Stabilization of organic amendments and remediation of pesticide-contaminated soils through microbial inoculation and complementary techniques.

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Abstract

Soil contamination by pesticides is a growing environmental concern due to its adverse effects on ecosystems and human health. This study seeks to address the remediation of pesticide-contaminated soil by stabilizing organic amendments and applying microbial inoculation. The experiment was conducted in four phases: (1) stabilization and conditioning of the organic amendment, (2) analysis of the eroded soil, (3) contamination of the eroded soil with pesticides, and (4) remediation of the contaminated soil using microbial inoculant and complementary techniques such as palm biochar, zeolite, and clay soil. The results indicate that the application of organic amendments and microbial inoculants significantly improved soil parameters including organic matter content, carbon and nitrogen levels, and porosity. The remediation process also demonstrated a reduction in pesticide concentration and an increase in soil fertility. This study highlights the effectiveness of organic amendments in remediation processes and microbial inoculation as sustainable techniques for soil remediation, offering a viable solution for the recovery of pesticide-contaminated soils.

Keywords

soil remediation, pesticides, organic amendments, microbial inoculation, soil fertility.

Introduction

Soil contamination by pesticides is an environmental problem of utmost importance, particularly in agricultural areas where excessive use of chemical pesticides has led to degradation of soil quality and alteration of ecosystems. Pesticides, such as imidacloprid, are widely used in agriculture due to their effectiveness in pest control. However, their persistence in the environment creates significant risks to soil health, water quality and biodiversity (Gavrilescu, 2005). Accumulation of pesticides and insecticides in soil can lead to inhibition of microbial activity, reduction of soil fertility and contamination of groundwater, ultimately affecting human health and food security. (Pimentel et al., 2005).

Recent studies have mentioned the need for sustainable remediation techniques to address soil contamination; thus, organic amendments such as compost and biochar have been shown to improve soil structure, increase microbial activity and reduce the bioavailability of contaminants (Lehmann et al., 2011). Furthermore, microbial inoculation has emerged as an important approach for pesticide biodegradation, as certain microorganisms can metabolize these compounds into less toxic forms (Singh et al., 2020). The combination of organic amendments and microbial inoculation offers a holistic approach to soil remediation, favoring both physical and biological recovery of contaminated soils.

This study aims to evaluate the efficacy of organic amendments and microbial inoculation in remediating pesticide-contaminated soil. The experimentation was carried out in four phases: (1) stabilization and conditioning of the organic amendment, (2) analysis of the eroded soil, (3) contamination of the eroded soil with pesticides, and (4) remediation of the contaminated soil using microbial inoculants.

Methods

Stabilization of organic amendments is a critical step in soil remediation processes, as it ensures the maturity and stability of organic matter prior to application. In this study, the "Bocashi" method was used for organic amendment stabilization. This method, which originated in Japan and has been widely adopted in Latin America, involves the aerobic decomposition of organic materials through the action of microorganisms (Gómez et al., 2010). The Bocashi method is particularly effective because it accelerates the decomposition process, reduces the volume of organic residues, and produces a stable amendment rich in nutrients and beneficial microorganisms.

Several studies have demonstrated the success of the Bocashi method in improving soil fertility and structure. For example, a study by Gómez et al. (2010) found that application of Bocashi compost significantly increased soil organic matter, nutrient availability, and microbial activity in degraded soils. Similarly, a case study in Colombia reported that the use of Bocashi compost improved soil fertility and crop yield in pesticide-contaminated agricultural fields (Rojas et al., 2015)

Remediation of pesticide-contaminated soil was achieved by applying microbial inoculant and complementary techniques such as biochar and clay soil. Microbial inoculation is a well-established technique for biodegradation of

organic contaminants, including pesticides. Certain microorganisms such as Pseudomonas and Bacillus species have been shown to degrade pesticides such as imidacloprid into less toxic compounds (Singh et al, 2020). In this study, microbial inoculum was prepared by culturing microorganisms from the stabilized organic amendment and diluting them in peptone water. The inoculum was then added to the contaminated soil, along with 100 mL of Degradex (0.3 g/L) and 100 mL of Safersoil (0.6 g/L).

The use of biochar as a complementary technique has also been widely studied for its high capacity to absorb organic contaminants and improve soil properties. Biochar is a carbon-rich material produced through pyrolysis of organic waste and its porous structure provides a habitat for microorganisms while adsorbing contaminants (Agegnehu et al., 2016). In this study, palm biochar was added to contaminated soil to enhance pesticide adsorption and promote microbial activity.

