obtained by subtracting the lithium content (cmol(+)/kg) from the original CEC value of untreated smectites.

2.3. Aflatoxin adsorption isotherms

Each smectite sample was saturated with Na, Ca, or Ba to test the effect of the exchange cation size and valence on the adsorption of aflatoxin. The samples were saturated three times with 1 M Na, 0.5 M Ca, or Ba chloride solutions, and then washed twice with DI water. Smectite dispersions were prepared to have a concentration of 2 mg/mL. The aflatoxin adsorption isotherm procedures were described in Grant and Phillips (1998) and Kannewischer et al. (2006). A 1000 ppm aflatoxin stock solution was prepared by dissolving 50 mg of aflatoxin B₁ in 50 mL of acetonitrile. A dilution was made to prepare an 8 ppm AfB₁ solution in DI water. The concentrations were varied as: 0.0, 0.4, 1.6, 3.2, 4.8, 6.4, and 8.0 ppm. Each isotherm point had a total volume of 5 mL. A volume of 50 μL of the smectite dispersion was used for each isotherm point. After overnight shaking at room T, the samples were centrifuged at 4500 rpm for 57 min and the aflatoxin concentration in the supernatant was quantified using a Beckman Coulter DU 800 UV-spectrophotometer at 362 nm wavelength. Aflatoxin adsorption isotherms were also performed for the charge-reduced Ba-montmorillonite and Ba-nontronite following the procedure described above. The aflatoxin B₁ adsorbed was plotted against the equilibrium concentration to fit the Langmuir model (LM). From the curve fitting, the maximum aflatoxin adsorption (Q_{max}) and the affinity constant (K_d) were obtained (Grant and Phillips, 1998). The smectite samples that had a poor fitting (low η²) were plotted with either the exponential Langmuir model (ELM) or the modified Langmuir model with q dependent affinity (QKLM) (Grant et al., 1998; Deng et al., 2012).

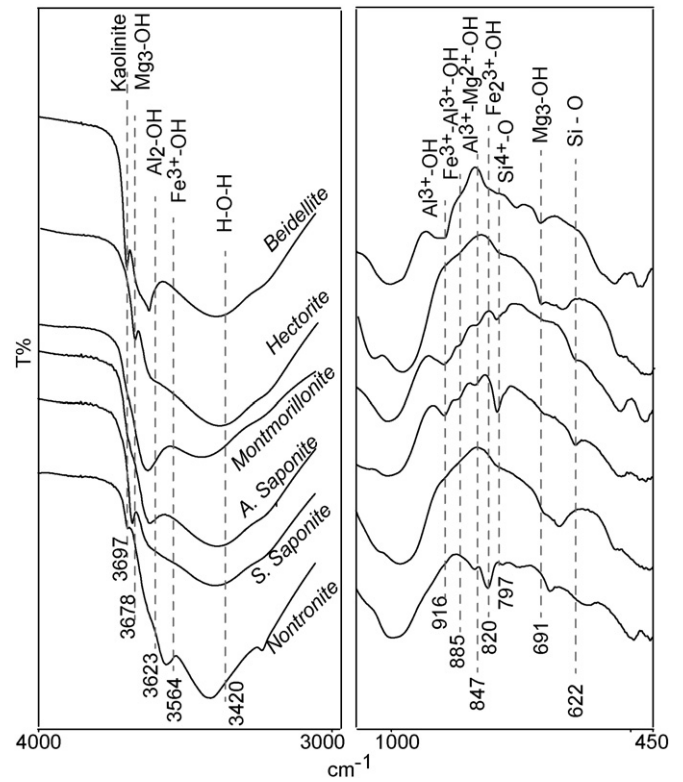


Fig. 2. FTIR spectra of clay mineral fractions. (%T: % of transmittance).

3. Results

3.1. Mineralogical composition of smectites

The fractionation of the bentonite samples yielded the following <2 μm clay contents for the samples: beidellite 76%, montmorillonite 88%, hectorite 95%, nontronite 73%, Spain saponite 72%, and Australia saponite 85%.

