



Bioremediation of Styrofoam-Contaminated Soil Using Mycorrhizal Fungi and Cattle Dung Manure to Sustain Soil Fertility Properties

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ABSTRACT

Sustainable agriculture relies on maintenance of soil health, but increasing production of plastics has resulted to polluting agricultural soils with polystyrene. The paper evaluated the effectiveness of mycorrhizal inoculation and cattle dung amendment to bio-restore Styrofoam polluted soil by assessing the impacts that these treatments produced on the qualities of the soil and cowpea (*Vigna unguiculata*) growth. The experimental design was entirely randomised; it was a factorial inoculation design, which consisted of mycorrhizal inoculation (10 g and 15 g), cattle dung (50 g and 100 g), and combinations of them. The height of the plants was followed over a five-week period, the physicochemical properties of the soils were analysed and the data were exposed to ANOVA and Duncan Multiple Range Test. Findings indicated that there were significant effects of treatment ($p \leq 0.05$). The greatest plant height (14.0 cm) and significant enhancement of soil organic matter (0.68%), available phosphorus (28.81 mg/kg) and cation exchange capacity (8.80 cmol/kg) were the products of the 50 g cattle dung treatment due to the absence of salinity stress. On the other hand, 100 g cattle dung led to slow germination and low growth (8.2 cm) because of initial phytotoxicity by high soluble salts and sodium concentration (0.79 cmol/kg). Inoculation of mycorrhizal alone at 10g gave moderately good growth (9.9 cm), which indicates increased nutrient mobilisation. The synergistic advantage of combining 10 g of mycorrhiza with 50 g of cattle dung was not found (7.8 cm), which indicates that there is no resource competition or nutrient saturation anymore. More importantly, in the combined 15 g mycorrhizacattle dung (100 g) the phytotoxicity was totally inhibited (0 cm) due to synergistic phytotoxicity of excessive salts, high exchangeable acidity (0.66 cmol/kg) and the displacement of sodium by cations. Medium levels of cattle dung (50 g) can thus be considered to be the most suitable option in terms of replenishing the soil with fertility and nourishing the growth of plants in the Styrofoam-contaminated soil, whereas higher application rates have adverse consequences. This paper gives a scientific method of recovery of plastic-polluted soils via selective organic modification.

Keywords: Organic Amendments, Mycorrhizal Associations, Bioremediation, Soil Restoration, Styrofoam Contamination.

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INTRODUCTION

Soil health is important to the terrestrial ecosystems because it constitutes a dynamic system, which provides vital services such as nutrient cycling, water filtration, and carbon sequestration. The different classes of microbes facilitate such functions by changing biological, chemical and physical characteristics of soil and microorganisms are indispensable in the breakdown of organic matter and recycles of nutrients [1]. Nevertheless, non-biodegradable pollutants persistence has turned out to be a growing challenge on the quality of the environment. Styrofoam is a trademarked brand of expanded polystyrene (EPS) which is a lightweight closed cell foam material that is made up of about 95-98 percent air and 2-5 percent polystyrene polymer. It has outstanding buoyancy, thermal insulation, and a low price of manufacture, which makes it widely used as packaging material, disposable food wrapping, and

construction insulation [2]. However, it has been widely used and it has produced immense accumulation on the environment. Styrofoam does not easily break down and its estimated environmental life of 500 to more than 1,000 years is due to its resistance to attack by the microbial enzymes as long polymer chains and rings forming aromatic structures comprise the styrofoam [3, 4]. Styrofoam changes the basic physical properties when it is added to soil. Styrofoam particles cause a reduction in the overall apparent density of the soil matrix because of their very low bulk density (1664 kg/m^3 as opposed to mineral soil $1200 - 1600 \text{ kg/m}^3$). At the same time, the porous and disordered form of fragmented Styrofoam increases the total soil porosity by forming more macropores and inter-particle voids. The effects of these physical changes are far-reaching in that reduced density leads to poor structural stability of the soil, and greater porosity increases preferential flow direction, which will accelerate water infiltration and free-surface flow of soluble nutrients especially nitrogen into the root zone. This has increased the rate of leaching disturbing the nitrogen cycle and hindering the acquisition of nutrients by plants [3, 4]. In addition to physical interference, Styrofoam has pernicious impacts on soil communities of microorganisms. The compound decreases the abundance and metabolism of major microorganisms in the soil that decompose organic matter, mineralize nutrients and maintenance of soil fertility. In particular, Styrofoam particles are able to adsorb microbial exoenzymes, prevent substrate accessibility, and form hydrophobic micro environments which inhibit microbial colonization and activity, which in turn inhibits degradation of organic materials and affects nutrient cycling processes [5].

