



Performance Evaluation of Terrazyme as Soil Stabilizer

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ABSTRACT: Literature published TerraZyme stabilization case studies are analyzed rationally for evaluating the performance of TerraZyme as a stabilizer. As a measure of the degree of stabilization, the percentage variation in soil index and engineering properties induced by enzyme treatment for various soil types are investigated. The literature shows inconsistent enzyme stabilization results. The causes of the inconsistency in test results and factors affecting the effectivity of enzyme stabilization are elaborated. The effect of soil characteristics on the efficacy of enzyme stabilization is analyzed. The results of enzyme treatment on soil properties such as Atterberg limits and indices, optimum moisture content, maximum dry density, unconfined compressive strength and California bearing ratio, for various soil groups with different enzyme doses and conditions of curing are also studied. An attempt has been made to quantify the attribute variation of treated soils, establishing, the underlying reasons, for the effectivity of enzyme stabilization and its performance within and among various soil groups. The effect of enzymatic treatment on each soil group is also classified. The research emphasizes the necessity of an elaborate organized study on the enzyme and soil characterization, to have a better knowhow of the effectiveness of enzymatic stabilization.

Keywords: Degree of Stabilization, Enzymes Stabilizers, Soil Classification, Soil Stabilization, Terrazyme.

1. Introduction

For improvement in engineering properties, the soil is stabilized either by chemical, or mechanical treatment or by other non-traditional methods such as Enzyme treatment (Mekonnen et al., 2020).

The enzyme treatment minimizes the mechanical compacting efforts and results in higher soil density, shear strength and lower permeability (Taha et al., 2013). Compared to other conventional stabilizers, the enzymes are cost-efficient,

environmentally sustainable and convenient to use. Conventional soil stabilizers like lime and cement lead to environmental pollution, with carbon emissions. Enzymes are also, energy-efficient as they can reduce the compacting efforts required for mechanical stabilization. As a biological system catalyst, enzymes check the reaction rate and lower the activation energy essential for the new product formation thus facilitating the conversion state (Scholen, 1995). The enzymes are more efficient compared to inorganic catalysts. They

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expedite the reaction rate up to 106 to 1012 times. Enzymatic reactions are sensitive to temperature and act better at moderate temperatures (35° C). The enzyme reactivity is adversely affected by high temperatures. Enzymes are also pH-sensitive and efficient at a pH around the value of 7 (Ryan and Norris, 2014).

TerraZyme is an electrochemical product with fermented vegetable extract, mainly consisting of nonionic surfactants and carbohydrates. It is reactive with soils with clay contents. As per the recommendations by Saini and Vaishnava (2015) the soil for effective TerraZyme stabilization should have a liquid limit of less than 30%, particle size passing 75 micron greater than 15%, clay content greater than 6%, pH between 4.5 to 9.5 and soil temperature greater than 15 °C,

The soil sample analysis summary (Nature Plus Inc., 2021) for the TerraZyme application consists of an enquiry into, the work type and soil test data. The soil data consists of results of sieve analysis, Atterberg limits (liquid limit WL, plastic limit PL), Plasticity Index (PI), compaction characteristics (Standard Proctor test), unsoaked California Bearing Ratio (CBR) value and soil pH. These are the assessment parameter for checking the soil suitability, enzyme dose requirements and effectivity of the proposed enzyme stabilization (Myint and Swe, 2014; Shaka and Shaka, 2016).

2. Literature Review

According to Tingle et al. (2007), stabilizing the performance of enzymes is soil-specific. Enzymes are suitable for the treatment of water affine, clayey soils with high plasticity and with organic content. According to Woll et al. (2008) in several field applications, the enzyme performance was adversely affected due to the deficiency of clay/fine content. The initial reaction of the enzyme with the organic matter in the soil forms a gel which after crystallization forms a bond with soil particles. Loss of

moisture by evaporation is essential for the crystallization of gels.

Organic matter and clay content are crucial for the enzymatic reaction, bond formation and consequent stabilization. For an effective enzymatic reaction, the soil must have the necessary clay contents. According to Bergmann (2000), at least 2% clay/fine is essential for enzymatic stabilization, and 10% to 15% clay content to ensure good stabilization. Enzyme products are effective for soils having 18% to 30% clay fines, a plasticity index between 2% to 10%, and a liquid limit of up to 30% (North Dakota DOT, 2014). According to Kestler (2009), the soil should have a plasticity index greater than 8% and a minimum clay content of 10%. Effective stabilization is obtained for soils with, plasticity indexes between 8 and 35% and clay content between 12 and 24% (even up to 30% for a few enzymes). The degree of stabilization is high at a water content 2-3% lesser than the Optimum Moisture Content (O.M.C.).

Literature also mentioned of treated SC, SM-GM and SP-Soils with 0-2% clay content also recorded some improvements in soil properties. However, as observed by Shankar et al. (2009) enzymatic stabilization is inefficient for soils having high cohesionless content.

Bio-Enzyme stabilization with suitable doses improves the index and engineering properties of soils and reduces the compressibility of soil. The improvement in the properties may be due to the reaction of bio-enzyme with soil minerals and other chemically active constituents. Hence the stabilizing effect of bio-enzyme on soil should be tested in the laboratory before the field application (Guthrie et al., 2015).

Bio-enzyme stabilized soils show a considerable improvement in shear strength. Shukla et al. (2003) observed that bio-enzyme stabilization of different soils with variable clay content has witnessed little to high improvement in physical properties due to their variable degree of reactivity with bio-enzymes. For silty to

sandy soils, there was an appreciable improvement in CBR and UCS values. A study on the effect of the enzyme on lateritic soil using UCS, CBR, compaction and permeability showed medium improvement in the physical properties of lateritic soil. Whereas it was ineffective in improving the consistency limits. Lateritic soil (Wl and PI greater than 25% and 6%, respectively) with higher enzyme dosage (200 ml / 2 m³ of soil) results in a 300% increase in CBR, a 450% increase in UCS and a 42% decrease in permeability for four weeks of curing. The enzyme is also ineffective for cohesionless soil. Venkatasubramanian and Dhinakaran (2011), tested three soils with Wl = 28%, 30%, 46% and PI 6%, 5% and 6%, respectively, stabilized with different enzyme dosages. The increase in UCS was 152 to 200% and CBR was 157 to 673%, respectively, after 4 weeks of curing. According to Isaac et al. (2003) the lateritic soil and clayey soil for a curing period of 8 weeks TerraZyme, has increased the CBR in the range of 136 to 1800 times the untreated soil value. Whereas the CBR increase was less significant about 700 percent silty soils and clayey-sandy soil

