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### Environmental and Engineering Performance of Terrazyme-Treated Clayey Soil

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#### ABSTRACT

This study examines the mechanical behavior of clayey soil treated with terrazyme, with concentration on how it affects soil characteristics. It assesses how different terrazyme dosages affect important geotechnical and environmental properties, such as Atterberg limits, compaction characteristics (OMC, MDD), strength (UCS, CBR), leaching potential (TCLP), and micro-structure. Terrazyme raised MDD from 19.71 kN/m<sup>2</sup> to 24.02 kN/m<sup>3</sup> and reduced OMC from 13.45% to 9.45% at a dose with a dilution factor of 1.6. It also enhanced soil plasticity, lowering the LL from 52% to 40% and the PI from 23% to 8%, while increasing the PL from 29% to 32%. At a dilution factor of 1.7, UCS raised from 150 kPa to 156.2 kPa, and after 28 days, it raised even more to 300.5 kPa. CBR values rose from 3% to 14% after 7 days and from 5% to 25% after 28 days due to the enhanced soil micro-structure. Terrazyme was relatively successful in bringing the concentrations of Hg, Ba, and Pb down to EPA standards; the most effective results were obtained at a dilution factor of 1.8. Analysis using a scanning electron microscopy (SEM) showed that the treated samples had a denser particle packing. In order to comprehend more about soil-enzyme interactions for geotechnical applications, this work emphasizes the necessity of conducting systematic research on enzyme-based soil stabilization.

**Keywords:** Consistency, Mechanical behavior, Geotechnical properties, Clayey soil, Terrazyme, heavy metal, Micro-structure.

#### INTRODUCTION

Clayey soils, which are common in Pakistan's

Punjab and Sindh, have unfavorable engineering characteristics, such as high plasticity, poor shear strength, and high settlement potential. In addition to

their low shear strength and bearing capacity, these soils present a major geotechnical difficulty when it comes to building infrastructure (Ardah et al., 2017; Jamil et al., 2024; Khajeh et al., 2020; Liaqat et al., 2019; Liu et al., 2020; Mekonnen et al., 2020; Onyelowe & Duc, 2020; Waleed & Alshawmar, 2025; Waleed et al., 2024). The sub-grade layer's cracks and eventual structural failure are caused by the clayey soil's swelling and shrinking behavior (Tara et al., 2025). To enhance its engineering characteristics, soil might be stabilized chemically, mechanically, or in other ways. Enzyme treatment decreases permeability and the need for mechanical compaction while increasing soil density and shear strength (Khan & Taha, 2015; Pooni et al., 2019). Traditional soil stabilizers, such as cement and lime, pollute the environment by emitting carbon (Modarres & Nosoudy, 2015; Mohammed et al., 2023; Saldanha et al., 2021). Enzymes may decrease the compaction effort needed for mechanical stabilization and are energy-efficient. They speed up the conversion state by controlling reaction rates and lowering the activation energy for the production of new products, because they are biological catalysts (Scholen, 1992).

Numerous investigations on the effects of terrazyme on different soil types have mostly concentrated on the

way in which it enhances the engineering properties of clayey soils, with little consideration to the environmental effects of terrazyme on soil. To determine whether terrazyme-treated clayey soils are a viable option for long-term soil stabilization, a thorough assessment of their engineering and environmental characteristics is necessary. By carefully assessing the behavior of clayey soil treated with terrazyme and offering insightful information about its applicability for the creation of sustainable infrastructure, this research aims to fill existing research gaps. This research aims to contribute to the broader context on environment and sustainable infrastructure development.

## MATERIALS AND METHODOLOGY

### Materials

To perform project tests on soil, the soil was collected from Rawalpindi, Pakistan. It was extracted from a site under construction. Hence, the sample was taken from a depth of 1 meter. The sample contained 0% gravel.

Table 1 summarizes the main geotechnical properties of clayey soil.

**Table 1. Geotechnical properties of clayey soil**

Soil type	Classification		Atterberg's limits			Grain size distribution			Moisture density relationship	
	USCS	AASHTO	LL (%)	PL (%)	PI (%)	Sand (%)	Clay (%)	Silt (%)	MDD (kN/m <sup>3</sup> )	OMC (%)
Clayey soil	CH	A-4	52	29	23	8	73	19	19.1	13.45

Terrazyme is an organic biopolymer based enzyme that gets produced when fermentation processes alter the chemical composition of fruit extracts, vegetables, sugar, and water. Terrazyme's pH ranges from 4.30 to 4.60, its specific gravity ranges from 1 to 1.09, and its vapor density is 1 (Waleed et al., 2023).