Styrofoam is leaching in the environment, releasing toxic chemicals such as styrene monomers, which damage the soil organisms and agricultural systems especially in the Sudan Savanna area where the contamination of soil has hampered the growth of plants, and the safety of food [2]. The process of soil stabilisation, associated with enhancing physical, chemical, and biological stability of soil against degradation, soil structure, and ecosystem functions, is promoted by the arbuscular mycorrhizal fungi (AMF) that produce glomalin to adhere soil particles into stable aggregates, which minimises soil erosion and nutrient leaching. The AMF establish symbiotic relationships with plant roots where they promote organic contaminant rehabilitation, nutrient-absorption, and stabilisation of the soil structure [6]. The use of livestock manure, as well as the use of mycorrhizal inoculation, also increases soil restoration, and comparative research has shown that cattle manure that features a balanced carbon/to-nitrogen ratio is better at improving soil organic matter, microbial biomass and mycorrhizal colonization than poultry or swine manure, which tends to contain greater concentrations of soluble salts [7]. These organic amendments stabilise the important nutrients, promote healthy microbial populations, and all contribute to the increase in soil fertility and resilience to Styrofoam contamination [8]. High throughput sequencing methods gave a wide perspective of microbial interaction in the degradation of pollutants. Cattle Dung may be described as the undigested remnant of the eaten foodstuff that is excreted by grass eating bovine animal species. As a combination of faeces and urine in the ratio 3:1, it is primarily composed of lignin, cellulose, and hemicelluloses; 24 various minerals, including nitrogen, potassium and traces of sulphur, iron, magnesium, copper, cobalt, and manganese exist in it [9,10].

Cattle dung harbours a rich microbial diversity, containing different species of bacteria (*Bacillus* spp., *Corynebacterium* spp., and *Lactobacillus* spp.), protozoa, and yeast (*Saccharomyces* and *Candida*) [10], which makes them appropriate for microbial degradation of contaminants [11-13]. Understanding how bacteria and fungi cooperate is extremely helpful when using organic fertilizer in conjunction with mycorrhizal inoculants. Bacteria have a relatively large surface area and therefore are also able to capture soluble pollutants easily. Fungi, particularly mycorrhizal species, secrete cellular enzymes that decompose resilient compounds such as polystyrene, cellulose, and lignin. The enzymatic degradation accelerates the decomposition of persistent pollutants, rendering the combination of mycorrhizal inoculation and organic amendments an effective remediation strategy [14].

Concerns regarding Styrofoam pollution are prevalent; however, there is a paucity of research on remediation methods in the Sudan Savanna. There is limited understanding of the interrelationships among polystyrene contamination, microbial dynamics, and soil fertility. Researchers have previously examined the impact of microplastics and spent engine oil on plant and soil health. Therefore, this study evaluates the efficacy of mycorrhiza inoculation and cattle dung amendment in alleviating Styrofoam-induced soil degradation.

Materials And Methods

The study was conducted at the Teaching and Research Farm of the Department of Soil Science at Federal University Dutse in Jigawa State, Nigeria. The research site is situated in the Sudan Savannah agro-ecological zone at a latitude of $11^{\circ}42'04''\text{N}$ and a longitude of $9^{\circ}20'31''\text{E}$, with an elevation of 457.36 meters above sea level as reported by Nkereuwem et al. [15]. The area demonstrates a hot topic, wet-dry weather, defined by two distinct seasons: the wet season from May to October and the dry season from November to April. The average annual temperature is roughly 26°C , with a winter range of 21°C (December to February) and a summer range of 38°C (April to May). Annual precipitation ranges from 681 mm to 743 mm, contributing to the region's semi-humid conditions [16]. The experiment was conducted in a screen house using plastic pots (anvils) measuring 25 cm top diameter, 18 cm

bottom diameter, and 25 cm height, each filled with 5 kg of air-dried, sieved soil. The pots were perforated at the base to ensure adequate drainage. A completely randomised design (CRD) with three replications was employed (Plate 1). Treatments were arranged in a $3 \times 2 \times 2$ factorial combinations comprising three levels of Styrofoam contamination (0%, 2%, and 5% w/w), two levels of mycorrhizal inoculation (10 and 15 g/pot), and two levels of cattle dung (50 and 100 g/pot), resulting in 12 treatment combinations replicated three times for a total of 36 experimental units.

A soil sample was randomly obtained from the Teaching and Experimental Farm at a location devoid of previous Styrofoam contamination, at an excavation depth of 0 - 15 cm, utilizing a shovel. The collected soil was air-dried and filtered through a 2-mm sieve to eliminate debris, in accordance with the methodology described by Adeleye et al. [17]. A composite soil sample was created by meticulously blending the sieved soil and potted.



Plate (1): Treatment application, and experimental layout

Styrofoam (0, 2, and 5% w/w) soil was mixed thoroughly with the soil, and thereafter, cattle dung (50 and 100 g/pot) was applied and mixed thoroughly with the soil to ensure even distribution (Plate 1). The soil mixtures were incubated for two weeks before planting to facilitate stabilization. Mycorrhiza (*Glomus Clarum*) inoculation (10 and 15 g/pot) was blended into the central third of the potted soil at planting according to Nkereuwem et al. [18]. Three (3) cowpea (*ALOKA LOCAL*) seeds were sown and later thinned to 2 stand/pot after germination. The pots were positioned in the experimental area with a spacing of 0.5 m between them and 1 m between replications, as recorded by Adeleye et al. [17].