Rauch et al. (1993) analyzed the effects of ionic, enzyme, and polymer stabilizers on two high-plasticity natural clays and three clays with one of the predominant clay minerals either kaolinite, illite, or sodium montmorillonite. Overall, there was no marked improvement in Atterberg limits, density, shear strength and swell potential of soils, still, there were individual cases of improvement. It was also concluded that higher doses may produce improvements in soil properties. Tingle and Santoni (2003) observed that enzymes treated in two soils of low and high plasticity have shown no improvement in saturated and unsaturated UCS tests of the strength of either soil. UCS tests on residual soil with three enzymes by Khan and Taha (2015); however, did not observe any improvement. A CL was treated with three enzymes but no betterment was observed. Similar variable results were obtained with other proprietary

bio-enzyme such as Permazyme, EarthZyme etc. According to Milburn and Parsons (2004), compaction test (water content at 1% less than the optimum) on the bio-enzyme Permazyme 11-X treated ML and SM soils (fines 88% and 30%, LL 30% and 20% and PI 7% and 3%, respectively) at manufacturer's recommended dosage, the dry density increases by only 4% and 1%, respectively or stiffness, wet-dry and leaching tests no improvement was recorded for 28 days of curing. However, freeze-thaw indicates a very small improvement. Stabilization was done by Brandon et al. (2010) using bio-enzyme (Permazyme) on six single sources and three blended soils. There was a decrease in PI for soil-1 and soil-2 and an increase of 44% for soil-6. There is no definite trend of improvement in the properties of treated soils. However, there is an increase of 6 to 64% in cohesion due to the agglomeration of the soil particles. Mgangira (2009) observed little or no improvement in PermaZyme 11-X and EarthZyme treated two native soils and three reference clays (illite, kaolinite and montmorillonite).

The laboratory test results are also affected by the duration and conditions of sample curing studied the effect of air-dry curing (drying) and desiccator curing on the properties of Terrazyme stabilized black cotton soil and red earth for various curing periods. Unsoaked CBR, UCS strength of both Terrazyme treated black cotton soil and red earth showed tremendous improvement with drying than laboratory desiccator curing. Free Swell Index for black cotton soil showed better improvement with drying. The air-dry curing (or drying) condition proved more efficient in treating both soils than the desiccator curing condition. However, Atterberg limits for both black cotton soil and red earth did not exhibit any difference in either drying or desiccator curing (Daigavane and Ansari, 2021; Vastrad et al., 2020; Sen and Singh, 2015).

The studies show varying improvements in soil properties from very small to

significant degrees which necessitate laboratory testing before actual field application. Mostly the details of enzyme composition are provided by the manufacturers. Since the enzymes are often reformulated; their product-specific testing must be conducted to check their suitability. On contrary, the enzyme stabilizers have no specific laboratory tests available to assess their on-site performance. Due to the unfamiliarity of the enzyme manufacturers with the design process, the significant benefits of the non-standard enzyme stabilizers have remained unexplored (Scholen, 1992).

All the literature-suggested parameters are included in the study to check the efficacy of TerraZyme stabilization for various soil groups. The research on TerraZyme stabilized soils has witnessed, dissimilar experimental and field performance. Hence, the research attempted to generalize the stabilizing effects or performance of the TerraZyme on different soils and checks soil suitability for stabilization (Indian Standards: 1498, 1970).

3. Methodology

The enzyme stabilizers act but, they can be effective under certain conducive circumstances only. It is therefore essential that the correct stabilizer and soil type for stabilization are chosen. The variation in a characteristic of soils needs systematic study to determine the enzyme's stabilization performance. At present, soil stabilization by enzymes is predominately based on empirical guidance from past experiences and is not much subjected to technical evolutions. The study on enzyme stabilization concluded that these products work but, they can be applied successfully only in certain conditions. It is therefore imperative that the correct stabilizer and material for stabilization.

The literature indicates that the random variable results of enzyme stabilization also it is uncertain how and under which

conditions the enzymes work better. The published results from literature about both untreated and TerraZyme-stabilized soils are checked to evaluate the effectivity of TerraZyme treatment. The objectives of the study are:

- To check the characteristics of soils that effects enzyme-based soil stabilization.
- To investigate the changes in soil index and engineering properties induced by enzyme stabilization.
- Analysis of enzyme-treated soil data from the literature to study performance evaluation of enzyme performance with various soil types.
- To investigate enzyme dose requirements to stabilize the various soil types.

The improvement in index and engineering properties of enzyme-stabilized soil can be an evaluation parameter for checking the degree of stabilization achieved. The improvement in soil properties is examined to resolve the effect of enzyme doses, curing conditions, and the type of soil. The laboratory results of Atterberg limits and indices, compaction characteristics, CBR, and UCS have been analyzed based on variations in soil type, enzyme doses and curing duration. The percentage difference in these results either positive (% increase) or negative (% decrease) is analyzed.

An attempt has been made to quantify the attribute variation of treated soils, establish, the underlying reasons, and correlations and to categorized the treated soil behaviour as per the individual soil groups. The literature on enzyme stabilization data consists of lots of parametric variations concerning enzyme types, doses, curing duration and type. The study attempts to maintain the uniformity of these parameters to the extent possible. However, considering the large quantity of data, only the minimum and maximum parametric value of the parameter for soil, enzyme doses and duration of curing are analyzed and presented. The lower value indicates the minimum percentage variation

in the property over the untreated sample, whereas, a higher value indicates the maximum percentage variation in the property after stabilization.

Considering the variation in literature results, the aspects of factors affecting the results of laboratory enzyme stabilization such as the enzyme application rates, sample preparation, sample curing conditions, test procedure etc. are also investigated and discussed (Muguda and Nagaraj, 2019).

The bio-enzyme stabilization is certainly economical and more sustainable stabilization technique. Several case studies discussed the economic benefits of TerraZyme stabilization for the construction of low-cost rural roads, road base course and subgrade stabilization. The contents of the research paper emphasize mainly the evaluation of the enzyme's stabilization performance. Hence the economical aspect of enzyme stabilization is not elaborated on.

3.1. Characterization of Soils

The soil type greatly influences the effectiveness of enzyme stabilization. Hence for the basis of comparison, various soils are classified as per IS Classification System (ISCS) as shown in Figure 1.

For better characterization of soils, an attempt was made to assess the degree of expansion and severity for fine-grained soils based on Atterberg's limits, free swell index and the presence of clay and mineralogical contents based on literature studies. However, conclusive remarks could not be made due to the non-availability of uniform soil data. For some of the soils, these comparison parameters cannot be applied as values are not falling within the defined ranges. The reason may be the empirical nature of the test and the lack of standard procedures.

The percentage of clay/fines and the mineral contents influence the stabilization mechanism. The soils are also classified based on plasticity and free swell index to check their principal clay minerals as shown in Appendix. This has been used as a simple parameter to the degree of suitability of clayey soil for an enzyme treatment.

3.2. Laboratory Tests

Improvements in the properties of bio-enzyme stabilized soil may be due to the chemical reaction of soil constituents with bio-enzymes. One of the parameters to check the degree of chemical reaction of the enzyme with soil is to check changes in the microscopic characteristics of stabilized soils.