A case study by Ortega et al. (2023) demonstrated the effectiveness of biochar in remediating pesticide-contaminated soil. The study found that the application of biochar reduced the concentration of pesticides in soil by 60% and improved soil fertility by increasing organic matter and nutrient availability. Similarly, a study by Coria (2006) highlighted the use of clay soil as an effective adsorbent for organic contaminants including pesticides.

The following table summarizes the procedures and techniques used in each phase of the experiment:

Table 1. Procedures to be carried out

PHASE	PROCEDURE			PHASE DEVELOPMENT
Stabilization	**Box 1**			For the stabilization process, a combination of raw materials (sludge and compost) was used due to the richness of nitrogen. The treatment focused on the evolution of organic matter as an indicator of the maturity of the amendment.
	Raw materials	**Weight**	**Prepared mix**	
	Sludge	400 g	The mixture was then subjected to drying in an oven at 105°C for 24 hours, followed by calcination in a muffle furnace at 500°C for 2 hours.	
	FORM	100 g		
	Pig slurry	100 mL		
	Nitrogenous sludge	25 g		
	Mature compost	500 g		
	Liquid nitrogen	100 mL		
	Rice husk	10 g		
	Box 2			
	Raw materials	**Weight**	**Prepared mix**	
	Sludge	300 g	The mixture was then subjected to drying in an oven at 105°C for 24 hours, followed by calcination in a muffle furnace at 500°C for 2 hours.	
	FORM	200 g		
	Pig slurry	200 mL		
	Nitrogenous sludge	50 g		
	Mature compost	400 g		
	Liquid nitrogen	150 mL		
	Rice husk	15 g		
	Box 3			
	Raw materials	**Weight**	**Prepared mix**	
	Sludge	200 g	The mixture was then subjected to drying in an oven at 105°C for 24 hours, followed by calcination in a muffle furnace at 500°C for 2 hours.	
	FORM	300 g		
	Pig slurry	300 mL		
	Nitrogenous sludge	100 g		
	Mature compost	300 g		
	Liquid nitrogen	200 mL		
	Rice husk	20 g		
Eroded soil	Soil analysis			The eroded soil was analyzed to determine its initial properties, including moisture content, organic matter, carbon, nitrogen, pH, and electrical conductivity (EC). The soil was dried in an oven at 105°C for 24 hours, and the organic matter content was determined by calcining in a

		muffle furnace at 500°C for 2 hours.
Soil contamination	Contamination with 2.4 mL of insecticide (imidacloprid)	The eroded soil was contaminated with pesticide, and its initial properties were determined, including moisture content, organic matter, carbon, nitrogen, pH and electrical conductivity (EC). The soil was dried in an oven at 105°C for 24 hours, and the organic matter content was determined by calcining in a muffle furnace at 500°C for 2 hours.
Remediation of contaminated soil	Application of stabilized amendment in different proportions (from best to worst) Application of microbial inoculum + Degradex (0.3 g/L) + Safersoil (0.6 g/L) Application of biochar and clay soil	Amendments were added in proportions ranging from the best (highest proportion) to the worst (lowest proportion), microbial inoculum, and conditioners to the contaminated soil to promote pesticide biodegradation; additionally, complementary techniques (biochar and clay soil) were applied to improve pesticide adsorption and soil structure.

Results

Stabilization of organic amendments

Three stabilization treatments were evaluated using the Bocashi method with different proportions of components. Table 2 presents the results of the experiment, which show that treatment 3 performed best overall, with the highest germination rate (84.38%) and a C/N ratio (23.79) within the acceptable range for agricultural soils, along with a pH close to neutrality (7.7).

Table 2. Physicochemical and biological parameters of Bocashi stabilization treatments

Parameter	Treat 1	Treat 2	Treat 3
Humidity (%)	47.67	47.68	46.69
Organic matter (%)	35.62	36.08	34.16
Carbon (%)	37.96	46.34	40.64
Nitrogen (%)	1.78	1.80	1.71
C/N ratio	21.32	25.60	23.79
pH	8.4	8.1	7.7
Conductivity (us/cm)	2983	2872	2961
Bulk density (g/mL)	0.65	0.63	0.87
Porosity	0.37	0.38	0.15
Germination (%)	70	78.13	84.38

The variation in the parameters studied, which show better results in treatment 3, are shown in Figure 1.