### Research Methodology

To assess terrazyme's effect on soil characteristics, the research study was systematically divided into three stages, as shown in Fig.1. These stages are as follows:

1. Untreated soil characterization;
2. Terrazyme dosage optimization;
3. Assessment of properties of treated soil.

### Phase-I: Properties of Untreated Soil

#### Modified Proctor Test

To ascertain the maximum dry density (MDD) at the optimum moisture content (OMC), a laboratory experiment was conducted adhering to the guidelines of ASTM D1557-12, as shown in Fig. 3. First, 4% water by weight was mixed, and for each experiment after that, the amount was increased by 3%.

#### Unconfined Compressive Strength Test

The Unconfined Compressive Strength (UCS) test followed ASTM D2166. Samples were molded to a 1:2 height-to-diameter ratio (8 cm x 4 cm), as shown in Fig. 2. Three specimens were prepared at the optimum

moisture content (OMC) and maximum dry density (MDD) from the modified Proctor test. The average UCS values were used for accuracy, and shear strength was calculated from the results.

### California Bearing Ratio Test

The California Bearing Ratio (CBR) test was conducted per ASTM D1883-16 to evaluate soil strength

(Fig. 2). It measures the penetration of a dynamic cone penetrometer (DCP) with an 8-kg hammer into a compacted specimen, correlating to *in-situ* CBR. Starting at 4%, the water content was increased by 3% for each test to find the ideal moisture state. The CBR value was derived from the penetration resistance recorded at set depths after compaction.

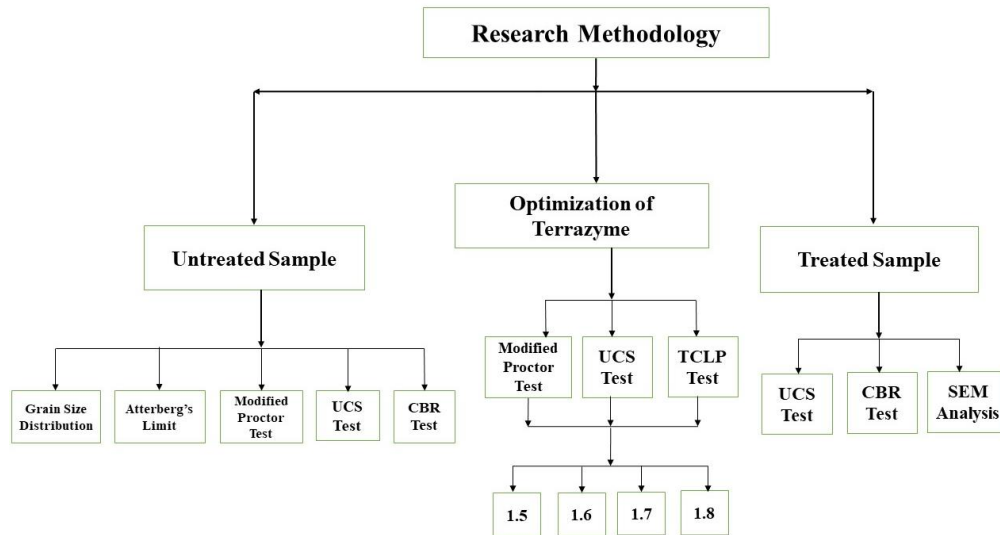


Figure 1. Research methodology

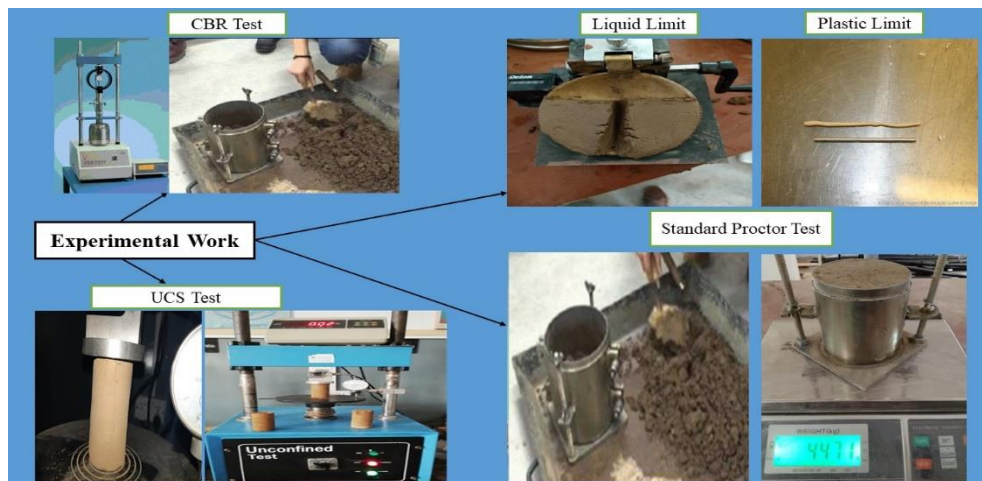


Figure 2. Soil tests performed in the laboratory

### Phase II: Optimization of Terrazyme

The second phase determined the optimal terrazyme dosage for soil stabilization, which depends on the soil's PI and MDD. The standard chart (Nature Plus) was used to determine the 1:100 dilution (factor of 1.6). This factor was multiplied by the soil mass (kg) to determine the enzyme quantity (ml). The optimum concentration

