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Mineralogical, Spectroscopic and Microstructural Characterization of Nigerian Bentonite Before and After Aflatoxin Detoxification of Stored Maize

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Abstract- ***Aflatoxin B_1 (AFB_1) contamination of maize poses a significant food-safety concern in Nigeria, creating a need for effective, affordable and locally available mitigation materials. This study characterized Nigerian bentonite before and after its application to AFB_1-contaminated stored maize using Fourier-transform infrared spectroscopy (FTIR), X-ray diffraction (XRD), and scanning electron microscopy coupled with energy-dispersive X-ray spectroscopy (SEM–EDS). FTIR analysis showed that the principal aluminosilicate framework was retained after treatment, with characteristic Si–O stretching and hydroxyl-related bands remaining evident, although changes occurred in water-associated absorption bands. XRD analysis identified montmorillonite as the dominant clay mineral, with minor quartz and feldspathic phases, and demonstrated persistence of the principal smectitic structure following application. SEM revealed irregular, flaky and aggregated particles, with observable differences in surface texture and aggregation after treatment. EDS confirmed silicon, iron and aluminium as the major elements, with relatively minor variations in elemental proportions between raw and post-treatment bentonite. Overall, the findings demonstrate that the Nigerian bentonite retained its fundamental mineralogical and aluminosilicate structure while undergoing measurable hydration- and surface-related changes following interaction with stored maize. These characteristics provide a favourable physicochemical basis for further investigation of the material as a locally sourced adsorbent for AFB_1 mitigation. However, quantitative adsorption, desorption, kinetic and toxicological studies are required to establish its detoxification efficiency, binding mechanisms and practical safety.***

Keywords: Bentonite; Aflatoxin B_1; Montmorillonite; FTIR; XRD; SEM–EDS; Stored maize; Clay adsorbent

I. INTRODUCTION

Aflatoxin contamination remains a significant food-safety challenge in maize, particularly in tropical and subtropical environments where elevated temperature, moisture and inadequate post-harvest handling favour the growth of aflatoxigenic Aspergillus species and subsequent toxin production. Among the major aflatoxins, aflatoxin B_1 (AFB_1) is regarded as the most toxic and is classified as a human carcinogen, making its occurrence in staple foods a major public-health concern (International Agency for Research on Cancer [IARC], 2012). Recent evidence confirms that mycotoxin contamination remains widespread in Nigerian maize. A systematic review covering 1,627 maize samples from different Nigerian agroecological zones reported frequent occurrence of aflatoxins, with contamination varying according to geographical location and season and with substantial proportions of samples exceeding recommended regulatory limits (Thierry et al., 2025). Similarly, Bashir et al. (2025) detected mycotoxins and aflatoxin-producing Aspergillus section Flavi in maize from north-western Nigeria, with 50% of the Flavi isolates identified as aflatoxigenic. These findings reinforce the need for effective, practical and locally adaptable approaches for reducing aflatoxin exposure while maintaining the nutritional and physicochemical quality of maize.

Adsorption with clay minerals has attracted considerable attention as a promising strategy for aflatoxin mitigation because of the strong affinity of certain smectite minerals for AFB_1. Bentonite is a naturally occurring clay dominated by montmorillonite, whose layered structure, high

specific surface area and exchangeable interlayer cations provide sites capable of interacting with AFB_1 molecules (Barrientos-Velazquez et al., 2024; Oladele et al., 2025). However, the binding performance of bentonite is not uniform and depends strongly on mineralogical composition, montmorillonite content, layer characteristics, exchangeable cations and other physicochemical properties. Recent experimental evidence demonstrated substantial differences in AFB_1 adsorption among bentonites with different mineralogical and structural characteristics, with calcium-rich montmorillonites generally showing strong adsorption performance (Daković et al., 2026). Likewise, selected Texas bentonites have demonstrated effective AFB_1 sequestration, although differences in adsorption capacity and clay properties were observed among samples (Barrientos-Velazquez et al., 2024). Therefore, evaluating the detoxification potential of bentonite requires not only determination of its AFB_1-binding efficiency but also detailed characterization of the mineralogical, chemical and structural properties that influence adsorption.