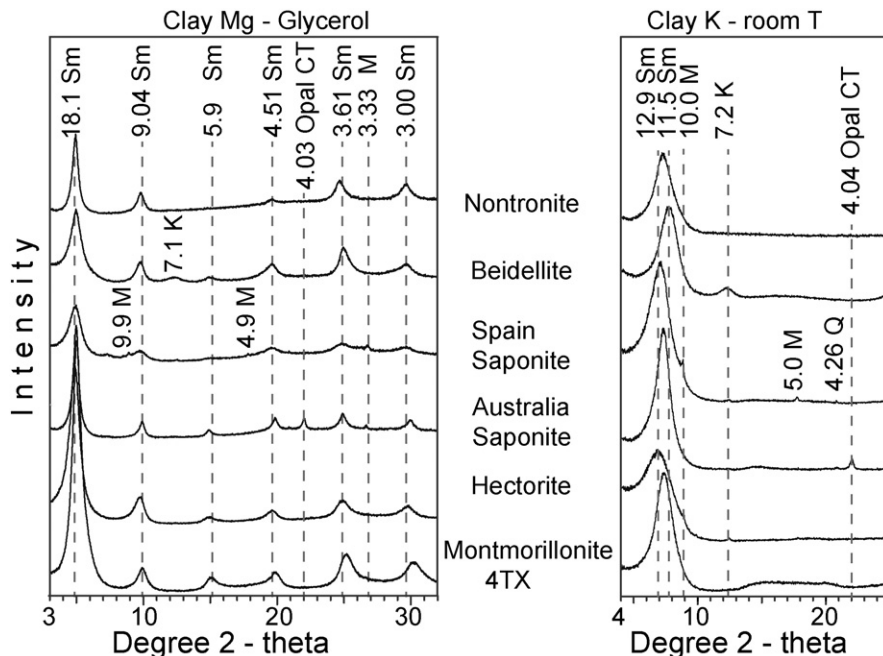


Fig. 1. XRD patterns of clay fractions Mg-glycerol solvation (left) and K-saturated clay fractions (right). (Sm: smectite, M: mica, K: kaolinite).

After Mg^{2+} saturation and glycerol solvation, all smectites showed an intense XRD peak with an expanded interlayer spacing of 18 Å, which indicated the dominance of smectite in all samples (Fig. 1). No other minerals were identified on the XRD patterns of the montmorillonite (4TX) or the nontronite samples but the FTIR spectrum of nontronite showed a weak 3699 cm^{-1} band of kaolinite (Fig. 2). The 7.1 Å kaolinite peak was present in beidellite and in the Spanish saponite. This peak was present in K-saturated hectorite sample only. The 10 Å and 3.33 Å peaks observed in Spanish saponite were due to presence of mica. The beidellite, hectorite, and the Australia saponite also contained mica that was only observed with TEM (data not shown). Additionally, the Australian saponite showed a peak at 4.04 Å , which was characteristic of opal-CT.

Distinct OH stretching vibration differences in $4000\text{ to }3000\text{ cm}^{-1}$ region (Fig. 2) were observed among the smectites. Beidellite and montmorillonite showed a distinctive band at 3623 cm^{-1} that was due to the dominance of Al^{3+} -OH in the octahedral sheet. The dominance of the octahedral Mg^{2+} in the hectorite and the Spanish saponite resulted in the band at 3678 cm^{-1} . This band was not present in the Australia saponite, instead a band at 3623 cm^{-1} was present, indicating a montmorillonite-like composition. Nontronite showed the distinctive 3564 cm^{-1} band due to octahedral Fe^{3+} -OH (Madejová, 2003). The

band at 3697 cm^{-1} confirmed the presence of kaolinite in beidellite and nontronite.

For beidellite, montmorillonite and the Australia saponite the $AlAl$ -OH bending vibrations occurred at 916 cm^{-1} , $Al^{3+}Fe^{3+}$ -OH bending at $\sim 885\text{ cm}^{-1}$, and $Al^{3+}Mg^{2+}$ -OH bending at $\sim 845\text{ cm}^{-1}$ (Gates, 2005). The major bands for hectorite and the Spanish saponite were observed at 691 cm^{-1} , which was attributed to the bending vibrations of Mg-OH. The distinctive bending vibrations at 798 and 622 cm^{-1} in Australia saponite were attributed to cristobalite (van der Marel and Beutelspacher, 1976).

3.2. Effect of interlayer cation on aflatoxin adsorption

The type of exchange cation strongly influenced the aflatoxin adsorption on all of the smectites. A similar trend was observed with all of the smectite samples: the lowest adsorption of aflatoxin was observed in the Na-saturated smectites with $Q_{max} < 0.3\text{ mol/kg}$ and $K_d < 0.1\text{ }\mu\text{M}^{-1}$, while the Ca- and Ba-exchanged smectites samples showed a higher adsorption with $Q_{max} > 0.3\text{ mol/kg}$ and K_d between $0.1\text{--}1.2\text{ }\mu\text{M}^{-1}$ (Fig. 3, Table 2). The intercalation of aflatoxin B₁ on the smectites was corroborated with the FTIR spectra. All the smectite samples showed the characteristic aflatoxin bands in the $1800\text{--}1200\text{ cm}^{-1}$