was determined by UCS tests conducted at dilution factors of 1.5, 1.6, 1.7, and 1.8. Before mixing, the water-soluble enzyme was dissolved, and specimens were cured so as to ensure a reaction. Compaction properties were examined using modified Proctor tests at different concentrations.

### Modified Proctor Test

The MDD and OMC were calculated using the modified Proctor test (D1557-12, 2021) to evaluate the efficacy of compaction and stabilization. Four Terrazyme dilution factors (1.5, 1.6, 1.7, and 1.8) were assessed, and the water content was raised by 3% increments from the starting 4%. The findings offered essential data for determining the optimal enzyme dosage for optimum stability and strength. Additionally, the investigation of the effects of enzymatic stabilization on compaction behavior was made easier by the differences in density and moisture content among parameters.

### Unconfined Compressive Strength Test

Four dilution factors (1.5-1.8) for soil stabilization were assessed using UCS testing (ASTM D2166). Using the modified Proctor test, samples have been made at the OMC and MDD and uniformly compacted. Shear strength was computed using the average UCS values, which ensured accuracy. This gave important information about the soil's ability to support loads when stabilized by enzymes.

### Phase-III: Properties of Treated Soil

Due to the third phase of the project which involved the properties of treated soil, UCS and CBR measurements were conducted using the previously determined optimal dosage of terrazyme.

### Unconfined Compressive Strength Test

In order to prepare UCS samples, terrazyme was dissolved in the modified Proctor test's optimal moisture content (OMC). The solution was well mixed with the soil to ensure uniform distribution. After that, the samples were placed in plastic bags and allowed to cure for 7, 14, 21, and 28 days, in order to measure the strength gain. Prior to testing, this curing time was necessary to ensure proper strength development and to promote the enzyme-soil reaction.

### California Bearing Ratio Test

The CBR test (ASTM D1883) was used to evaluate the strength of the soil treated with terrazyme. To assess long-term strength, gain, soil samples were compacted, treated with the recommended enzyme concentrations, and then cured for 7 days and 28 days. The cured samples were tested to determine the penetration

resistance and strength improvement from enzymatic stabilization.

### TCLP

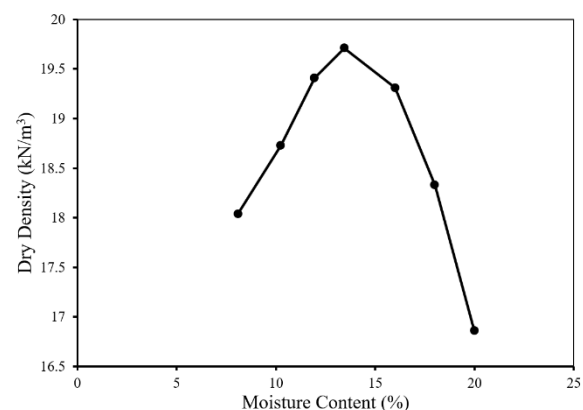
Samples were prepared using the US EPA Toxicity Characterization Leaching Procedure (Act, 2017). The concentrations of heavy metals and trace metal ions were determined using ICP-OES (Agilent 5800). Prior to analysis, samples were diluted 10-fold and 100-fold, then filtered through a 0.45  $\mu\text{m}$  organic membrane. All measured metal ion concentrations were found to be below US EPA environmental guidelines (Act, 2017).

## RESULTS AND DISCUSSION

### Phase I: Properties of Untreated Soil

#### Compaction of soil

The test began with an initial water content of 4% by weight, increasing by 3% for each subsequent trial. The maximum dry density was recorded as 19.71  $\text{kN/m}^3$  at an optimum moisture content of 13.45%, as shown in Fig. 3.



**Figure 3.** Dry density vs. moisture content of natural soil

### Unconfined Compressive Strength (UCS) Test

The UCS value of the soil was found to be 150 kPa.

### CBR Test

CBR was calculated based on the load values at standard penetrations of 2.54 mm and 5.08 mm. CBR at 2.54 mm penetration was found to be 3.20%, while at 5.08 mm penetration, it was found to be 3.35%. Since the higher value is considered for CBR evaluation, the final CBR value for the given soil sample is 3.35%.