Nigeria possesses considerable clay resources, including bentonitic materials that may offer inexpensive and locally accessible alternatives for mycotoxin management. Nevertheless, limited information is available on the mineralogical, spectroscopic, morphological and elemental characteristics of Nigerian bentonite before and after interaction with aflatoxin-contaminated maize. Such characterization is important because adsorption of AFB_1 may be accompanied by changes in surface chemistry, functional groups, mineral phases, morphology or elemental distribution. Fourier-transform infrared spectroscopy (FTIR) is particularly useful for identifying functional groups and structural changes in clay minerals, while X-ray diffraction (XRD) provides information on crystalline mineral phases and structural modifications. Scanning electron microscopy coupled with energy-dispersive X-ray spectroscopy (SEM–EDS) further enables assessment of surface morphology and elemental composition (Madejová, 2003). Consequently, this study characterized Nigerian bentonite before and after its application in the detoxification of AFB_1-contaminated stored maize using FTIR, XRD and SEM–EDS. Specifically, the study evaluated changes in the mineralogical, spectroscopic, morphological and elemental characteristics of the bentonite following its application. The findings provide insight into the behaviour of indigenous bentonite during AFB_1 adsorption and contribute to the assessment of its potential as a locally sourced mineral adsorbent for sustainable mycotoxin management.

II. MATERIALS AND METHODS

Bentonite Sample

Bentonite clay used in the study was characterized before and after its application in the treatment of stored maize. The untreated material was designated raw bentonite, whereas the material recovered following maize treatment was designated post-treatment bentonite.

The samples were subjected to FTIR, XRD and SEM–EDS analyses to determine changes in functional groups, crystalline mineral phases, surface morphology and elemental composition.

Fourier Transform Infrared Spectroscopy

Fourier-transform infrared (FTIR) spectroscopy was used to characterize the principal functional groups and structural vibrations of the bentonite before and after application to stored maize. Spectra were acquired using a PerkinElmer Spectrum 100 FTIR spectrometer equipped with a DTGS detector over 4000–400 cm^{-1} at a resolution of 4 cm^{-1}. Approximately 0.8 mg of powdered bentonite was thoroughly homogenized with 0.2 mg of spectroscopic-grade KBr and pressed into pellets for transmission analysis. Spectra were collected and processed according to the manufacturer's operating procedures, and characteristic absorption bands associated with O–H, H–O–H, Si–O and Al–OH vibrations were used to evaluate the mineral structure and possible changes following treatment. The KBr pellet approach is well established for routine clay-mineral characterization, while recent studies confirm the utility of FTIR for identifying structural and chemical changes in bentonite and related clay minerals (Madejová, 2003; Kuter et al., 2023; Zhang et al., 2023).

X-ray Diffraction Analysis

Powder X-ray diffraction (XRD) analysis was performed to characterize the mineralogical composition of the bentonite before and after application to stored maize using a Rigaku MiniFlex 600 diffractometer equipped with Cu Kα radiation (λ = 1.5406 Å), operated at 40 kV and 15 mA. Approximately 0.5–1.0 g of each sample was gently pulverized with an agate mortar and pestle, sieved to obtain a homogeneous powder, and back-loaded into zero-background silicon sample holders to minimize preferred orientation. Diffraction patterns were collected over a 2θ range of 5–70° at a step size of 0.02° and a scanning rate of 4° min^{-1}. The diffractograms were processed using SmartLab Studio II software, and crystalline phases were identified by matching observed peak positions and relative intensities with reference patterns in the International Centre for Diffraction Data (ICDD) Powder Diffraction File (PDF) database, consistent with established approaches for powder diffraction phase identification and materials characterization (Kabekkodu et al., 2024; Kabekkodu & Blanton, 2024).

Scanning Electron Microscopy and Energy-Dispersive X-ray Spectroscopy

Surface morphology and elemental composition of the bentonite samples before and after treatment were examined using a Phenom ProX desktop scanning electron microscope coupled with an energy-dispersive X-ray spectroscopy (EDS) detector. The samples were gently ground to approximately 100 mesh and mounted on aluminium stubs using double-sided carbon adhesive tabs before examination. SEM images were obtained at different magnifications to observe changes in particle shape, aggregation, surface texture and apparent pore structure. EDS spectra were collected from representative areas of each sample to determine the major elemental constituents using the instrument's standard semi-quantitative analysis procedure. The SEM–EDS approach is widely used for evaluating the microstructural and elemental characteristics of bentonite and other clay minerals, providing useful evidence of changes in surface features and elemental composition following treatment (Berhe et al., 2024; Quero-Jiménez et al., 2021; Tsige et al., 2023). Because EDS provides localized, semi-quantitative elemental information that may vary with sample heterogeneity and analytical conditions, the results were interpreted primarily for comparative characterization rather than as direct evidence of specific chemical reactions.

III. RESULTS

FTIR Characterization

The superimposed FTIR spectra of raw and post-treatment bentonite showed broadly similar spectral profiles, indicating retention of the principal aluminosilicate framework following application to stored maize.

Both samples exhibited a prominent absorption band in the region of approximately 3625 cm^{-1}, attributable to structural hydroxyl groups associated with the clay mineral framework. A broad band around 3440 cm^{-1} and an absorption near 1638 cm^{-1} were associated primarily with water-related vibrations. The strong band around 1035 cm^{-1} was assigned to Si–O stretching within the tetrahedral silicate structure.

