

# Immobilization of TNT and RDX Explosives from Contaminated Soil at Demining Sites in Amuru District, Northern Uganda, Using Rice Husk Biochar

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## Abstract

The study investigated using rice husk biochar (RHB) to immobilize TNT and RDX explosives in soil at demining sites in Amuru District, Uganda. RHB produced via pyrolysis at 550°C was applied at a rate of 5% w/w to soil samples spiked at 50 µg·g<sup>-1</sup> RDX or TNT. The Liquid chromatography-tandem mass spectrometer (LC-MS/MS) analysis showed that the RHB immobilized 56% - 67% of RDX and 55% - 69% of TNT. The biochar's porous structure and surface functional groups facilitated the explosive adsorption, reducing bioavailability. This demonstrates RHB's effectiveness as an environmentally friendly and cost-effective remediation strategy.

## Keywords

LIQUID Chromatography, Soil Remediation, Bioavailability, Anaerobic Pyrolysis

## 1. Introduction

The detection of high-explosive residues RDX and TNT in soil serves as a clear indicator of explosive device detonation (Yu et al., 2017). These chemical compounds are synthetic and do not occur naturally in the environment. Due to their toxicity and persistence, explosive residues pose significant risks to both humans

and ecological systems. Extensive research has been devoted to understanding the toxicological impacts of explosive compounds like TNT and RDX on plants, invertebrates, and animals (Robidoux et al., 2003).

Various remediation strategies have been explored to mitigate explosive soil contamination and health hazards associated with explosive contamination of soil. Conventional remediation methods often rely on the physicochemical properties of the adsorbents. However, these approaches are typically costly, and inefficient and may generate harmful environmental by-products (Singh & Singh, 2011). The success of any remediation effort is influenced by site-specific factors such as soil composition, moisture content, and soil pH, thereby making site assessment essential (Kumar et al., 2018). Regular monitoring is also crucial to enhance the effectiveness of contaminant degradation and ensure sustainable remediation outcomes.

Among emerging solutions, microbial remediation has shown great promise. Certain microorganisms can metabolize xenobiotic compounds, transforming them into less toxic or inert substances (Chatterjee et al., 2017; Sangwan et al., 2018). The efficiency of microbial degradation can further be enhanced using biocarriers such as biochar and eggshells.

Biochar, a carbon-rich material produced via pyrolysis of biomass under limited oxygen conditions, has gained attention for its soil-enhancing and environmental remediation properties (Wang & Wang 2019; Sharma et al., 2023). Biochar's effectiveness in immobilizing organic contaminants is attributed to its high cation exchange capacity, large surface area, stability, and rich carbon content (Rizwan et al., 2016). These properties can be tailored through modifications using acids, alkalis, metal ions, and oxidizing agents to target specific pollutants (Ahmad et al., 2012). Notably, biochar produced at lower pyrolysis temperatures tends to exhibit greater surface area and carbon content due to the formation of micropores (Chen & Yuan, 2011).

Recent studies have demonstrated the potential of biochar in remediating explosive-contaminated soils. For instance, Sharma et al. (2023) reported that a bioformulation combining coconut husk biochar and *Arthrobacter subterraneus* achieved degradation rates of 85.98% for RDX and 80.40% for HMX within 30 days. Similarly, Oh and Yoon (2016) observed that biochar amendments reduced the bioavailability of TNT, 2,4-DNT, and RDX to less than 10% of their initial concentrations within 10 days.

Despite these promising findings, there is limited research on biochar application for explosive remediation in Uganda. This study addresses the gap by investigating the use of RHB for the immobilization of RDX and TNT in soils collected from demining sites in Amuru District, Northern Uganda. The outcomes of this research offer valuable insights for environmental scientists and policymakers in Uganda, Africa, and beyond, contributing to the development of informed, sustainable remediation strategies for explosive-contaminated environments.

## 2. Materials and Methods

### 2.1. Reagents, Solvents, and Standards

All chemicals used were analytical grade. Explosive standards RDX and TNT were purchased from Accu Standards (New Haven, CT, USA) at a certified concentration of 1000  $\mu\text{g}\cdot\text{mL}^{-1}$  in acetonitrile: methanol (1:1). LCMS-grade methanol, LCMS-grade acetonitrile, LCMS-grade organic-free reagent water, ammonium formate, and formic acid were purchased from Arbbot Laboratory store, Uganda.

### 2.2. Instrumental Analysis

Instrumental analysis of samples was performed using the Shimadzu LCMS 8060 NX by the method reported by Ocaya et al. (2025). The mobile phase used contained formic acid and ammonium formate as buffers in this method. The LC-MS/MS instrument was operated in the Atmospheric Pressure Chemical Ionization (APCI) mode. The method had a run time of 13 minutes with the mobile phase starting at 5% B, which was increased to 95% B within 7.5 minutes, maintained for 2.5 minutes, dropped back to 5% B in the next 0.1 minutes, and finally maintained at 5% for the next 2.9 minutes before the next injection starts. The LC and MS conditions of the analytical instrument used to analyse the target analytes are shown in Tables 1-2, respectively.

**Table 1.** LC conditions of the analytical instrument.

<i>Analytical column</i>	Phenomenex kinetic XB-C18 (2.1 mm I.D. × 100 mm L, 2.6 $\mu\text{m}$ Thickness)
<i>Guard column</i>	Phenomenex security Ultra C18 2.1 mm I.D
<i>Mobile phase A</i>	10 mmol/L formic acid + ammonium formate-water
<i>Mobile phase B</i>	10 mmol/L formic acid + ammonium formate and methanol
<i>Flow rate</i>	0.3 ml/min
<i>Injection volume</i>	10 $\mu\text{L}$
<i>Column oven</i>	40 °C
<i>Elution mode</i>	Gradient