The particle size distribution (PSD) was measured utilizing the hydrometer procedure [16], and the soil's potential of hydrogen (pH) was quantified with a Hanna digital pH meter. The method of Walkley-Black was employed to analyze organic carbon content, whereas the Olsen method was utilized to ascertain available phosphorus. Exchangeable bases, such as calcium, potassium, magnesium, and sodium, were quantified using a Jenway flame photometer, while electrical conductivity (EC) was evaluated in accordance with FAO [19].

The growth parameter (plant height) of the test crop was measured across different weeks. Information on the soil's chemical properties was also generated through laboratory analysis.

Statistical Analysis

Data collected were subjected to Analysis of Variance (ANOVA) to identify significant differences ($p < 0.05$) among the treatments using Microsoft Excel, with significant differences between means assessed through Duncan's Multiple Range Test (DMRT).

Results And Discussion

Effects of Styrofoam Contamination on Soil Physicochemical Properties

The findings indicate that Styrofoam (SF) contamination significantly modified the physicochemical properties of the soil (Table 1), resulting in decreased fertility and nutrient imbalances. The contaminated process resulted in a slight increase in soil pH, trending towards salinity, as noted by Cella et al. [20]. This result aligns with the findings of Zhang et al. [21] and is comparable to the study conducted by Zhao et al. [22]. The reduction in organic carbon content in SF-contaminated soils may result from diminished microbial decomposition and restricted organic matter input. The findings align with the research conducted by Liu et al. [4], which suggested that microplastics may decrease soil organic matter. According to Cheng et al. [23], microplastics can alter the way nutrients function in the soil, and decreased phosphorus availability indicating that Styrofoam pollution hindered phosphorus's ability to decompose and migrate through the soil. The effect of Styrofoam contamination on soil properties is presented in Table 1. Statistical analysis revealed significant treatment effects on most parameters. Available phosphorus increased markedly at 2% Styrofoam contamination (18.90 mg/kg) compared to the control (2.30 mg/kg) and the 5% treatment (2.53 mg/kg) ($p < 0.001$). Additionally, the presence of Styrofoam may have altered microbial activity, leading to

enhanced mineralization of organic phosphorus pools. However, at 5% contamination, available P dropped to 2.53 mg/kg, suggesting that higher contamination levels may induce phosphorus immobilization or adsorption. The contamination by SF adversely impacted the soil's ability to retain and supply essential nutrients. Similarly, cation exchange capacity (CEC) was significantly higher at 5% contamination (3.45 cmol/kg) than at lower levels ($p = 0.009$). Soil pH increased progressively, with the 5% treatment recording significantly higher pH (6.03) than the control (5.69; $p = 0.024$). Conversely, organic carbon declined sharply from 1.20% in the control to 0.034% and 0.20% at 2% and 5% contamination, respectively ($p < 0.001$). Total nitrogen was significantly lower at 5% contamination (0.0073%) compared to 2% (0.0110%; $p = 0.018$). Particle size distribution shifted toward coarser texture with increasing Styrofoam levels, as sand content increased significantly ($p < 0.001$), while silt and clay decreased ($p = 0.003$ and $p = 0.011$, respectively), though the textural class remained sandy loam across all treatments. This increase is somewhat counterintuitive, as Styrofoam is non-polar and does not possess charged functional groups that directly contribute to CEC. A plausible explanation is that the incorporation of Styrofoam particles altered soil structure, potentially exposing more clay surfaces or organic matter exchange sites. Alternatively, the increase in soil pH from 5.69 to 6.03 at 5% contamination may have increased the negative surface charge of soil colloids, thereby enhancing CEC. The findings support the earlier study by Awet et al. [24], indicating that polystyrene nanoparticles modified soil microbial communities and functions, which may influence nutrient cycling and availability. The work of Amos et al. [25] demonstrates the role of organic carbon in enhancing soil Cation Exchange Capacity (CEC) through improved nutrient retention, indicating that its reduction can worsen contamination effects. The finding of this study aligns with the results of Eba et al. [26], which indicated that various contaminants can alter soil pH, organic matter, and nutrient availability, thereby impacting agricultural productivity.