The post-treatment sample displayed reduced intensity of the broad water-associated bands relative to the raw material. Other principal structural bands remained detectable, indicating that the dominant mineral framework was retained following application.

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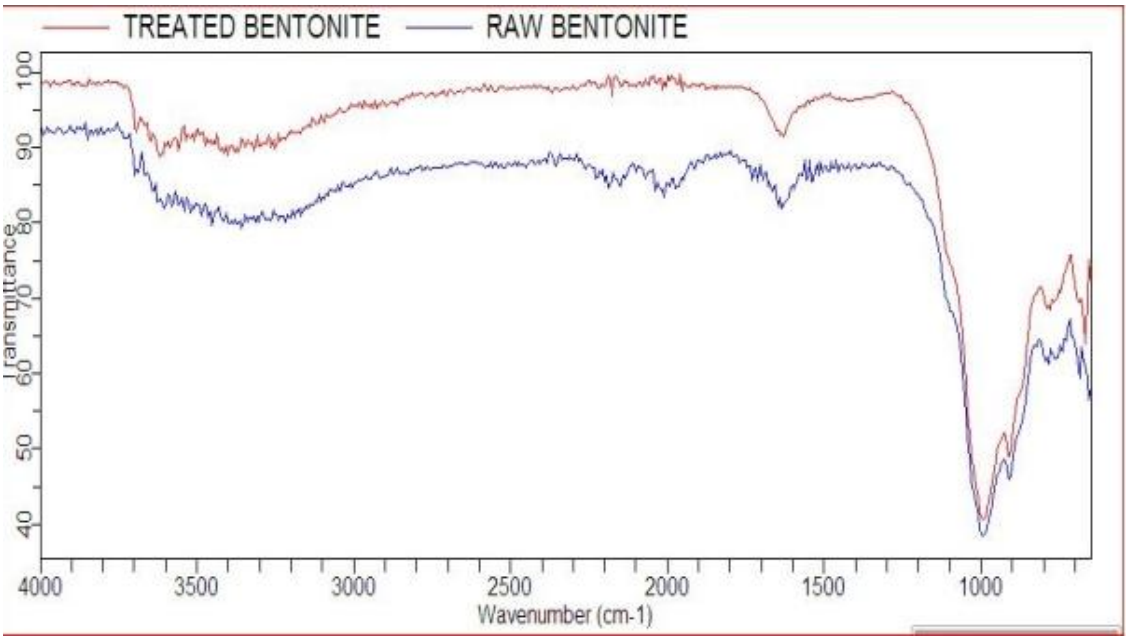

Figure 1. Superimposed FTIR spectra of raw bentonite and bentonite recovered after application to stored maize.

XRD Characterization

The XRD pattern of the raw bentonite displayed prominent reflections at approximately 2θ = 6.1°, 19.8°, 28.5°, 35.0° and 61.9°. The low-angle reflection around 6.1°, corresponding to a d-spacing of approximately 14.5 Å, was consistent with a hydrated smectitic phase and supported the identification of montmorillonite as the principal clay mineral.

Additional reflections were attributed to minor non-clay mineral phases, including quartz and feldspathic components.

The post-treatment bentonite exhibited major reflections at approximately 2θ = 6.2°, 19.7°, 26.6°, 35.0° and 61.8°. The persistence of the low-angle basal reflection demonstrated that the principal smectitic structure remained detectable after application to maize.

Relative to the raw material, some changes in peak intensity and breadth were observed, particularly at lower diffraction angles. These changes may reflect differences in hydration state, particle aggregation, preferred orientation or surface/interlayer interactions rather than complete transformation of the mineral phase.