**Table 2.** MS conditions of the analytical instrument.

<i>Nebulizing gas flow:</i>	3 L/min
<i>Drying gas flow:</i>	10 L/min
<i>Heating gas flow:</i>	10 L/min
<i>Interface temperature:</i>	300 °C
<i>CID gas:</i>	270 Kpa
<i>Interface current:</i>	0.1 $\mu\text{A}$
<i>Heating block temperature:</i>	400 °C
<i>DL temperature:</i>	250 °C

### 2.3. Preparation of Rice Husk Biochar

The rice husk biochar was prepared following the method described by Sarkar et al. (2019). Rice husk was obtained from Wairama Agroprocessors Ltd, located in Jinja city, washed with distilled water, and dried in the oven at 60°C for 12 hours. Dried rice husk (7 g) portions were wrapped tightly in aluminium foil to prevent air contact, placed into a 50 mL crucible and covered, introduced into a muffle furnace, and pyrolyzed at 550°C for 2 hours. The biochar was removed from the furnace after cooling, ground, and then sieved through a 355-micron mesh. The sieved biochar was wrapped in aluminium foil and stored in a tight container at room temperature for further analysis.

### 2.4. Characterization of Rice Husk Biochar

The physicochemical properties of RHB determined were electrolytic conductivity (EC), pH, carbon (C), calcium (Ca), magnesium (Mg), sodium (Na), phosphorus (P), potassium (K), silicon (Si) contents, and cation exchange capacity (CEC). These physicochemical properties were characterized using methods reported by Okalebo et al. (2002) and Severo et al. (2020).

The RHB's surface functional groups and surface morphology were analyzed using methods reported by Palniandy et al. (2019). The Fourier Transform Infra-Red (FTIR) spectroscopy Bruker Optik GmbH Vertex 70 model was used to analyze the functional groups of the RHB using the attenuated total reflection (ATR) in the range of 4000  $\text{cm}^{-1}$  to 250  $\text{cm}^{-1}$  at a resolution of 4  $\text{cm}^{-1}$ . The surface morphology of the RHB was studied using a ZEISS-Gemini 500 Field Emission Scanning Electron Microscope (FESEM). Samples were applied with a platinum coating before the imaging.

### 2.5. Soil Sample Collection

Soil samples were collected from areas very close to the demining sites with no history of demolition of munitions. Three sites considered for the study were Okidi, Bibia, and Elegu demining sites located in the Amuru district, northern Uganda. Samples were randomly picked using an auger in a horizontal layer up to a maximum depth of about 30 centimeters and placed in labeled plastic bags with zip locks. Approximately 1 kg of soil was randomly collected from ten sample points per demining site. Stones, plant roots, and leaves were removed before storage. The soil samples were placed in plastic buckets containing ice blocks and transported to the laboratory for analysis. Soil samples were tested to ensure no contamination from TNT and RDX, which are target analytes in the study. Only soil samples free from contamination of the explosive compounds TNT and RDX were thoroughly mixed and stored at a temperature of -4°C for subsequent analysis.

### 2.6. Soil Characterization

Soil physico-chemical parameters determined were pH, total organic carbon, total

nitrogen, total organic matter, C: N ratio, particle size distribution (percent clay, percent silt, and percent sand), and (CEC). Characterization of selected physico-chemical properties was determined by the method reported by Okalebo et al. (2002).

## 2.7. Immobilization Studies

Before analysis, the collected soil samples were air-dried at 25°C for 24 hours. The dried soil sample was sieved to a particle size of  $\leq 2$  mm. The sieved soil sample was homogenized by turning it several times in a plastic bag before the sub-sample was taken for analysis. Three replicates of 2-gram portions of the soil of each sample site were placed separately in 15 mL centrifuge tubes spiked to  $50 \mu\text{g}\cdot\text{g}^{-1}$  with the standard solution of either TNT or RDX and further spiked to 5% with RHB. The contents of the different centrifuge tubes were equilibrated using 50:50 (v/v) methanol: acetonitrile and then incubated at a temperature of 25°C. The contents of each centrifuge tube were analyzed using LC-MS/MS at intervals of 0, 5, 10, 15, 20, 25, 30, 35, 40, 45, 50, 60, and 70 days.

## 2.8. Determination of Immobilization Efficiencies

Extraction of soil samples was performed by the method reported by Numbera (2006) with slight modifications. 2 g of each soil sample was mixed with 10 mL of methanol-acetonitrile mixture (50:50 v/v) and extracted on an overhead shaker for 18 hours. Extracts were centrifuged at 5000 rpm for 5 minutes, and the supernatant was filtered through 0.22  $\mu\text{m}$  polytetrafluoroethylene filters. Filtrate from each sample (1 mL) was pipetted and placed in a vial for LC-MS/MS analysis. The percentage of contaminants removed was calculated using Equation (1).

$$\frac{\text{Spiking concentration} - \text{Concentration after time, } t}{\text{spiking concentration}} \times 100 \quad (1)$$

## 2.9. Statistical Analysis

The statistical analyses of the data generated were performed using Microsoft Excel 2016 (v16.0) and SPSS software version 20. One-way ANOVA test was used to test for statistical differences in the mean amounts of TNT and RDX immobilized from the soil samples obtained from the study sites.

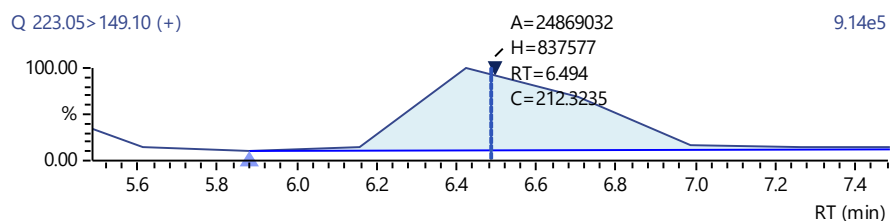
## 2.10. Ethical Consideration

The study was conducted after ethical approval by the Gulu University Research Ethical Committee (GUREC-2024-898) and the Uganda National Council for Science and Technology (UNCST-NS858ES). The bomb disposal experts pointed out the demining sites with brief information on vital aspects that greatly help determine the amounts of explosive compounds in soils. The sample collected was strictly for this study and not for any other purpose. All information on the study sites was kept private and treated with the confidentiality it deserved.