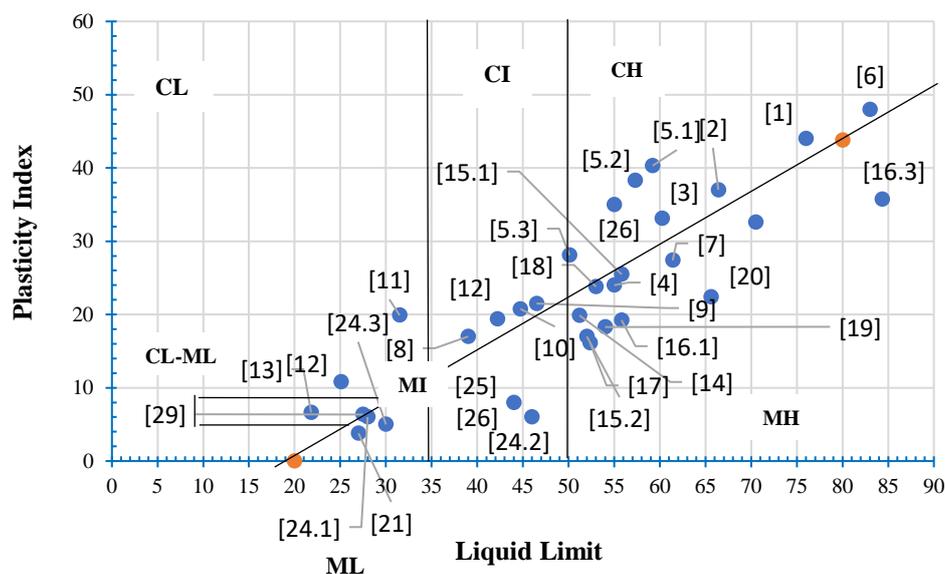


Fig. 1. Soil Classification for the soils included in the study

The changes in the microstructure of untreated and enzyme-treated soils with curing can be compared with advanced methods. (Indian Standard: 2720 (Parts 5,7,10,16), 1985). There are several methods suggested for checking the microstructural changes such as BET surface area analysis, Environmental Scanning Electron Microscopy (ESEM), X-Ray Diffraction (XRD) and X-Ray Fluorescence (XRF), Field Emission Scanning Electron Microscopy (FESEM), Scanning Electron Microscopy (SEM) (Zhang et al., 2013), etc. However, these micro-level observations seem to be too inconclusive to confirm the changes induced by enzymatic reactions with the curing. These changes are again variable with soil mineralogical contents.

Usually, the improvement in the index and engineering properties of stabilized soil is considered an evaluation parameter to check the degree of stabilization achieved. The consistency tests, proctor test, UCS, soaked/unsoaked CBR tests, Free Swell Index (FSI) test, swell pressure test, consolidation test, and permeability test may be carried out for evaluating the performance of stabilized soils. For the study, the result discussion is limited to the consistency test and the basic strength tests. These are the standard tests performed as per the procedure, material and equipment specifications defined by Indian Standards (IS). The section indicates the references and objectives of these tests.

- Index properties of the soil as per IS: 2720 (Part 5) (1985). The index properties such as liquid limit, and plastic limit are related to various properties of soil such as cohesion, and capillarity and also form the basis for soil classification and its specification as fill material.
- Compaction test as per IS: 2720 (Part 7) (1985). This test is used for determining the amount of water needed, for field compaction and as a measure of the degree of denseness that can be obtained at optimum moisture content. The water

density relation obtained from the test can be used for better control of these tests of the field compaction.

- Unconfined Compressive Strength (UCS) test as per IS 2720 (Part 10) (1985). UCS test is used for the determination of compressive and shearing strength of clayey soils not to be subjected to lateral pressure in an undrained condition.
- California Bearing Ratio (CBR) test as per IS: 2720 (Part 16) (1985). The CBR value is an index measuring the soil strength based on the condition of the material during the testing. It can be correlated with important parameters like the modulus of subgrade reaction, modulus of resilience and plasticity index.

3.3. Analysis of Stabilization Results for Various Soil Groups

There are several mixed examples from literature where this laboratory testing shows highly beneficial or no substantial improvement in the properties of enzyme-stabilized soils. These results are analyzed for enzyme stabilization.

3.4. Analysis for CH-Soil

In general, the CH-soil are clay of high plasticity, with Montmorillonite as the dominant clay mineral and with high expansivity and swelling potential.

3.4.1. Analysis of Consistency Limits for CH-Soil

Table 1 shows the maximum % variation in Atterberg limits for CH-soils. For CH-soil 1 maximum percentage reduction in liquid limit is 14.47% for an enzyme dose of 200 ml / 2 m³ of soil at the end of the 4th week. CH-soil 1 shows a continuous decrease in plastic limit with an increase in enzyme doses and period of curing. For CH-soil 1 there is a maximum reduction in PL of 28.13% at an enzyme dose of 200 ml / 2 m³ of soil at the end of the 4th week. (CH-soil 1- WL(%) = 76, WP (%) = 32, PI (%) = 44, Ws(%) = 8, IS(%) = 68 clay content is 21.61%, Active, highly plastic, specific gravity G = 2.62)