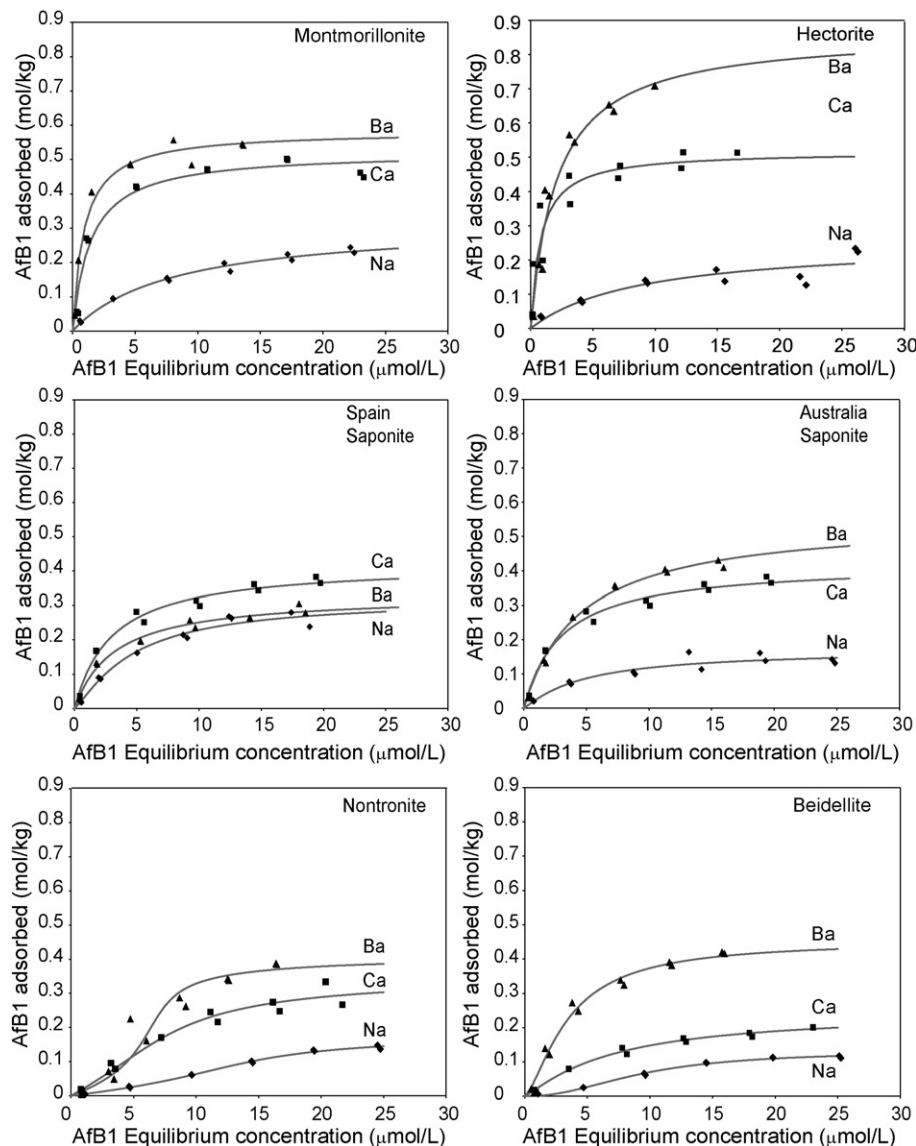


Fig. 3. Aflatoxin adsorption isotherms (room T) of smectites saturated with different cations.

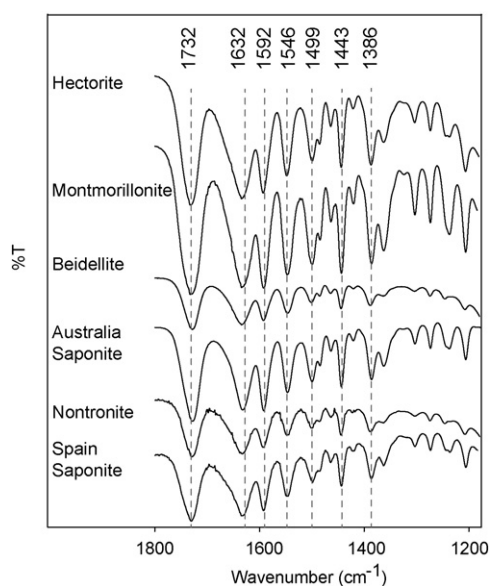


Fig. 4. FTIR spectra of AFB₁-smectite complexes showing the 1800 to 1200 cm⁻¹ region where the major AFB₁ bands occur. Data presented as % of transmittance (%T).

range. The strong 1732 cm⁻¹ band was attributed to the in-phase stretching vibration of the carbonyl groups in the aflatoxin B₁ molecule. The 1632 and 1592 cm⁻¹ bands were due to the out-of-phase stretching and bending vibrations, respectively (Deng et al., 2012). The FTIR spectra of the AFB₁-smectite complexes showed similar aflatoxin band positions for all samples (Fig. 4). The lack of major shifts on the IR bands indicated that the same aflatoxin adsorption mechanism occurred in all smectite samples.