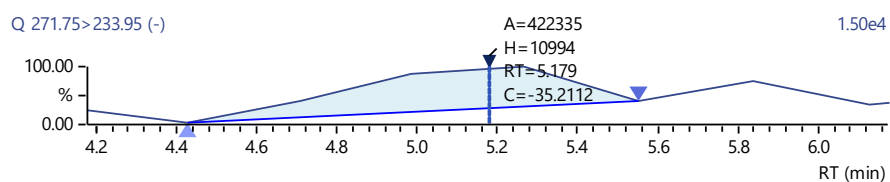
### 3. Results and Discussions

#### 3.1. Analysis of the Explosive Compounds

The retention times for RDX and TNT were about 6.494 and 5.179 minutes, respectively. The chromatograms for RDX and TNT are shown in **Figures 1-2**, respectively.

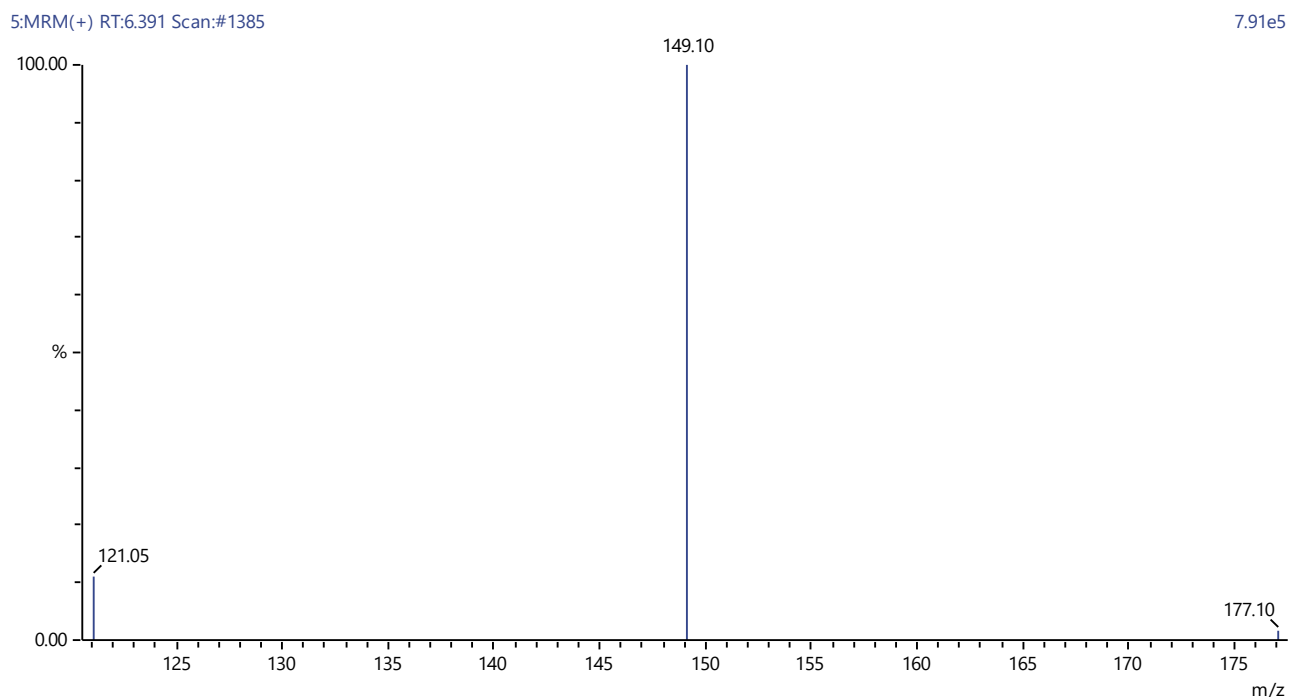


**Figure 1.** Chromatogram for RDX.



**Figure 2.** Chromatogram for TNT.

The MS spectrum for RDX is shown in **Figure 3**.



**Figure 3.** MS spectrum for RDX.

Mass Spectrometer (MS) Multiple Reaction Monitoring (MRM) transitions of the explosive compounds are shown in **Table 3**.

**Table 3.** MS-MRM transitions of the explosive compounds.

Explosive	Precursor ion	Product ion	Q1 Pre-Bias (V)	CE (V)	Q3 Pre-Bias (V)	Polarity
RDX	223.05	149.1 <sup>Q</sup>	-26	-19	-29	Positive
		177.1 <sup>Q</sup>				
		121.05 <sup>Q</sup>				
TNT	271.75	233.95 <sup>Q</sup>	18	15	16	Negative
		205.90 <sup>Q</sup>				
Picric acid	227.90	182.2 <sup>Q</sup>	25	18	18	Negative
		198.0 <sup>Q</sup>				
		63.1 <sup>Q</sup>				

<sup>Q</sup>Quantifier m/z and <sup>Q</sup>Qualifier m/z, Picric acid was used as an internal standard.

### 3.2. Physicochemical Properties of Soil

The pH values across the three sites, Okidi, Bibia, and Elegu, were relatively similar, ranging from 7.36 to 7.50. Okidi had the highest pH at 7.50, followed by Bibia at 7.46, and Elegu at 7.36. Total organic matter content varied among the demining sites, with Elegu having the highest percentage at 1.25% w/w, followed by Okidi at 1.03% w/w, and Bibia at 0.96% w/w. Total organic carbon content was highest at Okidi at 0.59% w/w, slightly higher than Bibia at 0.56% w/w, while Elegu had the lowest at 0.37% w/w. The carbon-to-nitrogen ratio was highest at Elegu at 1.72, indicating the highest proportion of carbon relative to nitrogen. Bibia had a ratio of 1.50, and Okidi had the lowest at 1.11. Total nitrogen content in Okidi was highest at 0.54% w/w, followed by Elegu at 0.42% w/w, and Bibia at 0.37% w/w. Bibia exhibited the highest cationic exchange at 43.48 meq/100 g, significantly higher than Elegu at 26.09 meq/100 g and Okidi at 17.37 meq/100 g. Bibia had the highest clay content at 26%, followed by Elegu at 18%, and Okidi at 8%. Similarly, Bibia had the highest silt content at 14%, followed by Okidi and Elegu at 12%. To the contrary, Okidi had the highest sand content at 80%, followed by Elegu at 70%, and Bibia at 60%. The textural class varied among the sites: Okidi was loamy sand, while Bibia and Elegu were sandy clay loam and sandy loam, respectively.