Table (1): Effect of Styrofoam contamination levels on selected physical and chemical properties of soil

Parameter	Styrofoam contamination level (%)			P-value
	0	2	5	
pH	5.69 ± 0.08 ^b	5.80 ± 0.10 ^{ab}	6.03 ± 0.06 ^a	0.024
Organic Carbon (%)	1.20 ± 0.11 ^a	0.034 ± 0.008 ^c	0.20 ± 0.03 ^b	<0.001
Organic matter (%)				
Total Nitrogen (%)	0.0093 ± 0.0012 ^{ab}	0.0110 ± 0.0010 ^a	0.0073 ± 0.0008 ^b	0.018
Av. Phosphorus (mg/kg)	2.30 ± 0.30 ^b	18.90 ± 1.20 ^a	2.53 ± 0.25 ^b	<0.001
CEC (cmol/kg)	2.65 ± 0.21 ^b	2.76 ± 0.19 ^b	3.45 ± 0.18 ^a	0.009
Particle size (%)				
Sand	70 ± 2 ^c	74 ± 1 ^b	80 ± 2 ^a	<0.001
Silt	16 ± 1 ^a	14 ± 1 ^{ab}	10 ± 1 ^b	0.003
Clay	14 ± 1 ^a	12 ± 1 ^{ab}	10 ± 1 ^b	0.011
Textural class (USDA)	Sandy loam	Sandy loam	Sandy loam	–

Values are means ± standard deviation ($n = 3$). Means within a row followed by different superscript according to Duncan's Multiple Range Test ($p < 0.05$)

Impact of SF-contamination on Soil Structure and Plant Growth

Table 2 illustrates the negative impacts of SF contamination on plant growth. In the control soil (0% SF), the test crop (cowpea) exhibited progressive growth, attaining an average height of 25 cm by day 28, compared with the plant heights obtained in soil with 2 and 5% SF contamination. Soils treated with 2% and 5% SF completely inhibited cowpea germination, resulting in no growth during the experiment. This outcome aligns with the findings of Liu et al. [4], who reported that microplastic contamination, including polystyrene, can alter soil properties, affect nutrient availability, and negatively impact plant development.

Table (2): Effect of SF-contamination on plant height of cowpea at different Days After Sowing (DAS)

Styrofoam contamination (%)	Plant height (cm)		
	7 DAS	14 DAS	28 DAS
0	5	15	25
2	NG	NG	NG
5	NG	NG	NG

NG: No growth

Effect of Mycorrhizal Inoculation (MI) on Styrofoam Biodegradation

Mycorrhiza inoculation markedly enhanced the chemical characteristics of SF-contaminated soil, as shown in Table 3. In comparison with 0% SF- contamination (pH 6.03, organic matter 0.12%, organic carbon 0.20%, available phosphorus 18.9 mg/kg, nitrogen 0.011%, and CEC 3.45 cmol (+)/kg), mycorrhizal inoculation at 10 and 15 g/pot improved the majority of the soil properties analyzed. The effect of mycorrhiza inoculation on soil properties in Styrofoam-contaminated soil is presented in Table 3. Statistical analysis using independent t-tests revealed significant improvements in most soil parameters with increasing mycorrhiza inoculation from 10 to 15 g/pot. Soil pH increased significantly from 6.30 to 6.92 ($p = 0.002$), while electrical conductivity rose from 0.42 to 0.76 ds/m ($p = 0.001$). Organic carbon and organic matter also increased significantly ($p = 0.026$ and $p = 0.021$, respectively). The most pronounced effect was observed in available phosphorus, which increased from 3.05 to 14.16 mg/kg ($p < 0.001$), and exchangeable potassium, which increased from 0.188 to 1.210 cmol/kg ($p < 0.001$). Conversely, calcium decreased significantly from 2.50 to 1.80 cmol/kg ($p = 0.010$). Cation exchange capacity (CEC) improved significantly from 4.06 to 5.82 cmol/kg ($p = 0.001$). Among particle size fractions, silt decreased from 16% to 12% ($p = 0.008$), while clay increased from 8% to 12% ($p = 0.008$), though the textural class remained sandy loam across both treatments. No significant difference was observed in total nitrogen ($p = 0.760$) or sand content ($p = 1.000$). The AMF inoculation enhanced soil health by improving physicochemical and biological properties in polluted soil, supporting previous findings by [27-29] regarding its role in plant growth and soil fertility under contaminated soil.

Table (3): Effect of mycorrhiza inoculation (MI) on soil properties of Styrofoam-contaminated soil