3.3. Effect of layer charge origin on aflatoxin sequestration by smectite

Both montmorillonite and hectorite, which had octahedral charge, showed the greatest effect of the exchange cation (Fig. 3). Ba-saturated montmorillonite had the maximum adsorption of 0.5 mol/kg, and hectorite had the highest adsorption of 0.8 mol/kg. Beidellite and the two saponites had similar isotherm shapes and maximum adsorption capacity. Aflatoxin adsorption isotherms in nontronite did not fit the Langmuir model well due to the low affinity that produced an S-type shape isotherm (Table 2). In this case, the modified Langmuir equation (QKLM) described by Grant et al. (1998) and Deng et al. (2012) was used to calculate the adsorption maxima (Q_{max}) and the affinity parameter (K_d). Although the aflatoxin adsorption affinity of nontronite remained poor, the maximum adsorption improved with the divalent cation saturation.

Montmorillonite 4TX and the nontronite had similar CEC values, 103.6 and 107.9 cmol(+)/kg respectively (Table 1), yet it was observed

Table 1
Characterization of smectite samples.

Sample	Clay mineral content (g/kg)	Non smectite minerals ^a	CEC (cmol(+)/kg)	Estimated layer charge ^b	Octahedral composition	Charge origin
Beidellite	764	kaolinite	73.5	0.34	Al ³⁺	Tetrahedral
Hectorite	964	kaolinite	86.9	0.33	Mg ²⁺ , Li ⁺	Octahedral
Montmorillonite 4TX	876	none	103.6	0.38	Al ³⁺ , Mg ²⁺	Octahedral
Montmorillonite 5OK	767 ^c	none	137.0	0.50	Al ³⁺ , Mg ²⁺	Octahedral
Nontronite	734	kaolinite	107.9	0.45	Fe ³⁺	Tetrahedral
Australia Saponite	853	Mica, Opal CT	105.7	0.41	Mg ²⁺	Tetrahedral
Spain Saponite	721	Mica	77.5	0.30	Mg ²⁺	Tetrahedral

^a Non smectite minerals present in the clay fraction.

^b Estimated smectite layer charge (charge/half unit cell).

^c Data from Deng et al. (2012).

Table 2
Smectite AFB₁ adsorption isotherm fit parameters with different cation saturation.

Sample	Exchange cation	Langmuir			
		Q _{max} (mol/kg)	K _d (μM ⁻¹)	η ²	
Montmorillonite (4TX)	Ba	0.58	1.11	0.96	
	Ca	0.52	0.69	0.95	
	Na	0.31	0.12	0.99	
Hectorite	Ba	0.87	0.48	0.96	
	Ca	0.52	1.27	0.89	
Australia Saponite	Na	0.25	0.11	0.82	
	Ba	0.55	0.22	0.99	
Spain Saponite	Ca	0.42	0.33	0.98	
	Ba	0.33	0.31	0.98	
	Ca	Exponential Langmuir			
		Q _{max} (mol/kg)	K _d (μM ⁻¹)	N	η ²
Beidellite	Ba	0.45	0.16	1.47	0.98
	Ca	0.25	0.106	1.15	0.99
	Na	0.13	0.008	2.15	0.99
Australia Saponite	Na	0.17	0.18	1.10	0.90
Spain Saponite	Na	0.31	0.15	1.22	0.96
		Modified QKLM			
		Q _{max} (mol/kg)	K _d (μM ⁻¹)	b	η ²
Nontronite	Ba	0.44	0.034	-3.77	0.96
	Ca	0.37	0.057	-2.29	0.98
	Na	0.19	0.024	-6.15	0.99
Montmorillonite (5OK)	Ba	0.45	0.033	-3.59	0.99

that montmorillonite showed higher adsorption capacity (Q_{max} > 0.5 mol/kg for the Ca- and Ba-exchanged samples) while AFB₁ adsorption in nontronite ranged between Q_{max} 0.3–0.45 mol/kg for the Ca and Ba saturated samples, respectively. The major difference was the low affinity in nontronite (K_d of 0.034–0.057 μM⁻¹) in comparison to montmorillonite (K_d of 0.69–1.11 μM⁻¹). The difference in adsorption can be related to the layer charge origin. In montmorillonite, the negative charge arose from the octahedral substitutions while in nontronite, it arose from the tetrahedral sheets. A similar trend was observed when hectorite was compared with beidellite and saponites: the samples had similar CEC values (Table 1) but the highest aflatoxin adsorption was observed in the octahedrally charged hectorite.