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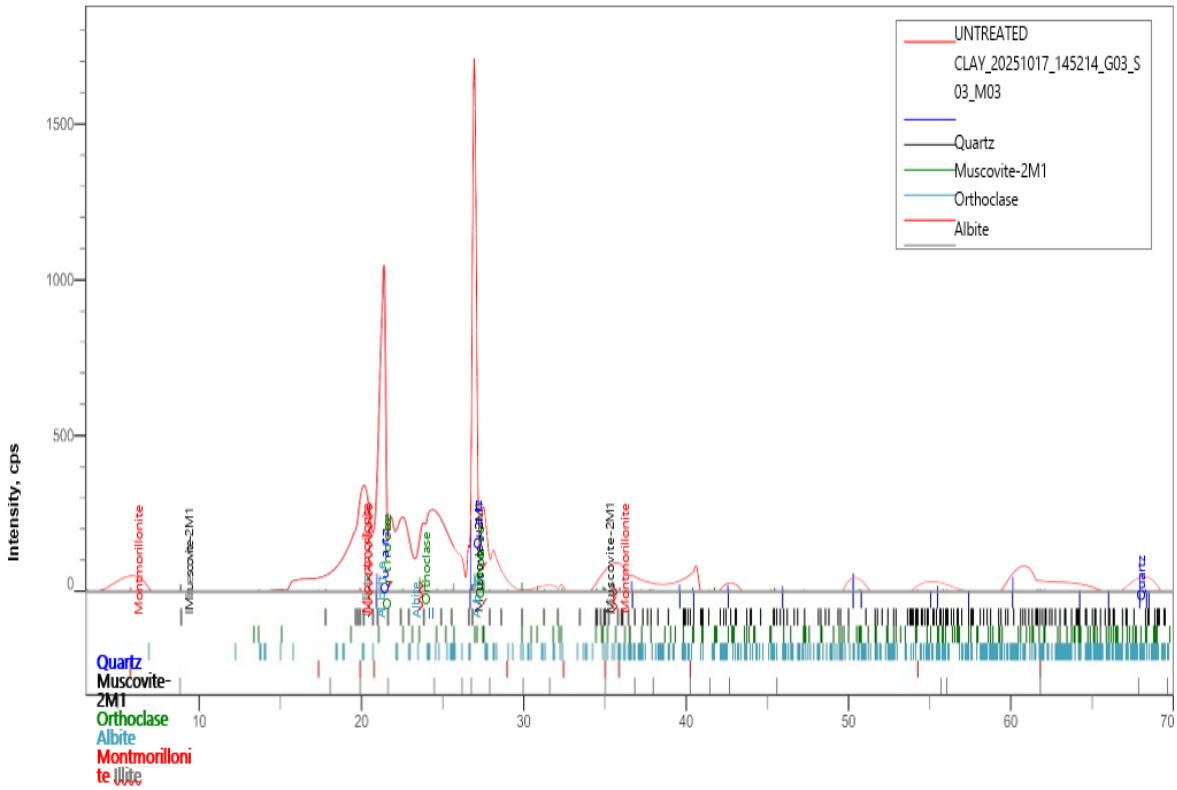

Figure 2: X-Ray diffraction (XRD) of the raw bentonite clay

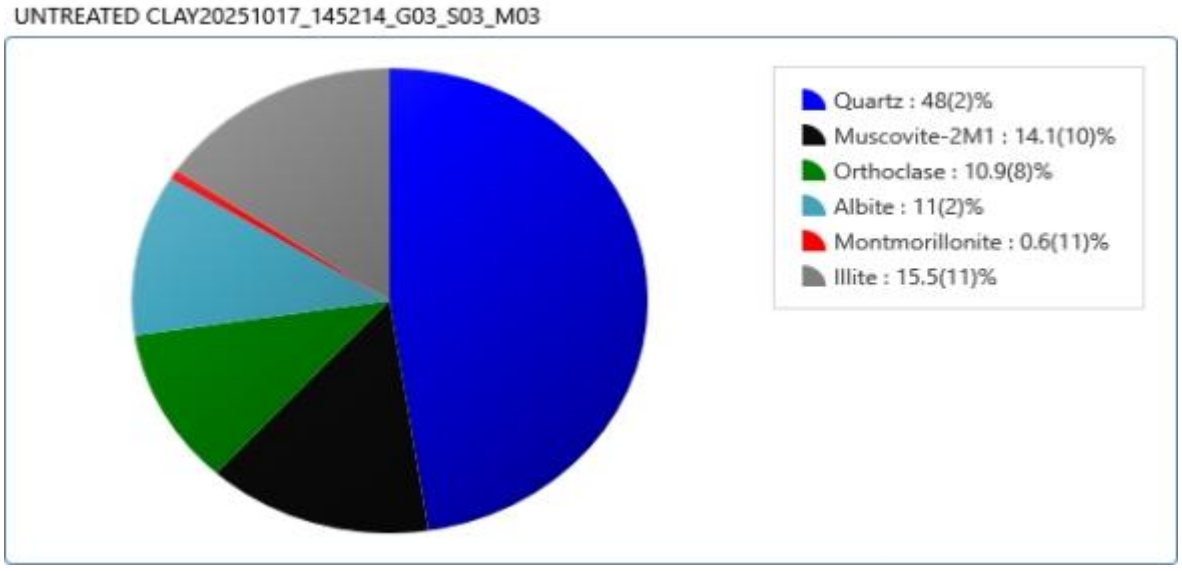

Figure 3: Pie chart representation of XRD diffractogram and mineral-phase distribution of raw bentonite.