Parameter	Mycorrhiza inoculation (g/pot)		t-value	p-value	Significant
	10	15			
pH	6.30 ± 0.12	6.92 ± 0.10	6.82	0.002	Significant
Electrical Conductivity (ds/m)	0.42 ± 0.04	0.76 ± 0.06	8.24	0.001	Significant
Organic Carbon (%)	1.60 ± 0.11	1.90 ± 0.10	3.46	0.026	Significant
Organic Matter (%)	2.75 ± 0.19	3.30 ± 0.17	3.66	0.021	Significant
Phosphorous (mg/kg)	3.05 ± 0.28	14.16 ± 0.92	20.36	<0.001	Highly Significant
Nitrogen (%)	0.0088 ± 0.0008	0.0090 ± 0.0007	0.33	0.760	Not Significant
Sodium [cmol(+)/kg]	0.130 ± 0.012	0.159 ± 0.010	3.24	0.032	Significant
Potassium [cmol(+)/kg]	0.188 ± 0.015	1.210 ± 0.087	20.15	<0.001	Highly Significant
Calcium [cmol(+)/kg]	2.50 ± 0.21	1.80 ± 0.15	4.66	0.010	Significant
Magnesium [cmol(+)/kg]	0.75 ± 0.07	2.00 ± 0.12	15.44	<0.001	Highly Significant
Exchangeable Acidity [cmol(+)/kg]	0.50 ± 0.06	0.66 ± 0.05	3.58	0.023	Significant
Total Exchangeable Bases [cmol(+)/kg]	3.56 ± 0.22	5.16 ± 0.24	8.44	0.001	Significant
Cation Exchangeable Capacity (CEC) [cmol(+)/kg]	4.06 ± 0.24	5.82 ± 0.27	8.40	0.001	Significant
Particle size (%)					
Sand	76 ± 2	76 ± 2	0.00	1.000	Not Significant
Silt	16 ± 1	12 ± 1	4.90	0.008	Significant
Clay	8 ± 1	12 ± 1	4.90	0.008	Significant
Textual class (USDA)	Sandy loam	Sandy loam	–	–	

Additionally, the soil fertility indices (N, K, and Mg) improved, thereby enhancing nutrient retention and the potential for plant growth with increased mycorrhizal inoculation. The CEC values demonstrated an increase from 3.45 cmol(+)/kg in Styrofoam-contaminated soil to 4.06 and 5.82 cmol(+)/kg for 10 g/pot and 15 g/pot mycorrhiza inoculations, respectively, indicating enhanced nutrient retention properties essential for sustainable crop productivity. The findings were corroborated by studies demonstrating that the addition of Arbuscular Mycorrhizal Fungi (AMF) can neutralize soil pH, which is optimal for the majority of crops [6].

Role of Cattle Dung (CD) in Enhancing Degradation

The chemical properties of the SF-contaminated soil showed significant enhancement with the incorporation of cattle dung as an organic amendment, in comparison to the control (0% styrofoam contaminated soil) (Table 4). The soil organic matter content exhibited an increase in both the 50 g/pot and 100 g/pot CD application, with the most significant rise observed in the 100 g/pot CD with a value of 3.27%. The increase in soil organic matter indicates enhanced microbial decomposition of organic materials, thereby augmenting the potential for carbon storage, which

is crucial for nutrient cycling and overall soil fertility. Organic materials were observed to enhance microbial growth and increase biological activity, as soil organic carbon levels rose from 0.2% in contaminated soil to 1.9% following treatment with 100 g/pot CD. The pH values increased from 6.03 to a neutral range of 7.13 to 7.15, which enhanced the enzymatic activity of microorganisms and facilitated nutrient absorption by the roots. The findings of this study match those of Bazie et al. [30], who indicated that the application of cow dung to contaminated soils enhances organic matter content in proportion to the amount applied, although there exists a threshold beyond which further application ceases to provide benefits to the soil. The results of this study concur with the earlier research by [31, 32], which demonstrated that adding organic materials, such as cow dung and cattle rumen digesta, can enhance the cleaning of contaminated soils by encouraging microbial activity, which increases nutrient accessibility. Gomez et al. [33] reported that similar amendments improve soil structure and quality, consistent with the observed improvements in soil physical properties in this study.

Table (4): Effect of Cattle Dung (CD) on soil properties of Styrofoam contaminated soil

Parameter	Cattle dung (g/pot)	
	50	100
pH	7.13	7.15
Electrical Conductivity (ds/m)	1.48	0.85
Organic Carbon (%)	0.4	1.9
Organic Matter (%)	0.68	3.27
Phosphorous (mg/kg)	28.81	18.76
Nitrogen (%)	0.029	0.019
Sodium [cmol(+)/kg]	0.3	0.79
Potassium [cmol(+)/kg]	2.2	1.3
Calcium [cmol(+)/kg]	4.9	1.75
Magnesium [cmol(+)/kg]	1.05	2.0
Exchangeable Acidity [cmol(+)/kg]	0.33	0.33
Total Exchangeable Bases [cmol(+)/kg]	8.47	5.84
CEC [cmol(+)/kg]	8.8	6.17
Particle size (%)		
Sand	74	78
Silt	14	8
Clay	12	14
Textural class (USDA)	Sandy loam	Sandy loam