3.4. Effect of layer charge density on aflatoxin adsorption

Montmorillonite 5OK, with high CEC (137 cmol(+)/kg), had an aflatoxin adsorption of 0.3 mol/kg and low affinity for AFB₁ (Figs. 5 and 6). As the CEC of this smectite was reduced by incorporating different amounts of Li in the octahedral sheet, the adsorption capacity increased. The highest aflatoxin adsorption (0.8 mol/kg) was observed when the CEC was reduced to 86 cmol(+)/kg. Further

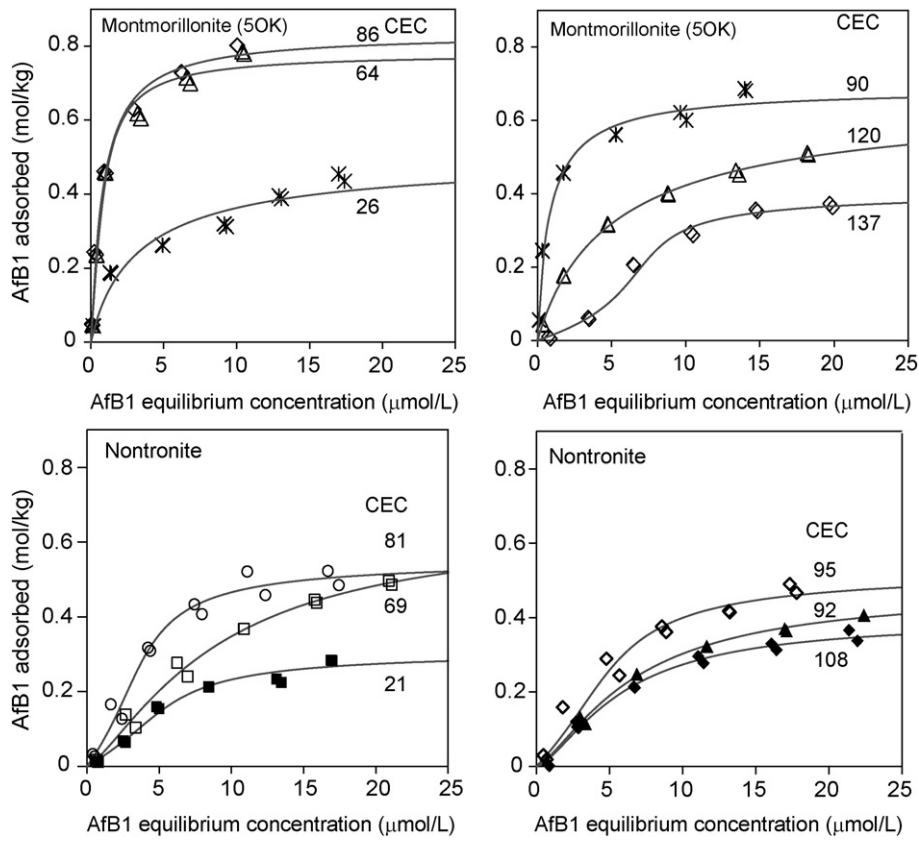


Fig. 5. Aflatoxin adsorption isotherms (room T) of charge-reduced montmorillonite 50K and nontronite. CEC data showed at the end of the plot.