## Phase II: Optimization of Terrazyme

The optimal terrazyme dosage for stabilizing high-plasticity soils was determined using standard tables (Nature Plus).

### Optimization for Modified Proctor Test

Dilution factors of 1.5, 1.6, 1.7, and 1.8 were selected using standard tables. Modified Proctor tests for each dosage showed that terrazyme significantly influences compaction, as seen in MDD and OMC variations. Based on the results, the enzyme improves particle arrangement and decreases voids to increase compaction and produce a denser soil matrix. With an OMC of 13.45%, the MDD for untreated soil (0% terrazyme) was 19.71 kN/m<sup>3</sup>. Terrazyme was added, and as Fig. 4 shows, the OMC decreased to 9.45% while the MDD first rose to a peak of 24.02 kN/m<sup>3</sup> at a dosage of 1.6. This indicates that through promoting enzymatic reactions that strengthen particle cohesion and result in higher density at lower moisture content, the enzyme strengthens soil bonding (Hinojosa et al., 2004). With values of 23.53 kN/m<sup>3</sup> and 21.67 kN/m<sup>3</sup> at 1.7 and 1.8

dosages, respectively, an insignificant decline in MDD was noted at higher dosages. The high enzyme content that alters the optimal soil structure is probably the cause of this loss. Better soil water efficiency is indicated by the simultaneous decrease in OMC with increased dosage. The 1.6 dosage had the lowest OMC (9.45%), indicating that the enzymatic treatment improves the soil's capacity to reach maximum density with less water. Excessive enzyme presence may be the cause of the little rise in OMC over this dosage (9.54% at 1.7 and 9.84% at 1.8), which could result in minor changes in the cohesion and moisture-retention properties of the soil. Prior research on soil stabilizing chemicals, such as enzyme additions, like terrazyme, showed enhancements in the soil's moisture-density relationship (Bara & Tiwary, 2023; Eujine et al., 2014).

Results indicate a 1.6 terrazyme dosage is optimal for peak MDD and lowest OMC. Beyond this, density gains decrease, likely from enzyme over-saturation. This confirms terrazyme's effectiveness in enhancing soil compaction, as well as improving strength and stability for geotechnical applications.

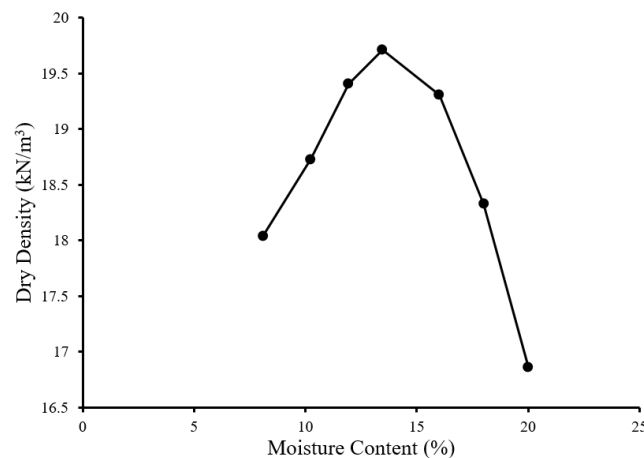


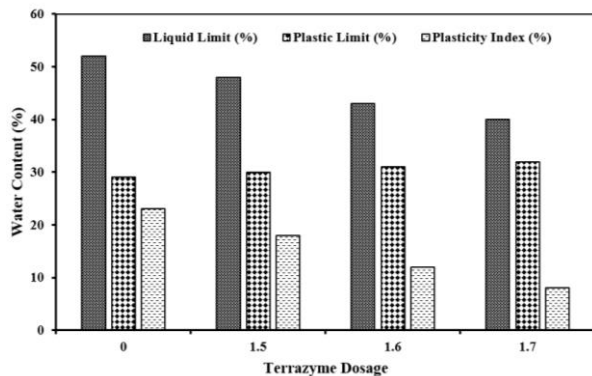
Figure 4. MDD vs. OMC at various dosages of terrazyme

### Effect of Terrazyme on Atterberg's Limits of Soil

As indicated in Figure 5, the LL dropped from 52% (untreated) to 40% at a terrazyme dosage of 1.7. This decrease suggests that the soil's capacity to retain water and change from a plastic to a liquid state has been much reduced. Terrazyme lowers the soil's capacity to absorb water by improving particle bonding and reducing the amount of free water present (Nadeem et al., 2023). As the terrazyme content rose, the PL went from 29% to 32%. Greater water absorption capacity before the soil becomes plastic is indicated by a higher PL. By

increasing particle cohesion and decreasing clay activity, terrazyme stabilizes soil and increases its capacity to retain water without deforming. The PI dropped from 23% to 8% as the terrazyme dosage increased, as depicted in Fig.5. Lower PI values imply the soil is less plastic and more stable. Terrazyme treatment reduces the plasticity of the soil, making it less prone to volume changes (swelling and shrinkage), and enhancing stability (Muguda & Nagaraj, 2019). Previous studies on the impact of enzymatic additives on Atterberg limits have reported varying results,