The physicochemical properties of the soil are shown in **Table 4**.

The immobilization of TNT (2,4,6-trinitrotoluene) and RDX (hexahydro-1,3,5-trinitro-1,3,5-triazine) in soil is influenced by several soil properties, including pH, organic matter content, cation exchange capacity (CEC), and particle size distribution. The data from Okidi, Bibia, and Elegu can provide insights into the potential for immobilization of these explosives at each site.

The pH values at Okidi (7.50), Bibia (7.46), and Elegu (7.36) are slightly alkaline. According to [Davis et al. \(2006\)](#), alkaline conditions can enhance the adsorption of TNT and RDX onto soil particles, reducing their mobility and bioavailability. The fate of explosive compounds in soil is dependent on soil pH. The amendment of soil pH to a value greater than 10.5 is an optimum condition for

**Table 4.** Soil physico-chemical properties.

Parameter	Okidi	Bibia	Elegu
pH	7.50	7.46	7.36
Total organic matter (% w/w)	1.03	0.96	1.25
Total organic carbon (% w/w)	0.59	0.56	0.37
Carbon: Nitrogen ratio	1.11	1.50	1.72
Total nitrogen (% w/w)	0.54	0.37	0.42
Cation exchange capacity (meq/100 g)	17.37	43.48	26.09
Particle size distribution: % Clay	8.00	26.00	18.00
% Silt	12.00	14.00	12.00
% Sand	80.00	60.00	70.00
Textural class	Loamy sand	Sandy clay loam	Sandy loam

decomposing explosives (Martin et al., 2012). Alkaline hydrolysis of soil by treatment with lime is effective in destroying TNT and RDX in solution (Martin et al., 2012). Davis et al. (2006) reported that 74% RDX and 83% - 99% TNT were degraded on lime treatment of explosive-contaminated soil. This suggests that all three sites may have a moderate capacity to immobilize these explosives due to their pH levels.

Soil organic matter is one of the most important soil constituents that significantly influences the sorption of organic compounds (Ran et al., 2007; Zhang & Chen, 2009). Organic matter and organic content play crucial roles in the adsorption of explosives. Elegu with the highest organic content (1.25% w/w) and Okidi with the highest total organic carbon (0.59% w/w) may have a greater capacity to immobilize TNT and RDX through adsorption onto organic matter. Therefore, Bibia, with the lowest organic matter and carbon content, may have a reduced capacity for immobilization. High amounts of soil organic matter are attributed to bacteria's decomposition of plant or animal residues by bacteria (Guo et al., 2020). The soil clay content, with the Okidi site having the lowest value, ranged from 8.00 to 26.00 percent w/w. The silt and sand in soil from the demining sites ranged from 12.00 to 14.00 and 60.00 to 80.00, respectively. The composition and texture of the soil are influenced by factors that include the microbial biomass and plant and animal residues, all of which have a high impact on the fate of the explosive compounds in the soil (Larson et al., 2008).

CEC measures the soil's ability to retain positively charged ions. While TNT and RDX are not cations, a higher CEC can indicate a greater capacity for adsorption and retention of various compounds. The main factors known to affect the values of CEC of soil include soil clay content, pH level and soil organic matter (Broomandi et al., 2020). Soil with a higher CEC value strongly adsorbs organic pollutants, thereby decreasing the bioavailability (Brannon et al., 2002; Chappell, 2011). Bibia with the highest CEC (43.48 meq/100 g) may have a higher potential for immobilizing TNT and RDX compared to Okidi (17.37 meq/100 g) and Elegu

(26.09 meq/100 g).

Also, soil texture affects the mobility of contaminants. Bibia with higher clay content (26%) and lower sand content (60%) may have better retention of TNT and RDX due to the higher surface area and adsorption capacity of clay particles. Okidi, with the highest sand content (80%), may have lower retention and higher mobility of these explosives. Elegu with intermediate clay (18%) and sand content (70%) may have moderate retention capacities.

Hence, the soil properties at Okidi, Bibia, and Elegu suggest varying capacities for the immobilization of TNT and RDX. Bibia, with its higher CEC and clay content, may offer the best conditions for immobilization, reducing the mobility and bioavailability of these explosives. Elegu, with higher organic matter and balanced texture, also shows potential for effective immobilization. Okidi, with higher sand content and lower CEC, may have the least capacity for immobilization, potentially leading to higher mobility of TNT and RDX. These insights highlight the importance of site-specific soil management practices to mitigate the environmental impact of explosive contaminants and enhance soil health and safety.

### 3.3. Physicochemical Properties of Rice Husk Biochar

The pH of the RHB was 9.70, indicating a highly alkaline nature. This higher pH can influence the chemical stability and immobilization of contaminants like TNT and RDX. The electrolytic conductivity of the biochar was at 2.54 dS/m, suggesting a moderate level of soluble salts. This property can affect the mobility and bioavailability of ions in the soil. The biochar contains 28.10% organic carbon, which is significantly high. This high organic carbon content can enhance the adsorption capacity of the biochar for organic contaminants such as TNT and RDX. The CEC of biochar was 34.67 meq/100 g, indicating a high capacity to retain and exchange cations. This property is crucial for the immobilization of contaminants, as it can enhance the retention of TNT and RDX in the soil. The elemental composition of the RHB was mainly 2.72% (Ca), 2.41% (Mg), 3.74% (Na), 4.22% (K), 3.88% (Si), and 3.43% (P). These elements contribute to the biochar's overall nutrient profile and chemical properties influencing its interaction with soil contaminants.