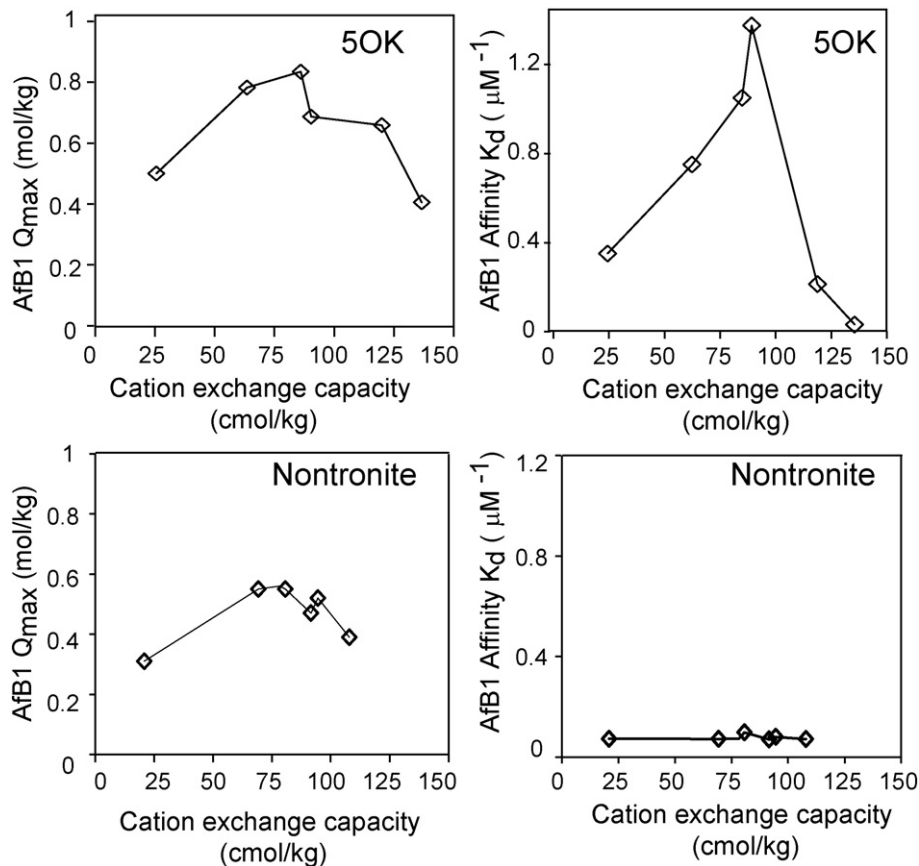


Fig. 6. Q_{max} and affinity (K_d) plots at different CEC values of the Li-treated montmorillonite 50K and nontronite.

reduction of the layer charge to 26 cmol(+)/kg caused the adsorption to decrease to 0.4 mol/kg. A similar trend was observed on nontronite. As the CEC was reduced, the adsorption capacity increased initially and reached a maximum at CEC of 81 cmol(+)/kg. Although there was an increase in the amount of aflatoxin adsorbed, the affinity of nontronite for aflatoxin was not improved in comparison to montmorillonite 50K (Fig. 6).

The effect of layer charge reduction of montmorillonite 50K on aflatoxin adsorption indicated that an optimal CEC of ~86 cmol/kg was required to reach the highest Q_{\max} of 0.8 mol/kg, when Ba was the exchangeable cation. Hectorite, a trioctahedral smectite with an octahedral charge origin, had a CEC of 87 cmol(+)/kg and showed the highest Q_{\max} of 0.87 mol/kg in the Ba-exchanged sample. Thus, Ba-montmorillonite and Ba-hectorite were able to adsorb similar amounts of aflatoxin at similar CEC values.

3.5. FTIR comparison of Li-treated montmorillonite and nontronite

The major structural changes on the high-charge dioctahedral smectite (50K) were observed at high Li saturation (85%) (Fig. 7). As the structural Li^+ increased, the intensity of the OH-stretching band at 3619 cm^{-1} reduced and a shoulder appeared at 3665 cm^{-1} . In the OH-bending region, the band due to the AlAlOH vibration at 914 cm^{-1} shifted to a higher wavenumber and the intensity of this band decreased. A shift to lower wavenumber was observed for the 622 cm^{-1} band. The band shifts were more evident as the Li^+ content was increased, indicating incorporation of Li in the octahedral sheet. On the contrary, no changes in the IR band position or intensities were observed for the nontronite samples for up to 50% of Li saturation. It is possible that the incorporated structural Li concentration was not high enough to produce significant shifts in the band positions of nontronite, but the Li quantification after nontronite digestion and the increase in aflatoxin adsorption indicated migration of Li to the smectite layers. Madejová et al. (2000) studied the perturbation of the infra-red bands of smectites by the incorporation of Li in their structures. A ferruginous smectite with a tetrahedral charge origin showed minor shifts on the Si–O bands, which suggested that Li^+ ion migrates to the hexagonal cavity formed by the tetrahedral sheets (Komadel, 2003).