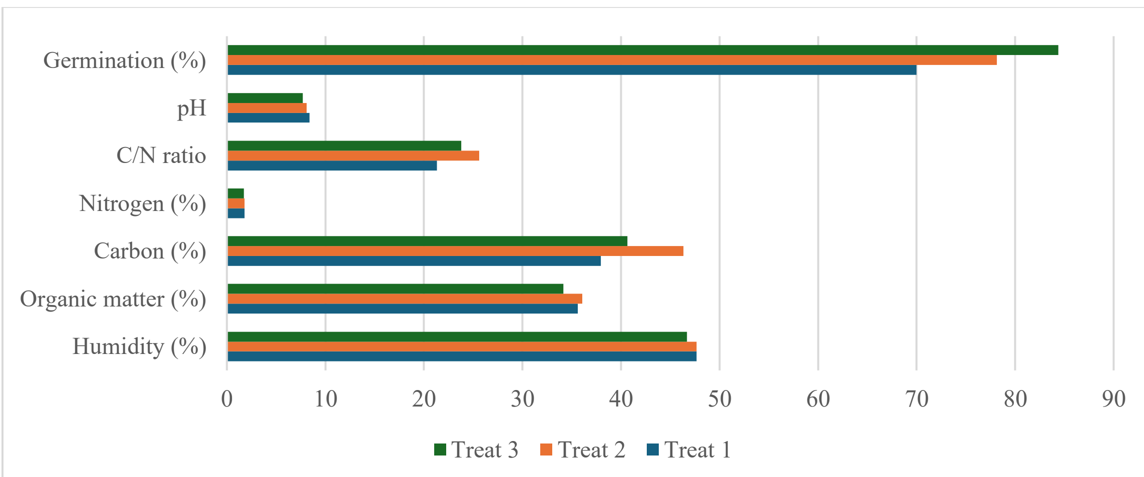

Figure 1. Variation in the physicochemical parameters relevant to the stabilization of organic amendments. Treatment 3 (green bar) shows better results and greater variation.

Characterization of eroded soil

The eroded soil showed low fertility and adverse conditions for microbial and plant development, as shown in Table 3. Noteworthy are its low organic matter (3.41%), nitrogen content (0.17%) and a very low germination index (1.93%), confirming its state of degradation.

Table 3. Initial characterization of eroded soil

Parameter	Amounts
Humidity (%)	14.36
Organic matter (%)	3.41
Carbon (%)	1.94
Nitrogen (%)	0.17
pH	8.2
Conductivity (us/cm)	220.3
Bulk density (g/mL)	0.80
Porosity	0.21
Germination (%)	1.93
Microbial communities	Gram-negative

Remediation of soil contaminated with imidacloprid

Application of microbial inoculum

The application of microbial inoculum showed a progressive reduction in COD (from 1140 mg/L to 367 mg/L) over three controls, indicating active biodegradation of the pesticide. However, turbidity remained high (>1000 NTU), suggesting the presence of colloidal material or microbial biomass in suspension (Table 4).

Table 4. Evolution of leachate quality parameters during remediation with inoculum

Parameter	Start	Control 1	Control 2	Control 3
COD (mg/L)	1140	554	373	367
Turbidity (NTU)	>1000	>1000	>1000	>1000
pH	7.3	8.3	7.4	7.1
Conductivity (us/cm)	385	511	483	402

Figure 2 illustrates the progressive reduction in COD during the bioremediation process, with a decrease of 67.8% between start-up and final control.

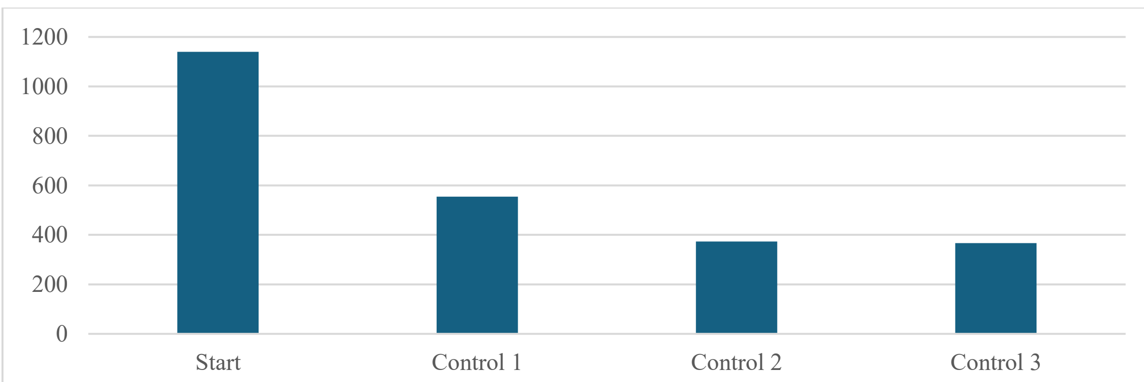

Figure 2. Progressive reduction of COD during the bioremediation process. A significant decrease in the parameter is observed between the beginning and the final measurement (control 3).