The physicochemical properties of the rice husk biochar are shown in **Table 5**.

The properties of RHB suggest it is highly effective for the immobilization of TNT and RDX explosives in soil. The biochar's high pH (9.70) can enhance the chemical stability of TNT and RDX, reducing their mobility and bioavailability in the soil. Alkaline conditions can promote the adsorption of these explosives onto the biochar, thereby immobilizing them. The high organic carbon content (28.10%) significantly increases the adsorption capacity of the biochar for organic contaminants. Organic carbon provides numerous binding sites for TNT and RDX, facilitating their immobilization.

The high CEC (34.67 meq/100 g) indicates that the biochar can effectively retain and exchange cations, which is beneficial for the immobilization of TNT

**Table 5.** Physico-chemical properties of biochar.

Parameter	Value
pH (water)	9.70
Electrolytic conductivity (dS/m)	2.54
Organic carbon (%)	28.10
Cation exchange Capacity (CEC) (meq/100 g)	34.67
Ca (%)	2.72
Mg (%)	2.41
Na (%)	3.74
K (%)	4.22
Si (%)	3.88
P (%)	3.43

and RDX. A higher CEC enhances the biochar's ability to adsorb and retain these contaminants, preventing their leaching into underground water. The presence of elements such as calcium, magnesium, sodium, potassium, silicon, and phosphorus contributes to the overall effectiveness of the biochar in immobilizing contaminants. These elements can interact with RDX and RDX, enhancing their adsorption and reducing their mobility in the soil. So RHB, with its high pH, organic carbon content, and cation exchange capacity, can be highly effective for the immobilization of TNT and RDX explosives in soil. Its properties suggest it can significantly reduce the mobility and bioavailability of these contaminants, making it a valuable amendment for contaminated soils. Using RHB can contribute to the remediation of explosive-contaminated sites, enhancing soil health and safety.

The physical properties of RHB are generally uniform regardless of the production methods (Kulkarni et al., 2014). RHB usually has a fine structure, is odourless, has a specific gravity of 2.3, is irregular in structure, is grey, and is <45 mm in size (Kulkarni et al., 2014). The morphology of RHB has been reported by Claoston et al. (2014) to be a honeycomb-like structure with many pores on the surface when pyrolyzed at 500°C. RHB pyrolyzed at 350°C has been reported to have pores that are not fully developed, while the regular pattern of pores is destroyed for RHB pyrolyzed at 650°C (Claoston et al., 2014). Pyrolysis temperature and residence time are key yield and physicochemical properties (Karam et al., 2022). Jia et al. (2018) reported the yield of RHB at 37.52 %w/w and 23.32 %w/w for pyrolysis temperatures of 350°C and 700°C on the rice husk, respectively, implying that the yield of RHB decreases with an increase in pyrolysis temperature. These findings are similar to the studies done by Claoston et al. (2014) and Nwajiaku et al. (2018). Pyrolysis of rice husks at longer residence time yielded lower amounts of RHB (Claoston et al., 2014; Jia et al., 2018).

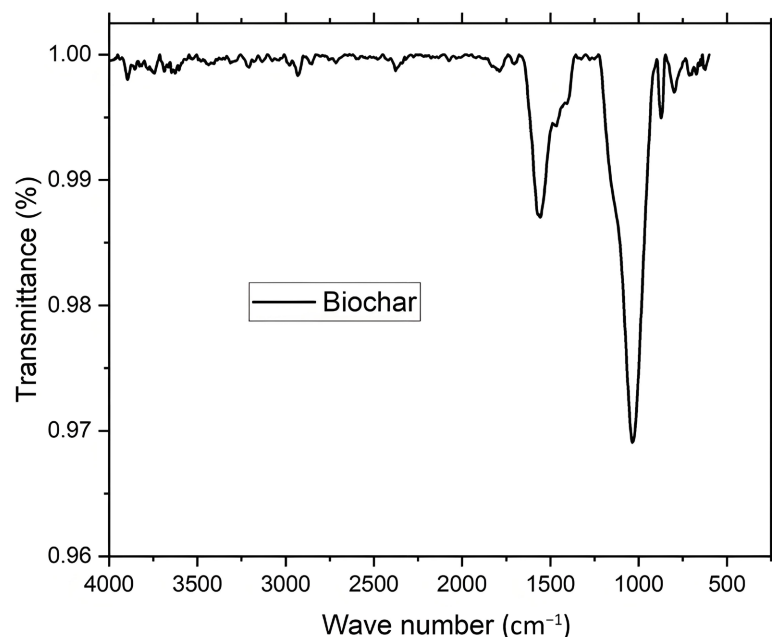
### 3.4. Fourier Transform Infra-Red (FT-IR) Spectrum of RHB

The Fourier Transform Infrared Spectroscopy (FTIR) analysis of RHB revealed

the following vibrational characteristics: 1550 - 1650  $\text{cm}^{-1}$  (N-H) bend range indicates the presence of amine groups, which can interact with nitro groups in TNT and RDX, potentially aiding their immobilization. 1000 - 1300  $\text{cm}^{-1}$  (Si-O-Si stretch) is characteristic of silicate structures, suggesting the presence of silica in the biochar. Silica can enhance the adsorption capacity of the biochar for organic contaminants, and the 840 - 890  $\text{cm}^{-1}$  (Si-H stretch) range indicates the presence of silicon-hydrogen bonds, which can contribute to the structural stability of the biochar and its interaction with contaminants.

FTIR data provides valuable insights into the potential of RHB for the immobilization of TNT and RDX explosives in soil. N-H bending vibrations suggest that amine groups are present in the biochar. These functional groups can form hydrogen bonds with the nitro groups in TNT and RDX, enhancing their adsorption onto the biochar surface. This interaction can significantly reduce the mobility and bioavailability of these explosives in the soil.