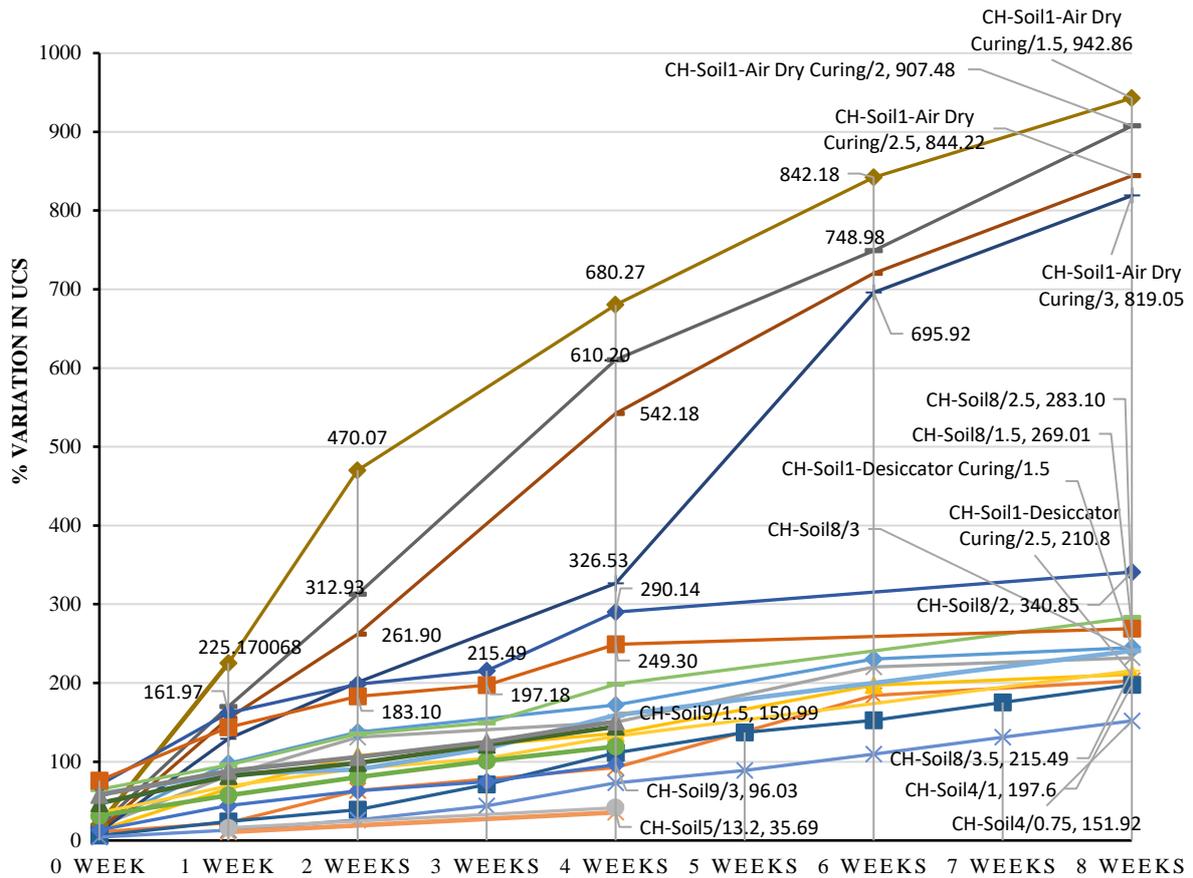


Fig. 2. Percentage variation in UCS for CH-soil

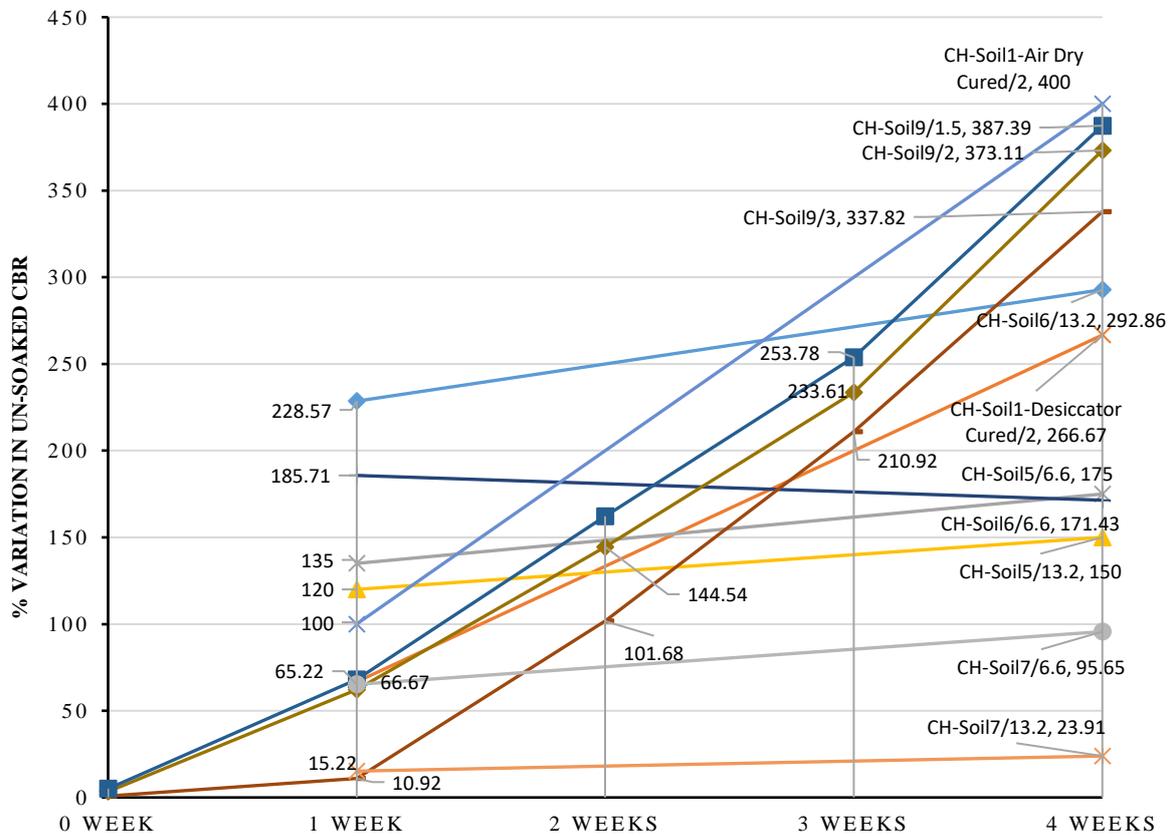


Fig. 3. Percentage variation in unsoaked CBR, CH-soil

UCS CH-soil1 gives a maximum percentage increase of 244.9% (Desiccator curing) and 942.9% (Air dry curing) for an enzyme dose of 200 ml /1.5 m³ of soil for 8 weeks of curing.

The maximum percentage increase in UCS value for CH-soils with enzyme doses of 200 ml / 1 m³, 1.5 m³ and 2 m³ shows uniform strength gain for a curing duration of 8 weeks. Whereas for CH-soil 5 (clay content 33%, FSI 170, Montmorillonitic) the percentage maximum increase in UCS value is 41.49% with 200 ml / 6.6 m³ enzyme dose for 4 weeks curing. The same soil shows a lesser improvement of 10.75% with a decrease in enzyme doses (200 ml / 13.2 m³) and curing duration (1 week)

Both CH-soil 4 (197.6%, 200 ml / 1 m³, 8 weeks) and CH-soil 9 (150.99%, 200 ml / 1.5 m³, 4 weeks) show comparable maximum percentage increase in UCS values may be due to the similar characteristic of CH-soil 4 (W_L(%) = 55, WP(%) = 31, PI(%) = 24 clay content is 66.78%, clay of high plasticity, specific gravity G = 2.45) and CH-soil 9 (W_L(%) = 61.4, WP(%) = 34, PI(%) = 27.4 clay content is 68.7%, clay of high plasticity, specific gravity G = 2.48, FSI = 72.8).

CH-soil 8, a BC soil (W_L(%) = 83, WP(%) = 35, PI(%) = 48, specific gravity G = 2.65, FSI = 78) has shown the maximum increase in UCS of 340.9% (200 ml / 2 m³, 8 weeks) due to its highly plastic nature.

3.6. California Bearing Ratio (CBR)

Test Results for Stabilized CH-Soil

Table 3 and Figure 3 show the % variation in CBR values-CH-soils.

CBR-CH-soil 1 air dry cured sample shows a CBR value increase of 400% as compared to 266.67% of increase for the

desiccator cured sample with the same enzyme dose of 200 ml/2 m³ of soil at the end of the 4th week.