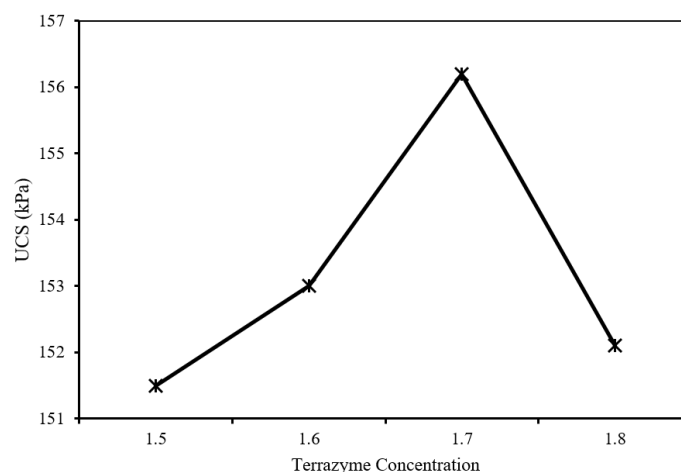
influenced by the enzyme type, soil characteristics, and treatment conditions (Eujine et al., 2014; Khan & Taha, 2015; Patel et al., 2018; Rajoria & Kaur, 2014; Renjith et al., 2020).



**Figure 5.** Liquid limit, Plastic limit and Plasticity index of soil treated with terrazyme

### Effect of Terrazyme on UCS of Soil

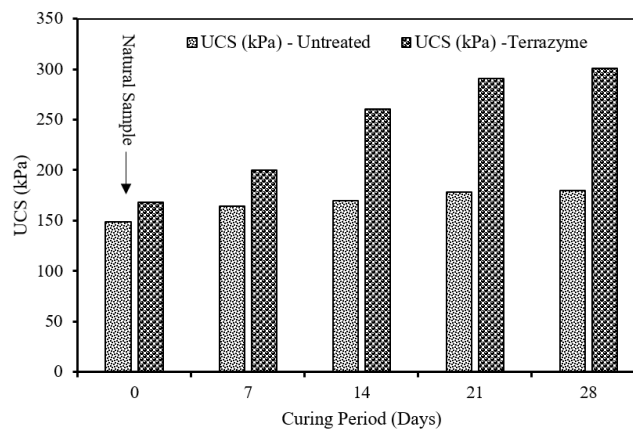
To evaluate the impact of terrazyme on soil strength, UCS tests were conducted at four different dilution factors: 1.5, 1.6, 1.7, and 1.8. Results showed that terrazyme-treated soil strength is dosage-dependent, with an optimal concentration for maximum improvement. The UCS values obtained for different terrazyme dosages are as follows: 151.5 kPa for 1.5, 153 kPa for 1.6, 156.2 kPa for 1.7, and 152.1 kPa for 1.8 as shown in Fig. 6. The highest UCS value of 156.2 kPa was recorded at a dilution factor of 1.7, indicating that this concentration provides the most effective stabilization. Due to improved interlocking, decreased voids, and strengthened particle bonding, UCS increased with terrazyme dosage up to 1.7. A little drop in UCS at a dilution factor of 1.8 indicates over-saturation, in which too much enzyme results in ineffective compaction. For maximal strength and cohesion for enhanced geotechnical performance, a dosage of 1.7 is proposed.



**Figure 6.** UCS of soil treated with varying amounts of terrazyme

In accordance with UCS data, terrazyme stabilization significantly increases strength over the duration of cure times. The UCS of the untreated soil was 150 kPa. The efficiency of the enzyme was shown by a noticeable strength increase after 7, 14, 21, and 28 days of curing with terrazyme (optimal dilution factor 1.7). The UCS rose from 150 kPa (untreated) to 200.12 kPa (treated) during the 7-day curing period, demonstrating the terrazyme's early-stage reactivity with soil particles (Fig. 7). The initial bonding improvement and decrease in voids within the soil matrix are responsible for this improvement. This UCS values increased to 260.23 kPa, 290.35 kPa, and 300.5 kPa, respectively, as the curing times increased to 14,

21, and 28 days. The trend demonstrates how the enzymatic process gradually strengthens soil structure over time. By strengthening inter-particle cohesion and generating a denser matrix from biological reactions, terrazyme improves UCS. In addition to increasing load-bearing capacity and decreasing permeability, this process allows for additional densification and strength gain through continuous curing. Prior investigation showed the same pattern, with UCS rising following varying curing times (Ramesh & Sagar, 2015). Overall, the findings indicate that terrazyme successfully enhances soil's mechanical properties, as seen by an increasing trend in UCS over time.