The FTIR spectrum of RHB is shown in **Figure 4**. The functional groups observed in the FTIR spectrum of RHB are shown in **Table 6**.



**Figure 4.** FTIR spectrum of Rice Husk Biochar.

**Table 6.** Functional groups observed in the FTIR spectrum of RHB.

Wave number ( $\text{cm}^{-1}$ )	Vibrational characteristics
1550 - 1650	N-H bend
1000 - 1300	Si-O-Si stretch
840 - 890	Si-H stretch

The Si-O-Si stretching vibrations indicate the presence of silicate structures

within the biochar. Silicates are known for their high surface area and adsorption capacity, which can enhance the immobilization of organic contaminants like TNT and RDX. The presence of these structures suggests that the biochar can effectively adsorb and retain these explosives, preventing their leaching into underground water.

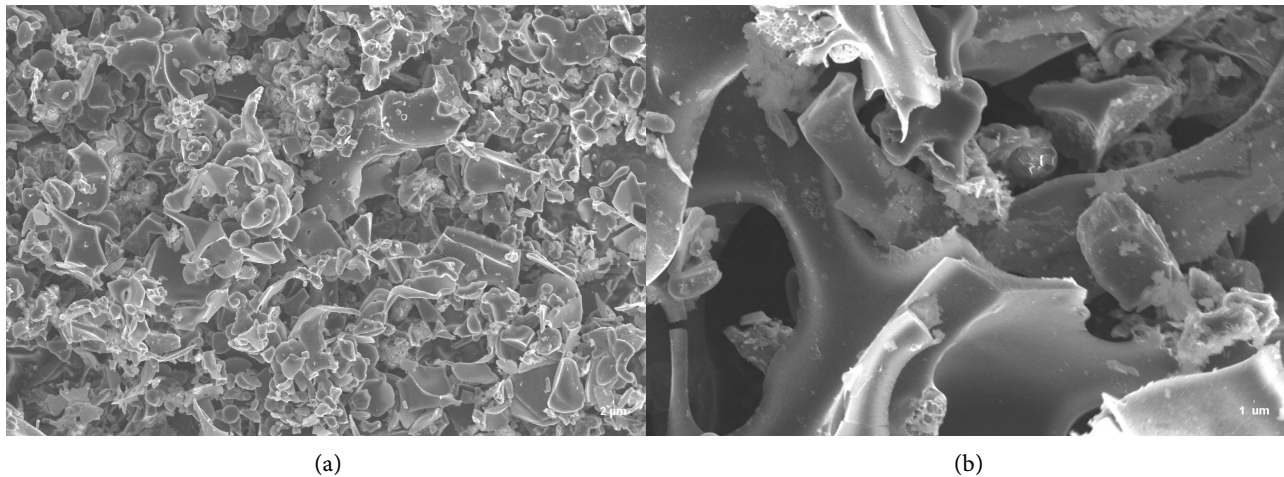
The Si-H stretching vibrations indicate the presence of silicon-hydrogen bonds, which contribute to the structural stability of the biochar. The stability is crucial for maintaining the integrity of the biochar in the soil environment, ensuring its long-term effectiveness in immobilizing the organic explosive contaminants. Si-H bonds also suggests that the biochar can interact with soil minerals, further enhancing its adsorption capacity.

### **3.5. Scanning Electron Microscope (SEM) Morphology of Rice Husk Biochar (RHB)**

The SEM images show that the RHB retains some of the original plant cell structures despite the high pyrolysis temperature. The cell walls appear to be well-preserved, indicating that the biochar maintains its structural integrity. The tissue morphology of the RHB displays a porous network with interconnected voids. These voids are remnants of the plant's vascular system, which has been carbonized during pyrolysis. The biochar exhibits a high density of micropores and mesopores. These pores are crucial for the adsorption of contaminants, providing a large surface area for interaction with TNT and RDX molecules. The surface morphology was seen through the Scanning Electron Microscope (SEM). The SEM micrographs reveal the variety of shapes and the micropores on the surface of RHB. The pores are honeycomb-like with cylindrical in shape and are responsible for the absorption of the TNT or RDX from the contaminated soil. The surface morphology and number of RHB pores depend on pyrolysis temperature. The number of pores on RHB reduces with increased in pyrolysis temperature during production (Claoston et al., 2014). Higher pyrolysis temperature increases ash content and causes cracks and shrinkages on the surface of RHB (Claoston et al., 2014).

SEM morphology of RHB provides valuable information on its potential for the immobilization of TNT and RDX explosives in soil. Despite high pyrolysis temperatures, the preservation of biomass structures in RHB, suggests that the biochar maintains its mechanical strengths and stability. This structural stability is essential for the long-term immobilization of contaminants, as it ensures that the biochar remains effective over extended periods. The porous network observed in the SEM images indicates that RHB has a high surface area, which is beneficial for the adsorption of TNT and RDX. The interconnected voids and channels can trap and hold these explosive molecules, reducing their mobility and bioavailability in the soil.

The SEM of RHB reveals the details insights into its biomass structures, tissue morphology, and pore characteristics (Figures 5(a)-(b)).



**Figure 5.** (a) Scanning Electron Microscope micrograph of biochar at 2  $\mu\text{m}$ ; (b) SEM micrograph of biochar at 1  $\mu\text{m}$ .

This porous structure enhances the biochar's capacity to act as a physical barrier, preventing the leaching of contaminants into underground water. The high density of micropores and mesopores in RHB is particularly advantageous for the immobilization of TNT and RDX. These pores provide numerous explosives adsorption sites for the explosives, facilitating their retention within the biochar matrix. The large surface area associated with these pores increases the likelihood of interactions between the biochar and the contaminants, enhancing the overall effectiveness of the immobilization process.