Among all the CH-soils, CH-soil 5 has recorded the least increase in soaked CBR value at a lower enzyme dose of 200 ml /13.2 m³ (CH-soil 5- WL(%) = 59.2, WP(%) = 18.9, PI(%) = 40.3, clay content 33%, G = 2.72, FSI = 170, Montmorillonitic). Such similar low improvement in CBR is also observed for CH-soil 7 (15.22%-unsoaked and 33.33%-soaked CBR), at low enzyme dose. However, the soil CBR-CH-soil 6 shows an increase of 292.86% in CBR value at the lower dose of 200 ml / 13.2 m³, when compared with the higher dose of 200 ml / 6.6 m³ of the same soil at the end of the 4th week with unexpected reduction trend. However, the same CH-soil 6 with 200 ml / 6.6 m³ of the soil gives a 400% increase in CBR value compared to an increase of 366.67% for a dose of 200 ml /13.2 m³ of soil at the end of 1st week of curing. CH-soil 5 and CH-soil 7 have greater FSI of 150, and 170, respectively and high montmorillonite contained with high swelling and expansivity. These soil on stabilization shows a high decrease in plasticity index. The same soil shows only moderate gain in soaked CBR values (Un-soaked CBR 175%, 4 weeks, 95.65%, 4 weeks % and soaked CBR 60%, 1 week, 122.22%, 1 week, respectively). CH-soil 6 having 20.2% clay content show better performance for both unsoaked (292.86%, 4 weeks) and soaked CBR (400, 1 week). CH-soil 1 having less percentage clay size fraction (21.61%) show a higher gain in UCS (942.86%) as compared to CH-soil 8 (UCS 340.85%) with a 30% clay size fraction.

Table 2. Maximum% variation in UCS values - CH-soils

Soil	CH-soil 1		CH-soil 4		CH-soil 5	CH-soil 8	CH-soil 9				
Doses	Curing type										
200	Desiccator		Air dry		1	13.2	6.6	3.5	2	3	1.5
(ml/m ³)	3	1.5	3	1.5							
% variation in UCS	10.88,	244.9,	10.88,	942.9,	197.6,	10.75,	41.49,	35.21,	340.9,	13.31,	150.99,
	(0	(8	(0	(8	(8	(1	(4	(0	(8	(0	(4
	weeks)	weeks)	weeks)	weeks)	weeks)	week)	weeks)	weeks)	weeks)	week)	weeks)

Table 3. Maximum % variation in CBR values - CH-Soils

Soil	Doses 200 (ml/m ³)	% Variation (+ve increase, -ve decrease)		
		Un-Soaked CBR		Soaked CBR
CH-soil 1	2 (Desiccator curing)	66.67, (1 week)	266.67, (4 weeks)	-
	2 (Air Dry curing)	100, (1 week)	400, (4 weeks)	-
CH-soil 2	3.5	59.92, (0 weeks)		-
	0.25	207.72, (0-week)		-
CH-soil 3	0.25	480, (3 weeks)		-
CH-soil 4	1	-		71.64, (0 weeks)
	0.75	-		71.64, (0 weeks) 329.85, (8 weeks)
CH-soil 5	13.2	120, (1 week)		20, (0 weeks) 20, (1 week)
	6.6	175, (4 weeks)		60, (1 week)
CH-soil 6	13.2	292.86, (4 weeks)		300, (0 weeks)
	6.6	171.43, (4 weeks)		300, (0 weeks) 400, (1 week)
CH-soil 7	13.2	15.22, (1 week)		33.33, (0 weeks)
	6.6	95.65, (4 weeks)		122.22, (1 week)
CH-soil 9	3	0.84, (0 weeks)		-
	1.5	387.39, (4 weeks)		-

3.6.1. Analysis of CI-Soil

CI-soils are clays of intermediate plasticity with normal activity.

3.6.2. Consistency Limits for Stabilized CI-Soil

Table 4 shows the % variation in Atterberg limits in CI-soils. CI-soil 1 shows a maximum reduction of 10.26% in LL with a dose of 200 ml / 2.5 m³ for 4 weeks of curing after an initial increase of 12.82% in LL values in the first week. CI-soil 1 and CI-soil 2 both show a reduction in PL values up to 13.64% and 50% with an increase in dose and duration. The maximum percentage reduction in PL values for CI-soil 2 is 50% for or both doses of 200 ml / 0.8 m³ and 0.6 m³ of soil.

The soil CI-soil 2 shows a similar reduction trend in PI values with an increase

in curing duration. The maximum reduction for CI-soil 2 is 44.19% for an enzyme dose of 200 ml / 0.6 m³ of soil. However, PI values for CI-soil 1 decrease to 5.8 % at the end of the 4th week with an initial increase of 41.18% in the first week. However, the shrinkage limit for CI-soil 1 shows a continuous decrease with a maximum reduction of 22.22% for an enzyme dose of 200 bml / 2.5 bm³ of soil at the end of 4 weeks of curing.

3.7. Compaction Characteristics for Stabilized CI-Soil

As shown in Table 5 CI-soil 2 shows a maximum increase of 68.71% in MDD value. All CI soils show better improvements in MDD values for higher enzyme doses.

Table 4. Maximum% variation in Atterberg limits - CI-Soils

Soil	CI-soil 1		CI-soil 2		CI-soil 3	CL-soil 2	
	2.5	1.7	0.8	0.6	1.5	13.2	6.6
Doses 200 (ml/m ³)	2.5	1.7	0.8	0.6	1.5	13.2	6.6
	12.82, (1 week)	-15.91, (0 weeks)	NS	-68.82, (3 weeks)	NS	NS	NS
% Variation (+ve increase, -ve decrease)	LL	-10.26 (4 weeks)					
	PL	-13.64 (4 weeks)	46.4, (0 weeks)	-50, (3 weeks)	-50, (3 weeks)	NS	NS
	PI	41.18, (1 week)	58.14, (0 weeks)	-44.19, (3 weeks)	-44.19, (3 weeks)	NS	-42.59, (1 week)
		-5.88, (4 weeks)					-58.33, (4 weeks)
	Ws	-22.22, (4 weeks)	NS	NS	NS	25.38, (0 weeks)	-47.22, (1 week)
							-59.26, (4 weeks)

Table 5. Maximum % variation in OMC & MDD in CI-soils

CI-soil	Doses 200 (ml/m ³)	% (+ve increase, -ve decrease)			Remark
		OMC		MDD	
CI-Soil 2	1.7	-0.38, (0 weeks)	-0.38, (1 week)	21.84, (0 weeks)	Standard proctor test
	0.6	-18.08, (6 weeks)		68.71, (6 weeks)	
CI-Soil 3	Untreated	-17.78, (0 weeks)		9.26, (0 weeks)	Modified proctor test
	2	-23.70, (0 weeks)		13.30, (0 weeks)	

3.8. Unconfined Compression Strength (UCS) for Stabilized CI, CL and CL-ML Soils

Table 6 shows % variation in UCS in CI, CL and CL-ML soils. For UCS-CI-soil 1 there is a continuous incremental increase in UCS values and the maximum value reaches around 827.78% for 200 ml / 2.5 m³ of the soil over the duration of 8 weeks.