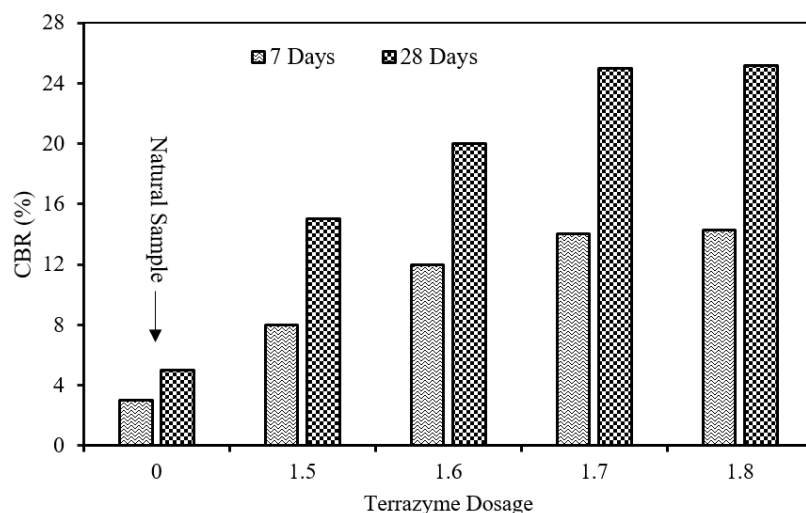


**Figure 7.** UCS of soil at different curing periods

### Effect of Terrazyme on CBR of Soil

Terrazyme dramatically increases soil strength, according to the CBR results. Over time, the enzyme improves penetration resistance and raises CBR values by strengthening particle bonds, decreasing voids, and increasing compaction efficiency. For both the 7-day and 28-day curing periods, the CBR values first rise significantly at a dosage of 1.5, suggesting early soil stability effects. The CBR values further improve when the dosage rises to 1.6 and 1.7, demonstrating the gradual improvement in soil strength brought about by improved particle cohesion and densification, as shown in Fig. 8. Strength development is also greatly influenced by the curing period, as the CBR values increased from 3% to 14% after 7 days of curing and

from 5% to 25% after 28 days. The CBR increase reaches an average beyond 1.7, suggesting the presence of an optimal terrazyme dosage, beyond which further enzyme application may not significantly improve soil performance. The CBR values at 28 days are consistently higher than those at 7 days, indicating the long-term improvement in soil stabilization. Enzymatic reactions progressively refine the soil micro-structure, forming a denser, higher load-bearing sub-grade suitable for pavements and other geotechnical applications. The findings of this study on the increase in CBR align with previous research on the impact of enzymatic additives on soil engineering properties (Kushwaha et al., 2018; Mekonnen et al., 2020).