Textural and CEC contrasts imply improved soil structure and capacity of cattle dung as organic soil ameliorant. The CEC of the polluted soil was 3.45 cmol/kg, which increased significantly to 8.8 cmol(+) kg(-1) with the addition of 50 g/pot of cattle dung, and then decreased to 6.17 cmol(+) kg(-1) with 100 g/pot cattle dung. These results support the findings of Rayne & Aula [34] who observed an increase in Cation Exchange Capacity (CEC) with the addition of manure to the soil, thus improving nutrient retention, resulting in improved soil fertility. The TEB increased significantly in the polluted soils up to 8.47 cmol/kg with 50 g/pot CD, the application of cattle dung at 100 g/pot resulted in decreased TEB (5.84 cmol(+)/kg). Higher magnesium (2.0 cmol(+)/kg) concentration in 100 g/pot CD shows greater soil conditioning in comparison with 1.05 cmol(+)/kg in 50 g/pot CD. Cattle dung application at 50 g/pot increased the CEC of the experimental soil from 3.45 cmol(+)/kg to 8.8 cmol(+)/kg, indicating that the soil's nutrient holding capacity had increased. Greater TEB concentration (8.47 cmol (+)/kg) was obtained at 50 g/pot cattle dung application, indicating an enhanced soil fertility. The findings of this research are in agreement with the report of Nkereuwem et al. [32], who reported an increase in soil properties due to cattle rumen digesta and poultry droppings.

Combined Effect of Mycorrhizal Inoculation and Organic Amendments

In this study, a mixture of 10 g MI and 50 g CD resulted in a pH of 7.18, which is favourable for microbial activity and nutrient availability, and serve as the superior treatment combination. This treatment consistently produced significantly higher values for most soil fertility parameters compared to the 15 g MI + 100 g CD treatment. This result is in agreement with the finding of Al Methyeb et al. [35] who reported that the application of these two treatments significantly improved the soil pH because the amount of MgO was able to compromise the soil, which encouraged microbial and plant growth. Research indicates that incorporating arbuscular mycorrhizal fungi (AMF) into organic amendments like cattle manure markedly enhances soil organic carbon and fertility [36]. The integrated treatments demonstrated a beneficial impact on phosphorus availability. The phosphorus concentration measured

16.899 mg/kg following the application of 10g MI and 50g CD, which was significantly greater than the 18.9 mg/kg observed in the contaminated soil and the 1.311 mg/kg recorded after the treatment of 15g MI and 100g CD. This enhancement aligns with findings by Samanhudi et al. [37], indicating that AMF inoculation can enhance phosphorus solubilisation and uptake in plants when used in conjunction with organic fertilizers.

Table (5): Effects of mycorrhiza inoculation (MI) and cattle dung (CD) on soil properties of Styrofoam contaminated soil

Parameter	Value		DMRT	Significance
	10 g MI+ 50 g CD	15 g MI + 100 g CD		
pH	7.18	6.9	a b	10 g MI + 50 g CD > 15 g MI + 100 g CD
Electrical Conductivity (ds/m)	1.23	0.28	a b	10 g MI + 50 g CD > 15 g MI + 100 g CD
Organic Carbon (%)	2	0.04	a b	10 g MI + 50 g CD > 15 g MI + 100 g CD
Organic Matter (%)	3.44	0.68	a b	10 g MI + 50 g CD > 15 g MI + 100 g CD
Phosphorous (mg/kg)	16.899	1.311	a b	10 g MI + 50 g CD > 15 g MI + 100 g CD
Nitrogen (%)	0.018	0.00019	a b	10 g MI + 50 g CD > 15 g MI + 100 g CD
Sodium [cmol(+)/kg]	0.24	0.008	a b	10 g MI + 50 g CD > 15 g MI + 100 g CD
Potassium [cmol(+)/kg]	1.91	0.14	a b	10 g MI + 50 g CD > 15 g MI + 100 g CD
Calcium [cmol(+)/kg]	3.15	1.9	a b	10 g MI + 50 g CD > 15 g MI + 100 g CD
Magnesium [cmol(+)/kg]	4.1	0.35	a b	10 g MI + 50 g CD > 15 g MI + 100 g CD
Exchangeable Acidity [cmol(+)/kg]	0.5	0.66	b a	15 g MI + 100 g CD > 10 g MI + 50 g CD
Exchangeable Bases [cmol(+)/kg]	9.4	2.398	a b	10 g MI + 50 g CD > 15 g MI + 100 g CD
CEC [cmol(+)/kg]	9.9	3.056	a b	10 g MI + 50 g CD > 15 g MI + 100 g CD
Particle size (%)				
Sand	79	77	a a	No significant difference
Silt	11	12	a a	No significant difference
Clay	10	11	a a	No significant difference
Textural class (USDA)	Sandy loam	Sandy loam		

Duncan's Multiple Range Test (DMRT) revealed significant differences between the two combination treatments across most soil properties (Table 5 & 6). The combination of 10 g mycorrhiza inoculation with 50 g cattle dung (10 g MI + 50 g CD) produced significantly higher values for organic carbon (2.00%), organic matter (3.44%), available phosphorus (16.90 mg/kg), total nitrogen (0.018%), exchangeable potassium (1.91 cmol/kg), exchangeable calcium (3.15 cmol/kg), exchangeable magnesium (4.10 cmol/kg), CEC (9.90 cmol/kg), and total exchangeable bases (9.40 cmol/kg) compared to the 15 g MI + 100 g CD treatment. Conversely, exchangeable acidity was significantly lower in the 10 g MI + 50 g CD treatment (0.50 cmol/kg) compared to the higher combination (0.66 cmol/kg). These findings indicate that the moderate combination of 10 g mycorrhiza and 50 g cattle dung is superior for enhancing soil fertility in Styrofoam-contaminated soil, while the higher combination (15 g MI + 100 g CD) resulted in diminished soil quality, likely due to nutrient imbalance or leaching losses.