Application of stabilized amendments

The application of stabilized amendments significantly improved soil properties, increasing organic matter, carbon, and nitrogen, and moderating the C/N ratio (Table 5).

Table 5. Effect of applying stabilized amendments to contaminated soil.

Parameter	Contaminated soil	Soil + Amendment
Humidity (%)	10.22	41.04
Organic matter (%)	5.56	9.70
Carbon (%)	3.17	5.53
Nitrogen (%)	0.28	0.48
C/N ratio	11.40	11.40
pH	7.3	8.2
Conductivity (us/cm)	385	801
Porosity	0.14	0.20

Effect of complementary techniques

The addition of biochar and clay soil to soil previously treated with amendments and inoculum further improved fertility parameters, with increases in organic matter (12.64%), carbon (7.21%) and porosity (0.27), confirming the synergistic effect of these techniques (Table 6).

Table 6. Further improvement of pesticide-contaminated soils using complementary techniques

Parameter	Soil + Amendment	Soil + Amendment + Techniques
Humidity (%)	41.04	28.88
Organic matter (%)	9.70	12.64
Carbon (%)	5.53	7.21
Nitrogen (%)	0.48	0.63
C/N ratio	11.40	11.40
Bulk density (g/mL)	0.82	0.74
Porosity	0.20	0.27
Conductivity (us/cm)	801	950.8

Discussion

The results show that the stabilization of organic amendment through the implementation of the Bocashi method is effective to produce mature and stable material with physicochemical and biological parameters suitable for use in eroded or contaminated soils. As shown in Figure 1, treatment 3 stood out for its high germination rate (84.38%), suggesting a low level of phytotoxicity and adequate compost maturity (Gómez et al., 2010). In addition, the C/N ratio obtained is close to the ideal range reported in the literature for stable amendments (Bernal et al., 2009), which favors the availability of nitrogen for plants and other microorganisms.

The characterization of the eroded soil confirmed its low agricultural capacity, with minimal amounts of organic matter (3.41%), nitrogen (0.17%), and an extremely low germination index (1.93%). These conditions of soil degradation justify the need for immediate intervention with the use of amendments and remediation techniques to restore its ecological functionality, especially in soils that have been affected by intensive agriculture (Pimentel et al., 2005).

During the bioremediation phase, the application of microbial inoculum to the soil showed a progressive reduction in COD (figure 2), from values of 1140 to 367 mg/L indicating active degradation of imidacloprid (the main component of the pesticide). This result is consistent with studies reporting the ability of microbial consortia, mainly of the genera Pseudomonas and Bacillus, to metabolize neonicotinoid pesticides (Singh et al, 2020; Cycón et al., 2017). In the same way, the addition of conditioners such as Degradex and Safersoil demonstrated an improvement in the bioavailability of the contaminant and stimulated microbial activity, as previously observed in agricultural soil bioremediation systems (Rojas et al., 2015).

The application of stabilized amendments (mainly in treatment 3) showed a significant improvement in soil fertility, increasing the content of organic matter, carbon, and nitrogen, favoring soil structure, moisture retention, and microbial activity, promoting the degradation of contaminants (Lehmann et al., 2011). Similarity, the increase in soil electrical conductivity can be attributed to the release of ions during the mineralization process of organic matter, a phenomenon documented in soil amended with compost (Wei et al., 2021).

The integration of complementary techniques such as biochar and clay soil showed a positive synergistic effect, improving soil porosity, carbon content, and cation Exchange capacity, supporting recent findings on the use of natural adsorbents in the remediation of pesticide-contaminated soils (Ortega et al., 2023; Coria, 2006).

The synergistic effect of complementary techniques is evident in Figure 3, which shows consecutive improvements in organic matter and carbon, as well as control over moisture percentages.