**Figure 8.** CBR of soil treated with different dosages of terrazyme at 7 and 28 days

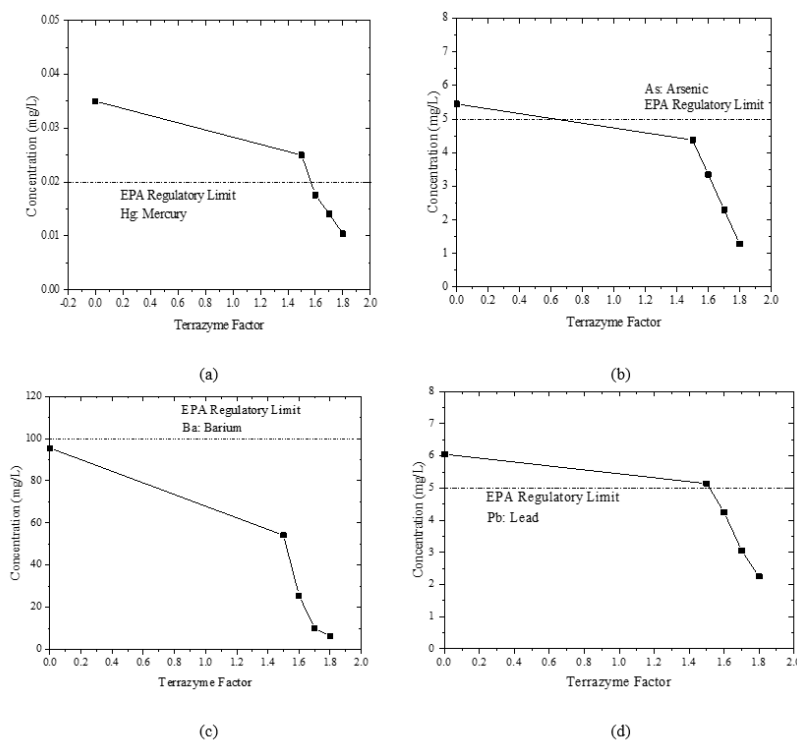
### TCLP Test Results

Compared with EPA regulation limits, the study

assessed the effect of terrazyme on heavy metal leaching. All terrazyme concentrations were successful

in bringing mercury (Hg) levels below the EPA limit of 0.02 mg/L; however, Fig. 9a shows that the most notable reduction (0.01 mg/L) occurred at a dosage of 1.8. This implies that terrazyme successfully lowers the toxicity caused by mercury. As seen in Fig. 9b, levels of arsenic (As) remained over the EPA limit of 5.0 mg/L even after terrazyme treatment gradually reduced concentrations. The involved chemistry of As might require the use of further stabilizing techniques, as seen by the lowest measured concentration (1.0 mg/L at 1.8 terrazyme), which suggests little effectiveness. As illustrated in Fig. 9c, all treated samples for barium (Ba) stayed within the EPA limit of 100.0 mg/L, with the greatest reduction (5.9 mg/L) occurring at 1.8 terrazyme concentration, indicating its effectiveness in immobilizing Ba. Terrazyme's capacity to reduce Pb leaching is further supported by the fact that Lead (Pb) levels remained below the EPA criterion of 5.0 mg/L, with the best reduction (2.0 mg/L) at 1.8 concentration (Fig. 9d).

Terrazyme was quite successful in bringing the levels of Hg, Ba, and Pb down to EPA standards; the best results were obtained at a concentration of 1.8. However, its inability to fully stabilize As suggests the need for complementary treatment methods. These findings highlight terrazyme's potential as a remediation agent while underscoring the necessity for further research on arsenic-specific solutions. Beyond leaching, there are important wider environmental effects. Terrazyme stabilization may lessen the long-term environmental responsibility and potential for groundwater pollution from treated soils, hence minimizing ecological and human health concerns, as evidenced by the successful immobilization of various heavy metals. Enzymatic stabilization is in line with sustainable remediation objectives, as, when examined through the perspective of a Life Cycle Assessment (LCA), this biocatalytic technique probably produces a lower environmental impact than conventional binders, like cement.



**Figure 9.** Change in heavy metal concentration with variation in terrazyme dilution factor for (a) Mercury (b) Arsenic (c) Barium (d) Lead

### Scanning Electron Microscopy

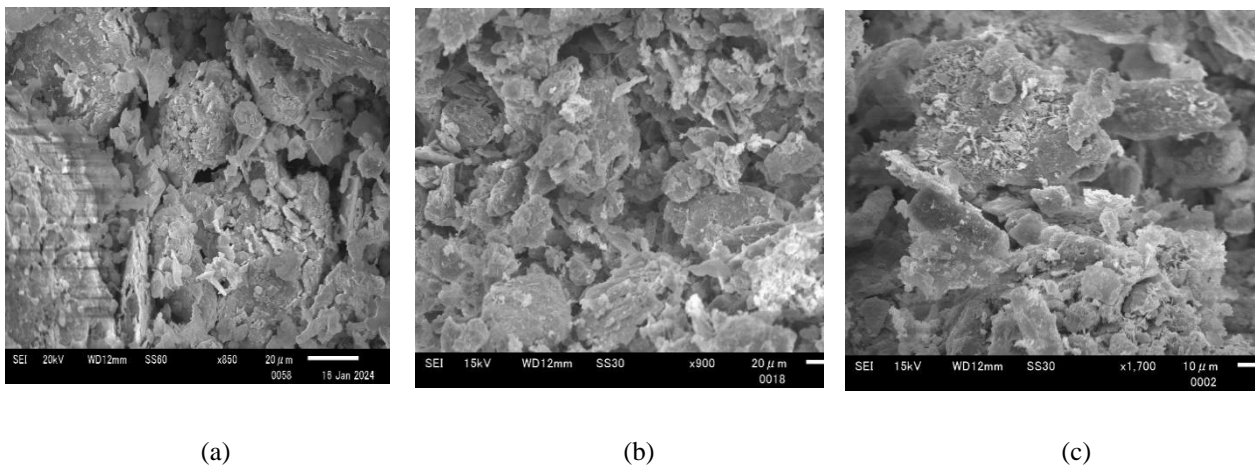
To observe the microstructural changes over time, terrazyme-treated clayey soil was analyzed using Scanning Electron Microscopy (SEM) at various curing times, such as 1 day, 14 days, and 28 days. The SEM