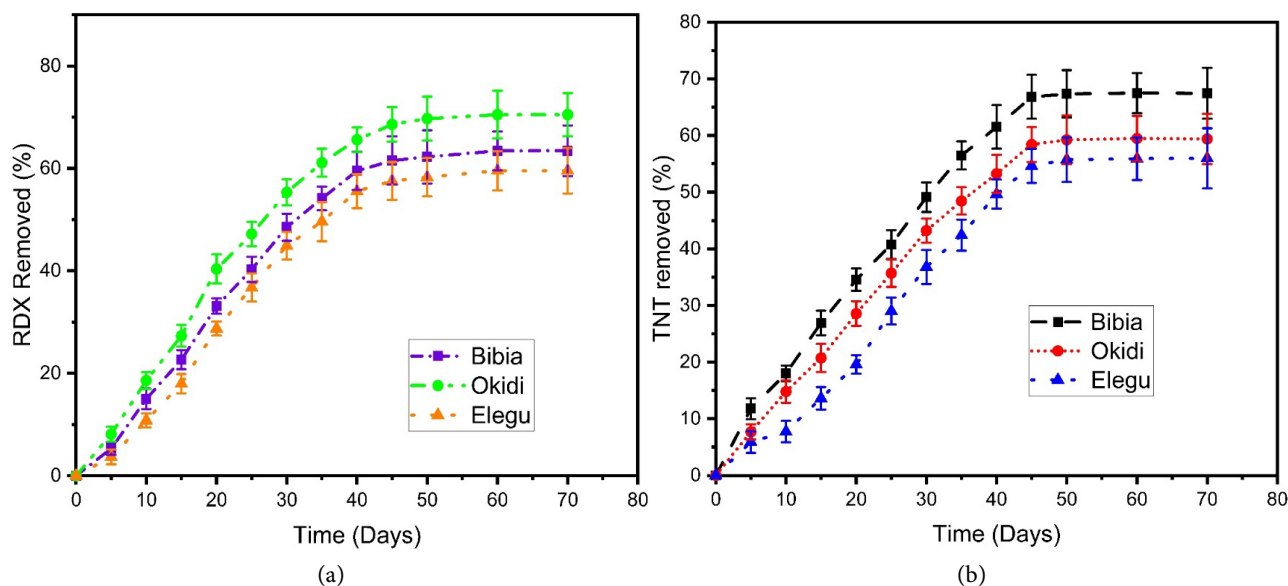
### 3.6. Immobilization Efficiency of Rice Husk Biochar

The graphs (**Figure 6**) illustrate the percentage of RDX 6(a) and TNT 6(b) removed over time for samples collected from three sites: Okidi, Bibia, and Elegu. The results from Okidi showed the highest percentage of RDX removal, reaching approximately 70% by day 50 - 60. Bibia follows closely behind Okidi, with slightly lower removal percentages throughout the experiment. Elegu exhibits the lowest RDX removal, achieving 50% by day 50 - 60. All samples demonstrate a rapid increase in RDX removal from day 0 to around day 20 - 30, after which the removal rate slows down and plateaus around day 50 - 60. Similarly, for TNT removal, results from Bibia indicated the highest percentage of TNT removal, reaching 70% after approximately 50 days. Okidi reached around 60% TNT removal after approximately 50 days, while Elegu exhibited the lowest percentage of TNT removal, reaching around 50% after approximately 50 days. Like RDX, all three samples demonstrated an initial increase in TNT removal over time before plateauing.

The results indicated that biochar in all three sites' samples effectively removes both RDX and TNT over time, but their efficiencies vary. This variation can be attributed to differences in soil properties such as pH, organic matter content, cation exchange capacity, and particle size distribution, which influence the immobilization of TNT and RDX. Treatment from Okidi's superior performance in

RDX removal suggests that its soil properties are highly conducive to the immobilization of explosives. Factors such as high organic carbon content and favourable pH may enhance the adsorption and retention of RDX molecules.

The amount of RDX and TNT immobilized by the rice husk biochar from soil obtained from the demining sites is shown in **Figures 6(a)-(b)**.



**Figure 6.** (a) Amount of RDX immobilized from soil obtained from the demining sites; (b) Amount of TNT immobilized from soil obtained from the demining sites.

The rapid initial increase in RDX removal indicates effective initial adsorption, likely due to the availability of numerous binding sites and favourable soil conditions. Samples from Bibia's performance, while slightly lower than Okidi, still showed significant RDX removal. The soil's high cation exchange capacity and clay content may contribute to its ability to retain RDX. The consistent removal rate suggests that Bibia's soil properties support sustained adsorption and immobilization of RDX over time. The observed sample from Elegu with lower RDX removal efficiency may be due to less favourable soil properties for adsorption. Factors such as lower organic carbon content and different particle size distribution could reduce the soil's capacity to immobilize RDX. One-way Analysis of variance reveals that there is no sufficient evidence to say that there is a statistically significant difference between the mean amounts of RDX and TNT immobilized from the soil samples obtained from the three study sites ( $P > 0.05$ ).

Adsorption involves the interaction between the adsorbate (TNT and RDX) and the adsorbent (soil particles and biochar). TNT molecules can interact with the soil particles through hydrogen bonding, Vander waals forces, and chemical bonding. The presence of functional groups such as amines (N-H bond) in biochar can enhance the adsorption of TNT by forming hydrogen bonds with its nitro groups. RDX molecules can be adsorbed onto soil particles through similar mechanisms. The high surface area provided by the micropores and mesopores in

biochar enhances the adsorption of RDX. In biochar, silicate structures (Si-O-Si stretch) can also contribute to the adsorption process. The adsorption of TNT by RHB involves a complex mechanism, with the Si-O-Si bond playing a vital role in  $\pi$ - $\pi$  interactions with the aromatic rings of TNT. In addition, the oxygen-containing functional groups on the biochar's surface act as electron donors, thereby forming hydrogen bonds with the  $-\text{NO}_2$  groups of TNT. Si-H bond adsorbs TNT through weak electrostatic interactions with the nitro groups of TNT. The N-H bonds on the RHB surface participate in hydrogen bonding with oxygen atoms in the  $-\text{NO}_2$  groups of TNT. RDX can be adsorbed in perpendicular or parallel orientations relative to the siloxane surface. The chemical can adopt different conformations, which have a greater influence on the adsorption on the Si-O-Si and Si-H bonds. RDX adsorption on Si-O-Si and Si-H bonds of RHB surface occurs through hydrogen bonding and chemical bonds with silicon and oxygen atoms (Xu et al., 2023).