Whereas for the same CI-soil 1 with desiccator curing the percentage increase in UCS value is 158.94% for an enzyme dose of 200 ml / 2.5 m³ of soil over the duration of 8 weeks. CI-soil 1 shows a 357.14% increase in un-soaked CBR content with doses of 200 ml / 2.5 m³ at the end of curing duration of 4 weeks (Table 7)

CI-soil 2 having the highest clay content (53%) among the CI group shows the maximum decrease in LL, PL and PI and an increase in MDD values. CI-soil1 having the lowest clay content (18.24%) among the CI group shows the maximum increase in UCS values.

3.9. Analysis of CL-Soil

CL-soils are clays of moderate plasticity maybe with normal activity.

3.9.1. Consistency Limits for Stabilized CL-Soil

CL-soil shows a consistent reduction in liquid limit values with a maximum reduction of 68.82% and at the end of the 3rd week with an enzyme dose of 200 ml / 0.6

m³ of soil.

3.10. Unconfined Compression Strength (UCS) Results for CL-Soil

For UCS-CL-soil 2 with a dose of 200 ml / 13.2 m³ there is an increase of up to 56% in UCS value for the duration of 4 weeks. However, for the same soil with an enzyme dose of 200 ml / 6.6 m³ the increase in UCS value is up to 68% for the same duration of 4 weeks.

3.11. California Bearing Ratio (CBR) Test Results for Stabilized CL and CL-ML Soils

When compared among CL and CL-ML soils, CL soil having less clay content shows a greater percentage improvement in both Un-soaked and soaked CBR values (Table 8).

3.12. Analysis of CL-ML Soil

CL-ML soils are silty clays slightly plastic and may be normal or inactive based on their clay content. Unconfined Compression Strength (UCS) results for stabilized CL-ML soil, show that for CL-ML soil with the higher enzyme dose of 200 ml / 0.5 m³ increase in UCS is up to 375% over a duration of 4 weeks. This increase in the UCS value for CL-ML soil (PI = 6.64%) is comparatively more than CI (Avg. PI = 19.76%) and CL (Avg. PI = 15.35) soils may be due to its low plasticity index.

Table 6. Maximum % variation in UCS in CI, CL and CL-ML soils

Soils	CI-soil 1				CL-soil 2		CL-ML
	Desiccator curing		Air dry curing		13.2	6.6	0.5
Soil / Doses 200 (ml/m ³)	4	2.5	4	2.5			
% UCS (+ve increase, -ve decrease)	7.73, (0 weeks)	158.94, (8 weeks)	7.73, (0 weeks)	827.78, (8 weeks)	11.33, (1 week)	68, (4 weeks)	375, (4 weeks)

Table 7. Maximum % variation in CBR values for CI-Soils

Soil	Doses 200 (ml/m ³)	(+ve increase, -ve decrease)	
		Un-soaked (%)	Soaked CBR (%)
CI-soil 1	3	214.29, (4 weeks)	NS
	2.5	357.14, (4 weeks)	NS

Table 8. Maximum % variation in CBR in CL-Soils and CL-ML Soils

Soil	Doses 200 (ml/m ³)	(+ve increase, -ve decrease)	
		Un-soaked (%)	Soaked CBR (%)
CL-Soil 2	13.2	63.64, (1 week)	266.67, (4 weeks)
	6.6	218.18, (4 weeks)	333.33, (4 weeks)
CL-ML	Untreated	56.88, (4 weeks)	NS
	0.5	185.32, (4 weeks)	NS

3.13. Analysis of MH-Soil

MH-soils are silts of high plasticity and their activity may be variable based on their clay content.

3.13.1. Consistency Limits for Stabilized MH-Soil

Table 9 shows the % variation in Atterberg limits in MH-soils. Both MH-soil 2 and MH-soil 3 shows a linear decrease in liquid limit values. The maximum reduction of 24.73% and 46.15% for enzyme doses of 200 ml / 2 m³ of soil for MH-soil 2 and MH-soil 3, respectively at the end of 2 weeks of curing. Plastic limit values for MH-soil 2 and MH-soil 3 also show a reduction in values with curing duration. The maximum reduction was 47.23% and 60% for MH-soil 2 and MH-soil 3, respectively for the same enzyme dose of 200 ml / 2 m³ of soil at the end of 2 weeks of curing. Plasticity index values however show inconsistent variation with enzyme doses and duration. For MH-soil 2 variation behaviour in the plasticity index is inconclusive.

For MH-soils, consistency indices show an inconsistent decrease in values with enzyme doses and duration however the trend is inconclusive. MH-soil 3 (Clay content = 39.43%, G = 2.36 FSI = 48) shows a maximum decrease in LL and PL = 46.15% and 60%, respectively. MH-soil 2 (clay content = 40%, G = 2.26, FSI = 55) shows the maximum decrease in IP (21.51%) MH-soil 10 (clay content = 53.8%, G = 2.72, FSI = 93) shows the maximum increase in IP (30.36%).

3.14. Compaction Characteristics for Stabilized MH-Soil

Table 10 shows a very slight increase in MMD for MH-soil even at the high dose of enzyme contents

3.15. Analysis of Unconfined Compression Strength (UCS) Results for Stabilized MH-Soils

Table 11 shows the % variation in UCS in MH-soils. UCS-MH-soil 1 shows the inconsistent variation of UCS with time and doses with a final rise of nearly 38% and 14% with doses of 200 ml / 2.5 m³ and 2 m³, respectively. UCS shows an overall decreasing trend of values with a total decrease of nearly 43% for the dose of 200 ml / 3 m³ of soil. UCS-MH-soil 2 (Clay content = 40%, G = 2.26, FSI = 55) shows the maximum increase in IP (30.36%) shows a continuous rising UCS raising trend, for all the enzyme doses. Though for all the doses with one-week curing the % increase in UCS is similar. There is a maximum increase of 135% for an enzyme dose of 200 ml / 2 m³ of soil at the end of the fourth week. For UCS-MH-soil 3 there is a rise in UCS for all enzyme doses. However, the dose of 200 ml / 2 m³ of the soil gives the highest increase of nearly 71% this dose also shows a high initial gain of strength of nearly 42% at the end of the first week. For UCS-MH-soil 7 there is an overall trend of consistent linear increase in percentage UCS values with a nearly 74% increase in UCS for a dose of 200 ml / 0.5 m³ of soil over the duration of 4 weeks. Similar is a rising trend of UCS values for MH-soil 9. For UCS-MH-soil 10 there is an

increase of nearly 77% for a dose of 200 ml / 6.6 m³ at the end of the 4th week.