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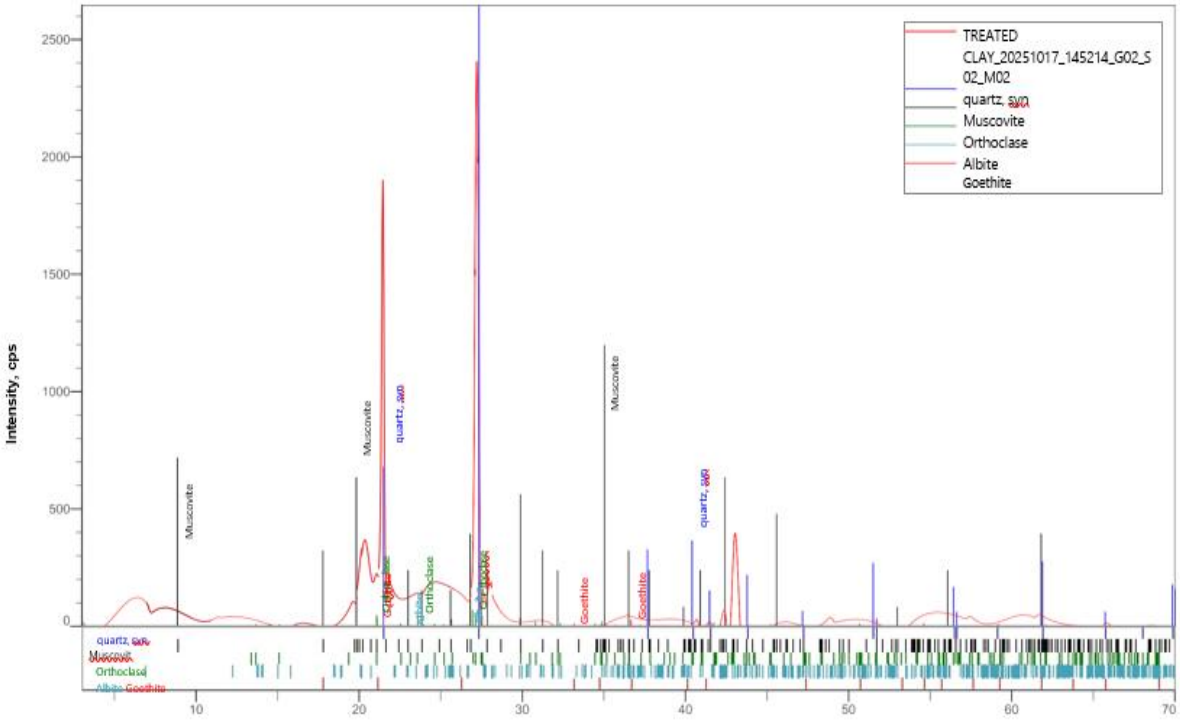

Figure 4: X-Ray diffraction (XRD) of the bentonite clay use for the maize treatment

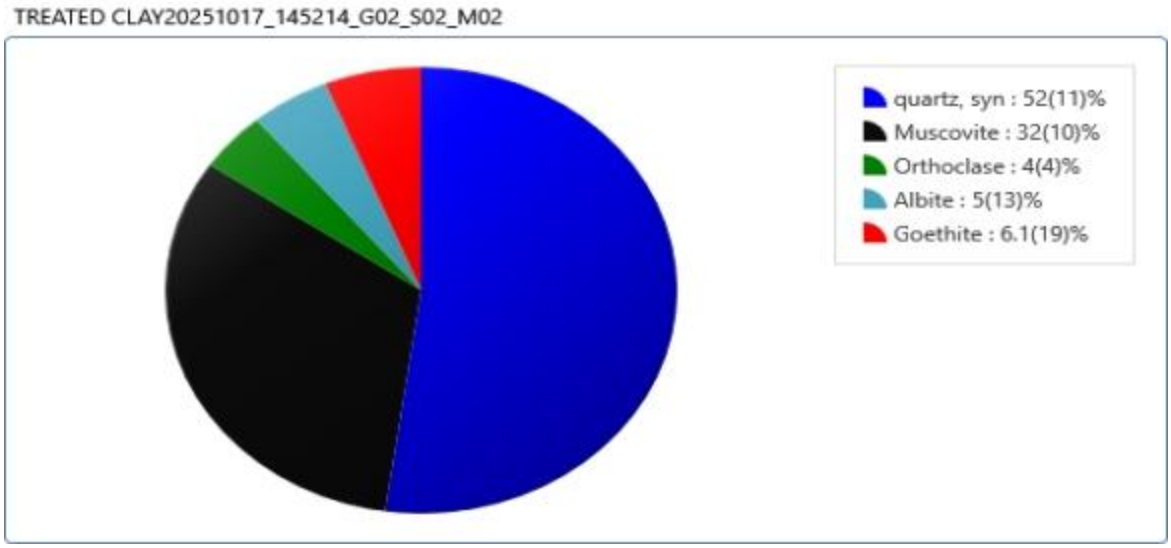

Figure 5: Pie chart representation of the XRD diffractogram and mineral-phase distribution of post-treatment bentonite.