Lingamdinne et al. (2015) reported that the adsorption of RDX on RHB occurs via electrostatic interaction and chemisorption through the nitrogen-oxygen bonds. The efficiency of remediation can be improved by treatment of the RHB with alkaline reagents, which activate the internal structure and increase surface area to improve adsorption and fixation capacities (Amen et al., 2020). Microbial degradation of RDX may occur during the immobilization process, hence reducing the amount of the explosive compound in the soil. Sharma et al. (2021) reported that a 6 mL inoculation of a microbial consortium of *Paenibacillus aestuarii* and *Arthrobacter subterraneus* was able to degrade 80.4% of RDX within 12 days. These environmentally friendly and inexpensive approaches to the remediation of RDX-contaminated soils should never be ignored.

TNT adsorbs to RHB via electrostatic interaction and chemisorption by the  $\pi$ - $\pi$  bonds and the nitrogen-oxygen bonds (Lingamdinne et al., 2015). Microbial degradation of TNT may also occur during the immobilization process, hence reducing the amount of the explosive compound in the soil. *Pantoe* sp. BJ2 can degrade 97.85% of  $100 \text{ mg}\cdot\text{L}^{-1}$  of TNT within 24 hours (Li et al., 2024). A *Pseudomonas* strain isolated from soil around an explosive factory grew with TNT as its sole source of nitrogen (Duque et al., 1993). *Enterobacter cloacae* strain PB2, which had previously been isolated due to its ability to utilize nitrate esters, has been found to slowly degrade TNT in soil by utilizing it as its sole source of nitrogen (Esteve-Núñez et al., 2001). Bacteria and fungi can be used for the bioremediation of explosive-contaminated soils. Bioremediation of explosive TNT in soil using fungi has aroused the interest of environmental scientists. The fungus *Phanerochaete chrysosporium* (wood white rot fungus), litter decay fungus, and white rot fungi have been reported by Esteve-Núñez et al. (2001) to bioremediate TNT.

The RHB prepared and used for the immobilization studies was not treated with acid, alkali, or metal. The physicochemical parameters in future studies need to be altered by treatment of the RHB before immobilization studies to improve the efficiency for the immobilization of TNT or RDX from the explosive-contami-

nated soil. Factors of pyrolysis temperature and retention time on the production of the RHB for the immobilization of TNT and RDX also have to be established to produce biochar with a higher Immobilization efficiency. These conditions for the production of RHB greatly influence on the porosity, functional groups, and mineral content, which in turn affect the immobilization efficiency. It has been reported by [Kulkarni et al. \(2014\)](#) that RHB contains  $\text{Al}_2\text{O}_3$ ,  $\text{Fe}_2\text{O}_3$ , and  $\text{SiO}_2$ . The immobilization efficiency of the RHB can be improved by physical modification during the charring, in which some pores are made open and activation processes, increase the pores and the specific surface area on the biochar. The microbial modification uses microorganisms, which act together with RHB to improve the adsorption capacity and remediation effectiveness for target pollutants ([Bhatnagar et al., 2013](#)). The microbial modification also improves the physicochemical properties of RHB, such as structure, redox potential, and pH ([Li et al., 2023](#)). Chemical modification approaches include functional group, metal, acid, and alkali modifications. The data generated in the present study show the effectiveness of RHB in the immobilization of TNT and RDX from soil. Since rice husk is cheap and readily available, it can be pyrolyzed, and its biochar can be utilized for the immobilization of explosive-contaminated soils.

#### 4. Conclusion

The study investigated RHB's potential for immobilizing TNT and RDX explosives in soil. FTIR analysis reveals that RHB contains functional groups and structures beneficial for this purpose. Specifically, N-H bending vibrations suggest potential hydrogen bonding with nitro groups, while Si-O-Si and Si-H stretching vibrations indicate high adsorption capacity and structural stability. These characteristics enhance RHB's ability to immobilize TNT and RDX explosives in soil.

SEM analysis further supports the RHB's suitability for immobilizing TNT and RDX. The biochar retains preserved biomass structures, porous tissue morphology, and a high density of micropores and mesopores, all of which contribute significantly to its adsorption capacity. These properties make RHB a promising amendment for remediating explosive-contaminated soils, thereby improving soil health and safety.

The study highlights that soil properties significantly influence the immobilization of TNT and RDX. Among the soils analyzed, Okidi's soil shows high potential for effective immobilization, followed by Bibia and Elegu. The varying efficiencies of TNT and RDX removal across different soil samples underscore the need for site-specific soil management practices. Key soil properties such as pH, organic matter content, cation exchange capacity, and particle size distribution play crucial roles in the adsorption and immobilization of contaminants. Therefore, soil amendments should be tailored to the specific characteristics of each site to optimize remediation efforts.

The study also demonstrates that biochar, particularly RHB, is highly effective in immobilizing TNT and RDX. The high pH, organic carbon content, and cation

exchange capacity of biochar enhance its adsorption capacity. Incorporating biochar into contaminated soils can improve the retention of explosives, reducing their mobility and bioavailability. This practice is particularly beneficial in areas with high contamination levels. The plateau observed in the removal of RDX and TNT suggests an optimal period of biochar's effectiveness. Continuous monitoring of soil conditions and contaminant levels is essential for long-term success. Regular maintenance, including reapplication of biochar or other amendments, may be necessary to sustain contaminant immobilization. These findings emphasize the importance of tailored soil management practices and the potential of biochar as a valuable tool for environmental remediation.

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## Conflicts of Interest

The authors declare no conflicts of interest regarding the publication of this paper.

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