3.16. California Bearing Ratio (CBR) Test Results for Stabilized MH-Soils

Figures 4 and 5 and Table 1² show % variation in CBR for MH-soils. CBR MH-

soil 2 gave a maximum % increase in soaked CBR value of 423.08% for 4 weeks of curing with a dose of 200 ml / 2 m³ of soil. MH-soil 2 (Clay content = 40%, G = 2.26, FSI = 55) shows the maximum increase in CBR. (Prakash and Sridharan, 2004)

Table 9. Maximum % variation in Atterberg limits for MH-soils

Soil	MH-soil 2			MH-soil 3		MH-soil 8			MH-soil 10
	2.5	2	1.5	2	1.5	Untreated	0.4	0.1	6.6
doses 200 (ml/m ³)									
LL	-	-24.73, (2 weeks)		-46.15, (2 weeks)	-	-	-	--	-
PL	-	-47.23, (2 weeks)		-60, (2 weeks)	NS	0.18, (0 weeks)		-18.42, (0 weeks)	NS
PI	-21.51, (2 weeks)	-	13.8, (1 week)	-17.65, (2 weeks)	29.41, (0 weeks)	-2.82, (0 weeks)	-8.98, (0 weeks)	-	30.36, (1 week) -16.52, (4 weeks)

Table 10. Maximum % variation in OMC & MDD for MH-soil and ML-soil

Soil	Doses 200 (ml/m ³)	% OMC (+ve increase, -ve decrease)		% MDD (+ve increase, -ve decrease)	
MH-soil 8	Untreated	-2.22, (0 weeks)	NS	-0.02, (0 weeks)	NS
	0.4	-4.89, (0 weeks)	NS	1.28, (0 weeks)	NS
	0.3	-11.11, (0 weeks)	NS	5.83, (0 weeks)	NS
	0.1	-6.67, (0 weeks)	NS	2.58, (0 weeks)	NS
ML-Soil	3	-41.18, (1 week)	-41.18, (2 weeks)	0.00, (1 week)	0.56, (2 weeks)
	2.5	-17.65, (1 week)	-11.76, (2 weeks)	2.12, (1 week)	4.47, (2 weeks)
	2	-41.18, (1 week)	-17.65, (2 weeks)	1.84, (1 week)	5.03, (2 weeks)
	1.5	-23.53, (1 week)	-17.65, (2 weeks)	1.12, (1 week)	2.79, (2 weeks)

Table 11. Maximum % variation in UCS in MH-soils

Doses Soil	MH-soil 1		MH-soil 2		MH-soil 3		MH-soil 7		MH-soil 9		MH-soil 10		
	3	2.5	2	1.5	2.5	2	1.5	0.5	3	1.5	13.2	6.6	
% UCS kPa (+ve increase, -ve decrease)													
200 (ml/m ³)													
	-42.7, (4 weeks)	109.60, (1 week)	135, (3 weeks)	51.6, (1 Week)	109.8 (3 weeks)	12.9, (1 week)	70.9, (3 weeks)	5.13, (0 weeks)	73.3, (4 weeks)	10, (0 weeks)	77.5, (4 weeks)	2.62, (1 week)	76.5, (4 weeks)

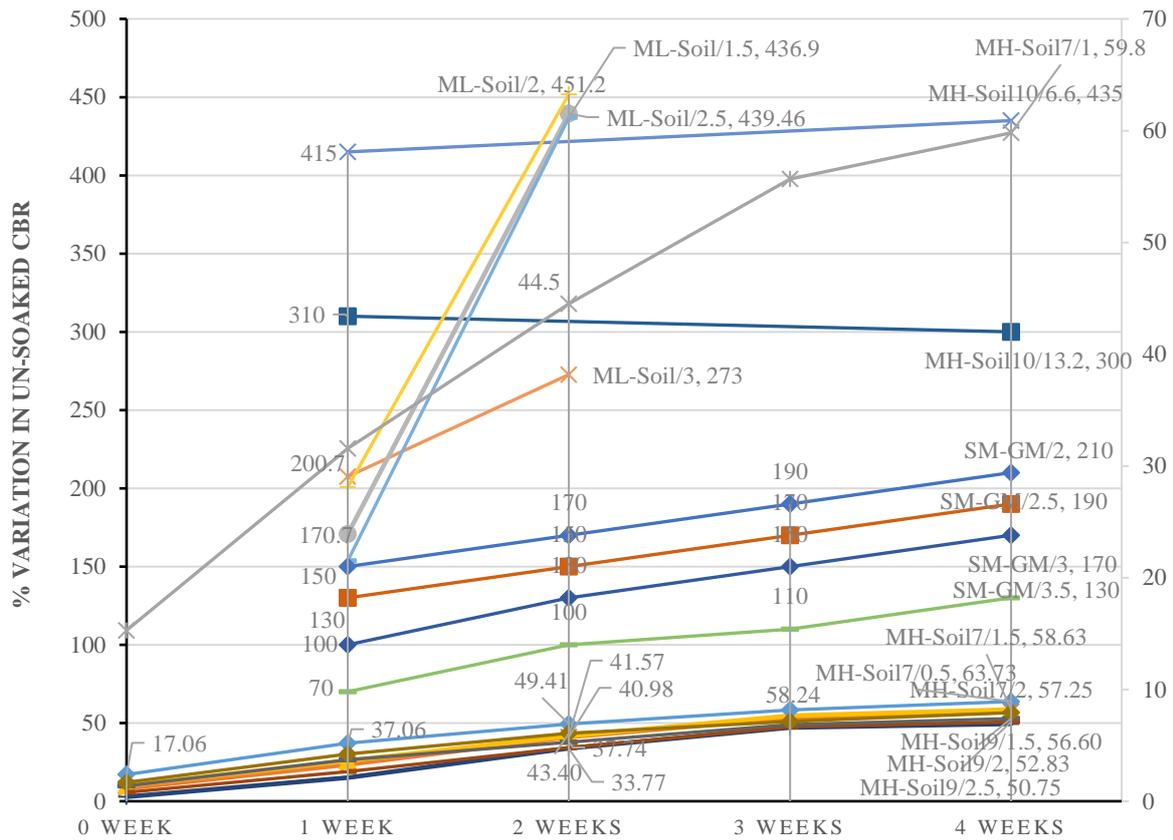
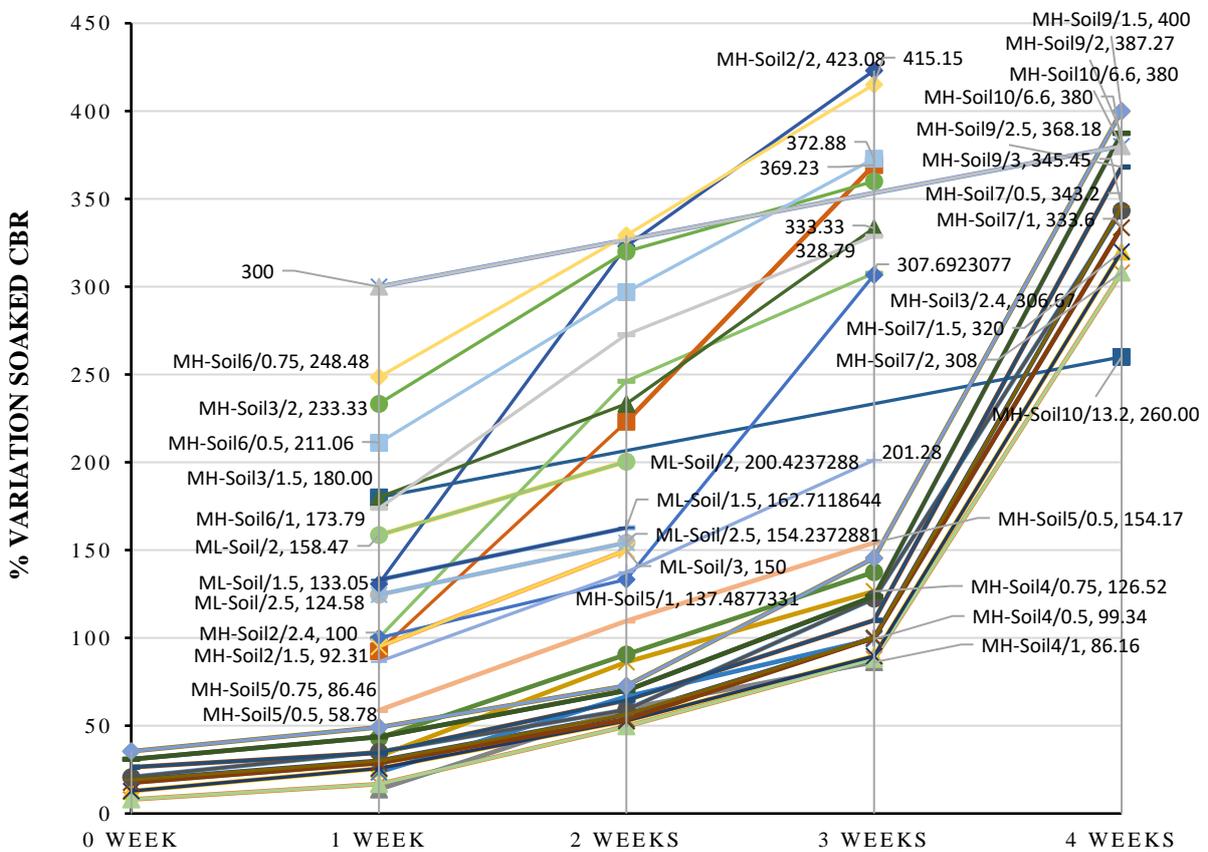