SEM Characterization

SEM examination revealed heterogeneous particles with irregular, flaky and plate-like morphologies. The particles occurred both individually and as aggregates, consistent with the morphology commonly observed in smectite-rich clay materials.

The post-treatment sample exhibited apparent differences in surface texture and particle aggregation relative to the raw bentonite. These differences indicate that exposure to the maize matrix and associated moisture conditions may have altered the surface state of the clay.

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However, morphological changes observed by SEM should be interpreted cautiously because sample preparation, drying and particle dispersion can themselves influence clay morphology.

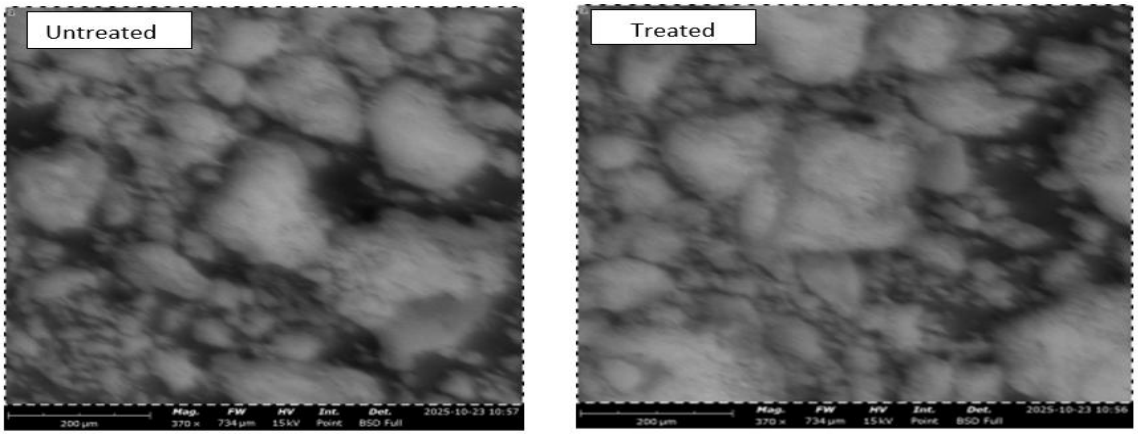

Figure 6. SEM micrographs of raw and post-treatment bentonite at representative magnifications.

EDS Elemental Composition

EDS analysis showed that silicon, iron and aluminium were the dominant elements in both raw and post-treatment bentonites

Table 1. EDS elemental composition of raw and post-treatment bentonite

Element	Raw bentonite Atomic Conc. (%)	Raw bentonite Weight Conc. (%)	Post-treatment Atomic Conc. (%)	Post-treatment Weight Conc. (%)
Si	53.59	43.59	57.33	45.76
Fe	19.75	31.94	18.41	29.22
Al	17.65	13.79	16.33	12.52
K	3.40	3.85	3.19	3.55
Ca	1.60	1.86	1.59	1.82
Ti	0.89	1.24	1.29	1.75
Mg	1.29	0.91	1.19	0.82
Mn	0.51	0.82	—	—
Cl	0.52	0.54	—	—
S	0.42	0.39	—	—
Ag	0.14	0.44	0.15	0.46
Zr	0.24	0.64	—	—
Sn	—	—	0.53	1.79
Cs	—	—	0.00	1.70
Cr	—	—	0.00	0.62

The principal compositional pattern was retained after maize treatment. Silicon increased from 43.59 to 45.76 wt%, whereas iron and aluminium decreased from 31.94 to 29.22 wt% and from 13.79 to 12.52 wt%, respectively. Smaller changes were observed for potassium, calcium, magnesium and titanium. The

appearance or disappearance of minor elements should be interpreted with caution because EDS measurements are influenced by sampling location, surface heterogeneity, matrix effects, detection limits and normalization of the measured spectrum.