4. Discussion

4.1. Effects of exchangeable cation on aflatoxin adsorption

The exchange cation (Na, Ca, Ba) had profound effects on the adsorption of non-ionic compounds by smectites (Johnston et al., 2004; Aggarwal et al., 2006; Liu et al., 2012; Deng et al., 2012). The effects of the cations, however, differed from compound to compound. The adsorption of atrazine by beidellite and saponite was enhanced with Cs^+ and K^+ saturation but low adsorption was observed on Ca^{2+} and Na^+ saturated smectites (Aggarwal et al., 2006). The lower hydration energy of Cs^+ and K^+ allowed the cation to get closer to the surface charge and reduce the interlayer space. Closer layers enhanced the adsorption of atrazine due to the planar molecule oriented parallel to the clay mineral surface and tight bonding to the hydrophobic surfaces (Aggarwal et al., 2006).

The present study demonstrated that the interlayer cation exerted the same effects on the adsorption of aflatoxin for all the smectite samples. The higher adsorption was observed in the Ba-saturated smectites, this observation in agreement with previous reported experiments (Deng et al., 2012). The lower hydration energy of Ba resulted in less amount of water held in the interlayer space, which allowed more space for aflatoxin molecules.

4.2. Effect of layer charge density on aflatoxin adsorption

The affinity of smectites for neutral organic compounds was also determined by the structural charge of the smectite. In the early studies by Laird et al. (1992), the adsorption of atrazine by different smectites showed an inverse correlation between the smectites affinity and the CEC. Jaynes and Zartman (2011) observed a similar effect of the smectites charge density on the adsorption of aflatoxin in corn meal solution, in which proteins and other organic compounds were present. At low aflatoxin concentrations, the mycotoxin retention was higher for the low-charge smectite (CEC = 76.4 cmol(+)/kg) in comparison to the high-charge smectite (CEC = 125 cmol(+)/kg (Jaynes and Bigham, 1987). The results by Deng et al. (2012) also demonstrated that high layer charge density can significantly reduce the affinity and maximum adsorption of aflatoxin molecules. When a high charge smectite

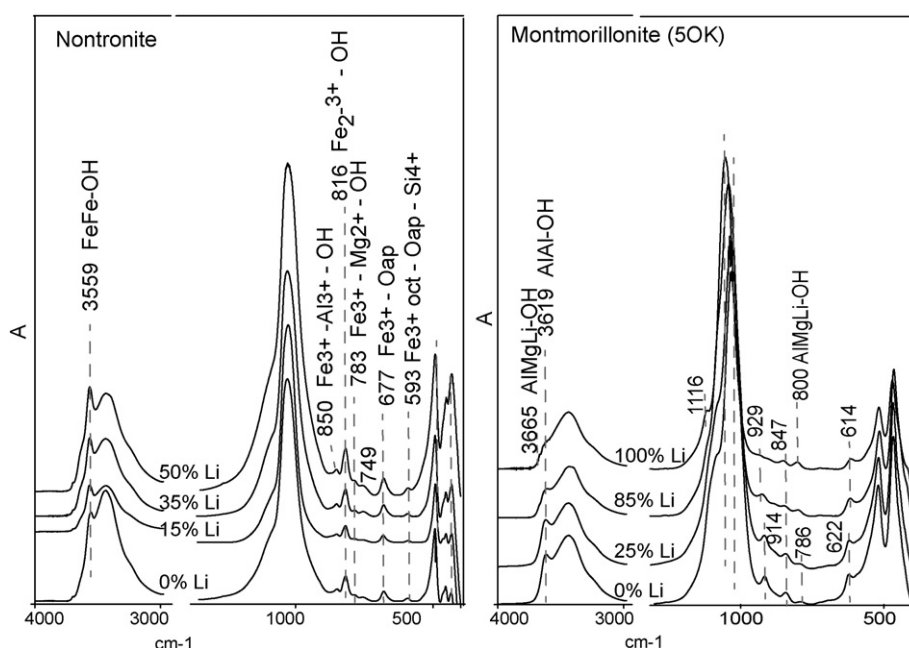


Fig. 7. FTIR spectra of Li-treated montmorillonite 50K and nontronite at 0% humidity.

(50K)(0.49 charge/half unit cell) was used, the maximum aflatoxin adsorption of the Ba-saturated smectites was 0.4 mol/kg. The aflatoxin adsorption isotherm of this high charge smectite did not fit the Langmuir model. In contrast, a low charge smectite (0.34 charge/half unit cell) adsorbed aflatoxin up to 0.61 mol/kg.

Jaynes and Zartman (2011) observed an increase in the aflatoxin adsorption in corn meal when the layer charge density of a high charge smectite was reduced by Li-treatment. Similarly, reducing the layer charge of montmorillonite 50K and nontronite, increased the adsorption of aflatoxin in both samples. As the CEC was reduced, a maximum toxin adsorption was reached at an optimal CEC around 86 cmol/kg. The main difference was that Li-nontronite only adsorbed up to 0.5 mol/kg, while Li-montmorillonite (50K) reached a maximum adsorption of 0.8 mol/kg at the similar CEC value. In addition, the low adsorption affinity in nontronite at the original CEC was increased as the layer charge was reduced. The results indicated that it is possible to modify high charge density smectites to improve their adsorption for aflatoxin.