Table (6): Significant Differences on mycorrhiza inoculation (MI) and cattle dung (CD) on soil properties of Styrofoam-contaminated soil

Category	10 g MI + 50 g CD	15 g MI + 100 g CD	Superior Treatment
Organic Carbon (%)	2.00 ^a	0.04 ^b	10 g MI + 50 g CD
Organic Matter (%)	3.44 ^a	0.68 ^b	10 g MI + 50 g CD
Phosphorus (mg/kg)	16.899 ^a	1.311 ^b	10 g MI + 50 g CD
Nitrogen (%)	0.018 ^a	0.00019 ^b	10 g MI + 50 g CD
Potassium (cmol/kg)	1.91 ^a	0.14 ^b	10 g MI + 50 g CD
Calcium (cmol/kg)	3.15 ^a	1.90 ^b	10 g MI + 50 g CD
Magnesium (cmol/kg)	4.10 ^a	0.35 ^b	10 g MI + 50 g CD
CEC (cmol/kg)	9.90 ^a	3.056 ^b	10 g MI + 50 g CD
Exchangeable Acidity (cmol/kg)	0.50 ^b	0.66 ^a	10 g MI + 50 g CD (lower acidity)

Implications for Sustainable Agriculture

The application of mycorrhizal inoculation and cattle dung significantly influenced the growth performance of cowpea in SF-contaminated soil for the period monitored. The results highlight that the highest growth rates were observed in the 50 g/pot cattle dung application, which exhibited a steady increase from 7.8 cm in Week 1 to 14 cm in Week 5, as shown in Plate 2, and Table 7. This suggests that cattle dung provides essential nutrients that support plant growth in contaminated environments. Mycorrhizal treatments also enhanced plant height compared with treatments without mycorrhiza. The result highlights the beneficial effect of mycorrhiza in enhancing plant growth. These findings align with the previous study by [27, 28], emphasizing the positive impact of organic amendments and mycorrhizal fungi on plant growth in stressed conditions. Studies by [15, 38] demonstrated that amendments with cattle dung significantly enhanced soil fertility and microbial activity, thereby facilitating improved plant establishment.

Table (7): Growth response of cowpea plants across treatments from Styrofoam-contaminated soil (means \pm SD, n=3)

Treatment	Week 1 (cm)	Week 2 (cm)	Week 3 (cm)	Week 4 (cm)	Week 5 (cm)
50g CD	7.8 \pm 0.5 ^a	12.0 \pm 0.8 ^a	12.4 \pm 0.7 ^a	13.2 \pm 0.6 ^a	14.0 \pm 0.8 ^a
10g MI	3.8 \pm 0.3 ^b	5.0 \pm 0.4 ^c	5.8 \pm 0.4 ^c	6.1 \pm 0.5 ^c	9.9 \pm 0.6 ^b
15g MI	2.4 \pm 0.2 ^c	5.0 \pm 0.3 ^c	6.0 \pm 0.5 ^c	6.4 \pm 0.4 ^c	8.0 \pm 0.5 ^c
10g MI + 50g CD	3.9 \pm 0.3 ^b	4.4 \pm 0.4 ^c	5.9 \pm 0.4 ^c	6.4 \pm 0.5 ^c	7.8 \pm 0.6 ^c
100g CD	0.0 \pm 0.0 ^d	4.2 \pm 0.3 ^c	5.8 \pm 0.4 ^c	6.0 \pm 0.4 ^c	8.2 \pm 0.5 ^c
15g MI + 100g CD	0.0 \pm 0.0 ^d	0.0 \pm 0.0 ^d	0.0 \pm 0.0 ^d	0.0 \pm 0.0 ^d	0.0 \pm 0.0 ^d
p-value	<0.001	<0.001	<0.001	<0.001	<0.001