IV. DISCUSSION

The combined FTIR, XRD and SEM–EDS results demonstrate that the Nigerian bentonite retained its principal montmorillonite-rich aluminosilicate structure after application to stored maize, although changes in hydration, particle aggregation and surface elemental composition were evident. The FTIR spectrum showed a strong absorption around 1035 cm^{-1}, characteristic of Si–O stretching in the silicate framework, while the bands around 3440 and 1638 cm^{-1} were associated mainly with O–H stretching and bending of adsorbed or interlayer water. Similar FTIR features have been reported for montmorillonite-containing materials, confirming that Si–O and O–H vibrations are useful indicators of the structural and hydration characteristics of smectitic clays (Shah et al., 2024; Berhe et al., 2024). The persistence of these characteristic bands after treatment indicates that contact with the maize matrix did not cause major disruption of the bentonite framework. The reduction in the intensity of the water-associated bands is more plausibly related to changes in interlayer hydration and moisture status during treatment, drying and subsequent analysis rather than to destruction of the clay structure. This interpretation is consistent with recent work showing that water content and interlayer hydration can substantially influence the physicochemical behaviour and basal spacing of montmorillonite (Long & Wang, 2024; Skoubris et al., 2024). Thus, the observed FTIR changes are better interpreted as evidence of altered hydration or surface environment rather than formation of a fundamentally different mineral phase.

The XRD findings provide further evidence for structural preservation. The prominent low-angle reflection at approximately 2θ = 6.1–6.2°, corresponding to a basal spacing of approximately 14–15 Å, is consistent with a hydrated smectitic phase and supports the identification of montmorillonite as the principal clay mineral. The persistence of this reflection after maize treatment indicates that the expandable smectite structure remained substantially intact. The additional reflections attributed to quartz and feldspathic minerals further demonstrate that the material is a heterogeneous bentonite containing associated non-clay phases rather than pure montmorillonite. Comparable mineralogical heterogeneity has been reported in natural bentonites and other clay deposits, where montmorillonite commonly occurs together with quartz, feldspar and other accessory minerals (Berhe et al., 2024). The relatively small changes in peak intensity and breadth between the raw and post-treatment samples should therefore not be interpreted as evidence of major mineral transformation. Instead, they may arise from changes in hydration, particle aggregation, preferred orientation or the distribution of clay particles during sample preparation. Recent studies of montmorillonite similarly demonstrate that changes in interlayer hydration and structural environment can alter diffraction behaviour without necessarily destroying the underlying clay framework (Long & Wang, 2024; Dongmo et al., 2024). The present XRD results therefore indicate mineralogical stability following application to maize, while suggesting that the physicochemical environment of the clay was modified during treatment.

The SEM observations complement the spectroscopic and diffraction results by showing irregular, flaky and aggregated particles typical of smectite-rich clay materials. The differences in surface texture and aggregation between untreated and post-treatment bentonite suggest that exposure to the maize matrix and associated moisture conditions modified the physical state of the clay. Similar aggregated, layered and heterogeneous morphologies have been reported for montmorillonite-based materials examined by SEM, reflecting the tendency of fine clay particles to form tactoids and aggregates during drying and sample preparation (Shah et al., 2024). Importantly, the apparent increase or alteration in aggregation should not be interpreted as direct evidence of increased adsorption-site availability because SEM morphology is strongly influenced by sample grinding, drying, mounting and dispersion. Likewise, the EDS results confirmed Si and Al as major

constituents, accompanied by Fe and smaller amounts of K, Ca, Mg and Ti, which is consistent with the aluminosilicate nature of montmorillonite-rich bentonite (Berhe et al., 2024). Silicon increased from 43.59 to 45.76 wt%, whereas Fe and Al decreased from 31.94 to 29.22 wt% and 13.79 to 12.52 wt%, respectively. These differences indicate variation in the analysed surface regions but should not be regarded as definitive evidence of cation exchange, dissolution or elemental leaching. EDS is a localized and semi-quantitative technique, and the measured composition can be affected by particle heterogeneity, analytical position, matrix effects and spectrum normalization. Consequently, the present SEM–EDS findings are best interpreted as evidence of changes in surface compositional distribution rather than proof of a specific chemical mechanism.

The preservation of the montmorillonite structure is nevertheless important when considering the potential of the Nigerian bentonite for AFB_1 adsorption. Recent research has established that AFB_1 binding by smectitic clays is governed not simply by the presence of montmorillonite, but by a combination of mineralogical and physicochemical properties, particularly layer-charge density, exchangeable cations, interlayer accessibility and the size and polarity of non-polar surface domains (Deng et al., 2024). This is particularly relevant to the present material because the EDS results indicate the presence of Ca, Mg, K and other exchangeable or associated elements, while XRD confirms a hydrated smectitic phase. Deng et al. (2024) demonstrated that differences in exchangeable cations and layer-charge density can produce substantial differences in AFB_1 adsorption among smectites. Similarly, Barrientos-Velazquez et al. (2024) reported that bentonites from different sources varied in their ability to bind AFB_1 and reduce aflatoxicosis, demonstrating that clay source and physicochemical characteristics are important determinants of performance. These findings provide a stronger basis for interpreting the present Nigerian bentonite as a potentially useful adsorbent, but they also demonstrate why mineral identification alone cannot be used to predict its adsorption capacity.