4.3. Effect of layer charge origin on aflatoxin adsorption

Most of the available studies have observed an increase on the adsorption of non-polar organic compounds like atrazine and dioxins by smectites with tetrahedral charge (Aggarwal et al., 2006). A comparison of the atrazine adsorption on two smectites (beidellite and montmorillonite) with similar CEC but different layer charge origin demonstrated the higher binding affinity of the smectite with tetrahedral charge (beidellite) (Aggarwal et al., 2006).

The results from the Li-treated montmorillonite and nontronite indicated the influence of the layer charge origin. The maximum adsorption of both Li-treated smectites were reached at a CEC about 80 cmol/kg, but montmorillonite had a higher adsorption than nontronite. This observation indicated that smectites with octahedral charge, such as montmorillonite, are better AFB₁ binders than smectites with charges arising from the tetrahedral sheets. Another evidence of the charge location effect was observed by comparing aflatoxin adsorption on hectorite and beidellite. The CEC of beidellite was similar to hectorite but the adsorption of aflatoxin was significantly lower in the tetrahedrally charged beidellite. The greater affinity and adsorption capacity of smectites with octahedral charge was attributed to more diffused layer charge that reduced the repulsion of aflatoxin molecules and allowed more interlayer expansion in comparison to the more localized layer charge on tetrahedral charged smectites. Another possible explanation could be the structural configuration of aflatoxin. Atrazine and dioxins are planar compounds that benefit from a closer or reduced interlayer spacing. The aflatoxin molecule is not completely planar, as it possesses a terminal furan ring that is tilted in the cis configuration. The reduced layer charge of Ba-nontronite increased the adsorption but the more localized charge and the lower hydration energy of the Ba cations can reduce the swelling of the interlayer, which could restrict the accessibility for aflatoxin molecules. The results of the present study indicated that smectites with high tetrahedral charge can be improved to adsorb more aflatoxin, but they were not as effective as smectites with octahedral charge.

4.4. Effect of octahedral cation composition on aflatoxin adsorption

The aflatoxin adsorption experiments by Jaynes and Zartman (2011) showed a slight difference in the adsorption capacity between hectorite and montmorillonite, and both are octahedral charge smectites with similar CECs (89 and 76 cmol/kg, respectively). The trioctahedral smectite (hectorite) adsorbed more aflatoxin in comparison to the dioctahedral smectite (montmorillonite), but the reason of this effect was not addressed. The higher adsorption of the hectorite in this study in comparison to montmorillonite (4TX) was mainly attributed to the lower CEC of the hectorite.

The influence of the octahedral cation composition was also addressed by comparing the AFB₁ adsorption of similar charge origin and layer charge smectites. In beidellite and saponite, the charge arose from the tetrahedral sheets and both have a similar CEC (70 cmol/kg). The main difference was the dominant octahedral cation: Al in beidellite and Mg in saponite. The adsorption isotherms were very similar indicating a negligible effect of the octahedral cation composition.

5. Conclusions

The results corroborated previous observations that the exchangeable cation strongly affected the AFB₁ adsorption and demonstrated that the effect was similar on all smectites. The exchangeable cation influenced the size and polarity matching between the adsorbing domains and AFB₁ molecules in the interlayer. The adsorption capacity of the smectites was influenced by the layer charge density. As the negative charge sites on the surface increased, the non-polar sites for the sorption of aflatoxin decreased. The results also demonstrated that the low adsorption of high charge smectites can be improved by reducing the CEC.

The charge origin had a slight effect on the aflatoxin adsorption. Smectites with octahedral charge adsorbed more AFB₁ than the smectites with tetrahedral charge. The CEC reduction treatment demonstrated that octahedral charged smectites were the best adsorbents for aflatoxin. The diffused layer charge in smectites with octahedral charge allowed more accessibility to the interlayer space and less repulsion for AFB₁ molecules than the smectites where the charge was of tetrahedral origin.

The octahedral structural composition of the smectites had negligible effect on the aflatoxin adsorption. Similar adsorption capacities were observed when comparing di- and tri-octahedral smectites with either octahedral charge origin (montmorillonite and hectorite) or tetrahedral charge origin (beidellite, nontronite, and saponite). The bonding mechanism previously described for montmorillonite remains the same for the other smectites with no effects from the charge origin and the layer charge density.

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