N.B: CD- Cattle dung; MI-Mycorrhiza inoculation

Duncan's Multiple Range Test (DMRT) revealed significant treatment effects on cowpea plant height across all weeks ($p < 0.001$). The 50 g cattle dung (50g CD) treatment consistently produced the tallest plants, with a mean height of 14.0 cm at week 5, which was significantly higher than all other treatments ($p < 0.001$). The 10 g mycorrhiza (10g MI) treatment emerged as the second-best treatment at week 5 (9.9 cm), significantly outperforming other mycorrhiza-based treatments. Treatments including 15g MI, 100g CD, and 10g MI + 50g CD were statistically similar at week 5 (7.8–8.2 cm), with no significant differences among them. The combination of 15g mycorrhiza with 100g cattle dung (15g MI + 100g CD) resulted in complete growth inhibition (0 cm) throughout the study, indicating phytotoxicity from excessive application rates. Notably, the 100g CD treatment exhibited delayed germination (0 cm at week 1) but recovered by week 2 and achieved final growth comparable to other treatments. These results demonstrate that moderate cattle dung application (50g CD) is optimal for cowpea growth in Styrofoam-contaminated soil, while high-rate combinations of mycorrhiza and cattle dung are detrimental.



Plate (2): Germination and comparative growth from week 1 – week 5

This result is in agreement with Dowarah et al. [39], who highlighted the role of MI in enhancing nutrient uptake, particularly when applied with moderate cattle dung, making it a promising strategy for improving contaminated soil quality. Notably, post-hoc analysis test identified 50 g/pot CD as the most effective amendment (12.32 cm mean height), significantly enhancing plant growth Figure 1. Similarly, a balanced combination of mycorrhiza and moderate cattle dung (10 g/pot MI + 50 g/pot CD, 7.8 cm) supported plant establishment, highlighting the importance of optimizing amendment concentrations for effective bioremediation. This result corroborates the findings of [15, 22] who reported enhanced plant growth due to mycorrhiza inoculation in contaminated soil. Conversely, a high level of organic inputs, such as 100 g/pot CD (8.2 cm) and 15 g/pot MI + 100 g/pot CD (0 cm), suppressed the growth, which may be attributed to nutrient imbalances or microbial succession. This result is also supported by findings reported in Tagele et al. [40] who implied that high organic amendment brought about potential interference with microbial nitrogen mineralization, which may cause nutrient leaching and declining plant productivity.

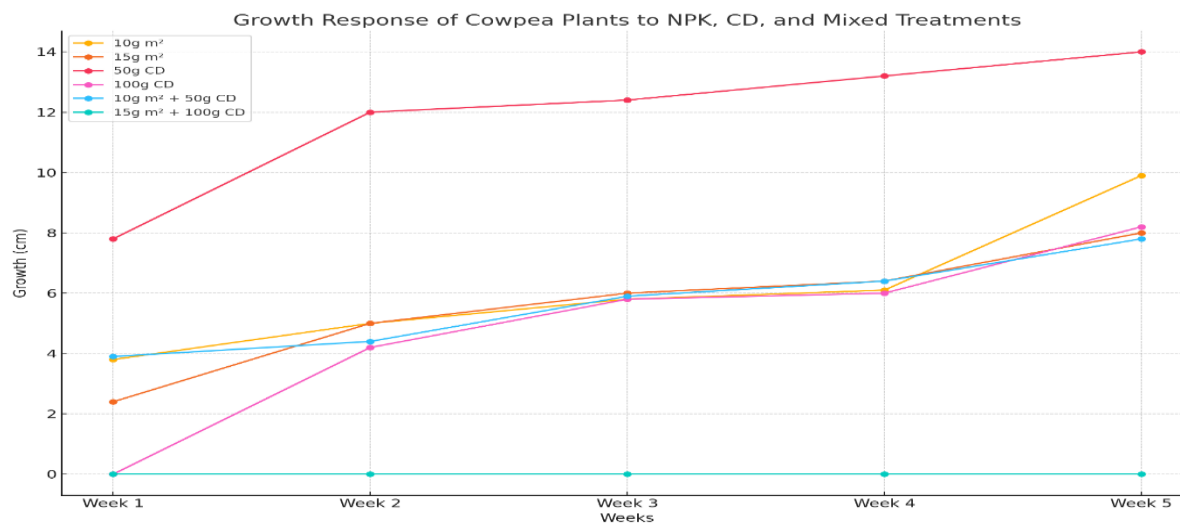


Figure 1. Growth response of cowpea plants to MI, CD, & MICD treatments in SF-contaminated soil

Conclusion

This study offers insights into the effective application of mycorrhizal inoculation and cattle dung for the remediation of SF-contaminated soil. It highlights the enhancement of plant growth and soil fertility, which can be attributed, at least in part, to favourable changes in soil physiochemical properties. Furthermore, the results indicated that SF contamination may influence soil enzymatic activity, which may have a potential impact on carbon, nitrogen, and phosphorus cycles in soil. Moderate organic additive, especially 50 g of cattle dung was shown to significantly promote plant height and nutrient storage and hence to be the most competent strategy for bioremediation. Future research should explore long-term microbial interactions, nutrient cycling, and large-scale applications to refine bioremediation strategies.

Conflict of Interest

The authors declare no conflicts of interest associated with this manuscript.

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