images at day 0 of the curing period showed that the soil particles were still relatively loose, had poor inter-particle bonding, and with apparent pores, as shown in Fig. 10a. Although some initial fine particle aggregation was observed, the binding action of terrazyme was not



fully developed due to the short curing duration. There was little cementitious compound production, and the structure still showed a dispersed distribution. After 14 days of curing, the micro-structure underwent significant changes, as seen in Fig. 10b. The SEM images displayed enhanced flocculation and particle aggregation, leading to a denser arrangement (Nadeem et al., 2023). The bonding of clay particles was supported by the enzymatic process, which decreased voids and increased soil compaction. The stability and strength of the soil were improved by the presence of gel-like substances, most likely CSH or other cementitious products. By the 28<sup>th</sup> day of testing, the

SEM analysis showed an extremely compact and closely bound soil matrix. As seen in Fig. 11c, the spaces were significantly decreased and the inter-particle interactions were more noticeable, suggesting strong cohesion between soil particles. A dense, interlocking structure has formed as a result of the complete development of the enzymatic stabilization. The soil's general stability was further enhanced by the cementitious compounds, which were more noticeable and filled the remaining pores. This change in soil micro-structure demonstrated that terrazyme gradually improved clayey soil's engineering characteristics.



**Figure 10.** SEM analysis (a) 0-day curing (b) 14-days curing (c) 28-day curing

When compared with conventional stabilizing methods, the increase in strength and decrease in flexibility is especially noticeable. Even though conventional agents, like cement and lime, offer significant cation exchange and cementitious pozzolanic products, they typically elevate the pH of the soil and need energy-intensive, carbon-intensive production procedures. In contrast, terrazyme acts as a biocatalyst, improving the properties of soil by means of biological reactions at a pH that is almost neutral. This makes it a potential lower-carbon choice for projects the main goal of which is not to achieve exceptionally high strength. From an economic standpoint, enzymatic stabilization presents an argument for practical use. The required dosage is typically relatively low (often less than 1% by volume) in comparison to dosages of lime or cement, which are typically given at 3%-8% by weight. Additionally, terrazyme-treated soils can frequently be compacted approximately to their natural moisture

content, saving money, time, and effort when drying or wetting to reach the ideal moisture content. It is a financially feasible choice for large-area stabilization projects, like sub-grade preparation and erosion control, because of its low material consumption and improved construction process, which can lead to noticeably lower overall project costs.

## CONCLUSIONS

This study presents the effects of terrazyme on the mechanical properties of clayey soil that was exposed to compaction, Atterberg's limit, TCLP and CBR tests.

- The soil's MDD is 19.71 kN/m<sup>3</sup> at an OMC of 13.45%. At a dosage of 1.6, the MDD rose from 19.71 kN/m<sup>3</sup> to 24.02 kN/m<sup>3</sup>, whereas the OMC declined from 13.45% to 9.45% as the terrazyme concentration increased.
- The addition of terrazyme decreased LL from 52% to

40% at a dosage of 1.7, PL increased from 29% to 32% and PI decreased from 23% to 8% overall. Terrazyme treatment improves overall stability by decreasing soil flexibility, which lessens susceptibility to volumetric changes, like swelling and shrinkage.

- The UCS of the natural soil was 150 kPa. The UCS showed a peak value of 156.2 kPa at a dosage of 1.7, demonstrating the greatest strength enhancement resulting from terrazyme stabilization.
- After 28 days of curing, the UCS rose from 150 kPa to 300.5 kPa, suggesting that terrazyme gradually strengthens the soil by enhancing densification and inter-particle bonding.
- After 7 days of curing, the CBR values increased from 3% to 14%, and then from 5% to 25% after 28 days. Terrazyme improves the micro-structure of the soil, creating a denser, more durable sub-grade that is appropriate for geotechnical and pavement applications.
- Terrazyme proved to be highly effective in reducing Hg, Ba, and Pb concentrations to within EPA limits, with the 1.8 concentration yielding optimal results

and demonstrating its potential as an environmentally friendly stabilizing agent.

## RECOMMENDATIONS

These results might only be applicable to particular soil types and environmental circumstances. More research is required to examine the performance of terrazyme stabilization on 48 days and 56 days across a variety of soil types in order to assess its wider application. In order to evaluate the long-term durability and impact of terrazyme stabilization under various geotechnical situations, future research should compare its performance with that of other additive types.

## Conflict of Interests

The authors declare that there is no conflict of interests.

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