The present observations are also consistent with recent experimental evidence that natural and modified montmorillonite clays can provide substantial AFB_1 adsorption and detoxification potential. Oladele et al. (2025) demonstrated that engineered montmorillonite materials can effectively adsorb AFB_1 and reduce its toxic effects, while emphasizing the importance of clay surface characteristics and modification in determining performance. Likewise, studies of raw bentonitic clay have reported high AFB_1 removal under appropriate experimental conditions, demonstrating the potential of naturally occurring bentonites as practical mycotoxin adsorbents (Barrientos-Velazquez et al., 2024; Oladele et al., 2025). However, adsorption performance is highly dependent on experimental conditions, including clay dose, particle size, pH, moisture, toxin concentration and gastrointestinal environment. Recent evidence further indicates that particle-size reduction and micronization can influence AFB_1 adsorption efficiency, highlighting the importance of physical characteristics in addition to mineral composition (Influence of Particle Size and Micronization, 2026). Consequently, the structural stability observed in the Nigerian bentonite should be regarded as a favourable prerequisite rather than direct evidence of superior AFB_1-binding capacity.

An important implication of the present findings is that the apparent changes observed after maize treatment may represent interaction between the clay surface, water and components of the maize environment rather than a transformation of the dominant mineral phase. AFB_1 adsorption by smectites is believed to involve interactions between the toxin and mineral surfaces or interlayer domains, with exchangeable cations and hydration influencing accessibility and binding strength (Deng et al., 2024). Therefore, the reduction in water-associated FTIR bands and changes in surface aggregation observed here may be relevant to the adsorption environment, although the available data do not permit the specific binding mechanism to be established. In particular, FTIR, XRD and SEM–EDS cannot independently distinguish AFB_1 adsorption from other possible interactions involving moisture, lipids, proteins or inorganic constituents of the maize matrix. Direct

identification of AFB_1–clay complexes would require complementary adsorption and spectroscopic investigations under controlled conditions.

Generally, the present findings compare favourably with recent studies of montmorillonite and bentonite materials by demonstrating that the Nigerian clay retained its fundamental mineralogical identity after application to stored maize while exhibiting measurable changes in hydration and surface characteristics. The agreement between FTIR and XRD results provides convincing evidence for preservation of the aluminosilicate framework, while SEM–EDS demonstrates associated changes in morphology and localized elemental composition. However, the results should be interpreted within the limitations of the characterization techniques. Structural preservation does not necessarily imply high AFB_1 adsorption, and changes in EDS elemental proportions cannot independently establish cation exchange or toxin binding. Therefore, the Nigerian bentonite should be considered a promising candidate for AFB_1 mitigation rather than a confirmed detoxification agent on the basis of characterization alone. Further studies should quantify AFB_1 adsorption and removal, determine adsorption isotherms and kinetics, evaluate the effects of clay dose, pH, moisture and particle size, determine cation-exchange capacity and specific surface area, and assess desorption and gastrointestinal stability. Such investigations would provide the mechanistic and performance evidence required to establish whether this locally sourced Nigerian bentonite can be developed into a safe, effective and economically viable material for sustainable aflatoxin management in maize and animal-feed systems.

V. CONCLUSION

The present study demonstrated that the Nigerian bentonite investigated was predominantly montmorillonite-rich and retained its principal aluminosilicate structure following application to stored maize. FTIR analysis confirmed the persistence of characteristic hydroxyl and silicate vibrations, while XRD demonstrated the continued dominance of the smectitic mineral phase. SEM analysis revealed irregular, flaky and aggregated particles, and EDS confirmed silicon, iron and aluminium as the major detected elements.

Comparison of raw and post-treatment bentonite revealed changes in water-associated FTIR bands, minor modifications in XRD peak characteristics, alterations in surface morphology and measurable differences in elemental proportions. These findings indicate that the bentonite interacted with the maize environment while retaining its fundamental mineralogical identity.

The results support the potential of locally sourced Nigerian bentonite as a candidate material for aflatoxin-management research. However, the present characterization should be regarded as a foundation for adsorption studies rather than definitive evidence of toxin-specific binding mechanisms. Further work should include quantitative AFB1 adsorption experiments, adsorption isotherms and kinetics, cation-exchange capacity, specific surface-area determination, desorption studies, contaminant screening and toxicological evaluation. Such investigations will be essential for establishing the efficacy, reproducibility and safety of Nigerian bentonite for practical application in maize and feed systems.

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