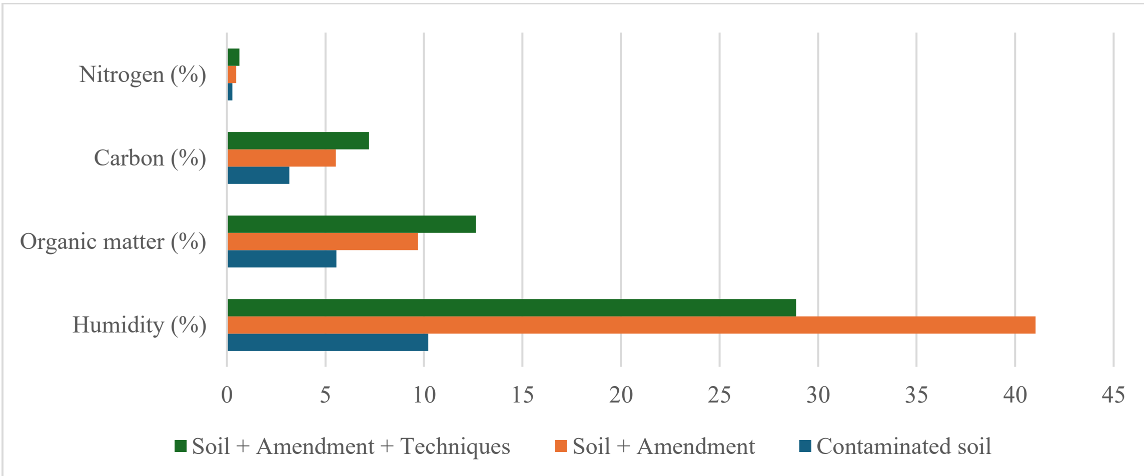

Figure 3. Variation in moisture, nitrogen, carbon, and organic matter in the bioremediation process, from contaminated soil to the addition of complementary techniques

This demonstrates the ability of biochar to absorb contaminants and provide a suitable habitat for degrading microorganisms (Lehmann et al., 2011), as well as the contribution of clay soil to the physical and chemical stabilization of the system (Ortega et al., 2023)

Finally, from an economic perspective, the Bocashi method and complementary techniques offer a more cost-effective alternative to chemical remediation. The production costs of Bocashi compost were estimated at $0.15 per kilogram using locally sourced materials (such as rice husks and manure), compared to $1.50 per kilogram for commercial bioremediation agents (Qambrani et al., 2017). Scaling up to field applications could reduce pesticide remediation costs by 60% to 70% on small farms, as demonstrated in similar case studies in Colombia (Rojas et al., 2015). Furthermore, the use of palm biochar, derived from agricultural waste, promotes the principles of the circular economy by valorizing waste and minimizing environmental impact. Challenges, such as soil types and climatic conditions, must be considered in pilot-scale trials to ensure scalability across diverse ecosystems.

Conclusions

The study demonstrates that stabilizing organic amendments using the Bocashi method is an effective technique for producing mature and stable materials, with Treatment 3 (200 g sludge + 300 g FORM + 300 mL slurry + 100 g nitrogenous sludge + 300 g mature compost + 200 mL liquid nitrogen + 20 g rice husks) showing the best overall performance, achieving a C/N ratio of 23.79 and 84.38% germination, indicating low phytotoxicity and high maturity.

The combined application of stabilized amendments and microbial inoculation proved to be an effective synergistic strategy for the remediation of soils contaminated with imidacloprid. A 67.8% reduction in COD and significant improvements in soil fertility parameters were observed: organic matter (74.5% increase), carbon (74.4%), and nitrogen (71.4%).

The integration of complementary techniques (palm biochar and clay soil) produced an additional synergistic effect, further improving porosity (35% increase), carbon content (30.4% additional), and cation exchange capacity.

This comprehensive approach offers a viable and sustainable solution for the recovery of agricultural soils contaminated with pesticides, simultaneously addressing contaminant remediation and soil fertility restoration, with potential application in sustainable agriculture systems.

While this study demonstrates the effectiveness of the proposed remediation strategy, future research should explore long-term field trials to assess the persistence of improvements over several crop cycles, adding phytoremediation with plants such as Sesbania cannabina, which have shown favorable results in saline-alkaline soils contaminated with pesticides (Gu et al., 2023; Tarla et al., 2020). In addition, research on scenarios with multiple contaminants, such as mixtures of neonicotinoids and herbicides, could broaden its applicability. These advances would further consolidate the approach for sustainable soil management in regions affected by pesticides.

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