Fig. 4. % variation in unsoaked CBR-MH soils



CBR MH-soil 6 (Clay content = 92%, $G = 2.78$, $FSI = 90.5$) shows a similar high increase of 415.15% for an enzyme dose of 200 ml / 0.75 m³ of soil. CBR MH-soil 3 gives the maximum percentage increase of 360% for a dose of 200 ml / 2 m³ of soil for 4 weeks of curing. CBR MH-soil 4 showed a continuous increase in soaked CBR up to 126.52% for an enzyme dose of 200 ml / 0.75 m³ of soil for 4 weeks of curing.

CBR MH-soil 7 shows an incremental increase in unsoaked CBR with an increase in doses and period of curing with a dose of 200 ml / 0.5 m³ of soil giving the highest increase of 63.73% at the end of the 4th week. MH-soil 9 shows a similar increase of 56.60% in unsoaked CBR for the same dose and duration of curing. However, both MH-soil 7 and MH-soil 9 give the minimum increases in the CBR values as compared to other soils. CBR MH-soil 7 and CBR MH-soil 9 show a similar consistent increase in the percentage soaked CBR with the maximum value of 343.20% and 400% for a dose of 200 ml / 0.5 m³ of soil and 1.5 m³ of soil respectively for 4 weeks curing. The % variation of soaked CBR for MH-soil 7 and MH-soil 9 showed similar trends with a

maximum percentage increase of 343.20% and 400%, respectively at the 4th week of curing. Both MH-soil 7 ($W_L(\%) = 52.35$, $WP(\%) = 36.20$, $PI(\%) = 16.15$ clay size 20.4, $G = 2.61$ silt of high plasticity, moderately plasticity with normal activity) and MH-soil 9 ($W_L(\%) = 54$, $WP(\%) = 35.71$, $PI(\%) = 18.29$, $G = 2.63$ Clay size 20.2, the silt of high plasticity, high plasticity with normal activity) have similar properties showing similar improvement on enzyme stabilization

CBR MH-soil 10 ($W_L(\%) = 65.6$, $WP(\%) = 43.2$, $PI(\%) = 22.4$, clay content = 53.8%, $G = 2.72$, elastic silt of high plasticity $FSI = 93$). Table 12 shows a very high percentage increase in unsoaked CBR values of 300% and 435% for enzyme dose of 200 ml / 6.6 m³ of soil at the end of the first and 4th week, respectively. The high improvement in unsoaked CBR value for MH-soil 10 may be due to the well-graded nature of soil with higher clay and silt content.

3.17. Analysis of ML-Soil

ML-soils are silts of low plasticity, slightly plastic in nature and may be inactive.

Table 12. Maximum% variation in CBR in MH-soils

MH-soils	Doses 200 (ml/m ³)	(+ve increase, -ve decrease)	
		Un-soaked CBR (%)	Soaked CBR (%)
MH-soil 2	2	NS	423.08, (3 weeks)
	1.5	NS	92.31, (1 week)
MH-soil 3	2.4	NS	100, (1 week)
	2	NS	360, (3 weeks)
MH-soil 4	1	NS	13.67, (1 week)
	0.75	NS	126.52, (3 weeks)
MH-soil 5	1	NS	42.98, (1 week)
	0.75	NS	201.28, (3 weeks)
MH-soil 6	1	NS	173.79, (1 week)
	0.75	NS	415.15, (3 weeks)
MH-soil 7	Untreated	6.86, (0-week)	NS
	2	NS	8.00, (0 weeks)
	0.5	63.73, (4 weeks)	343.20, (4 weeks)
MH-soil 9	3	2.83, (0 weeks)	19.09, (0 weeks)
	1.5	56.60, (4 weeks)	400, (4 weeks)
MH-soil 10	13.2	300, (4 weeks)	180, (1 week)
	6.6	435, (4 weeks)	380, (4 weeks)
ML-soil	3	NS	94.92, (1 week)
	2	451.79, (2 weeks)	200.42, (0 weeks)
SM-GM	1.5	153.57, (1 week)	NS
	3.5	70, (1 week)	NS
	2	210, (4 weeks)	NS

3.17.1. Consistency limits for ML-Soil and SM-GM Soils

Table 13 shows the maximum decrease of 27.78% in LL, the PI value increases initially and then decreases by 7.65% for an enzyme dose of 200 ml / 2 m³.

The SM-GM soil shows better stabilizing results with the maximum decrease in PI values of 63.3% 200 ml / 2 m³ at the end of the 4th day. The improvement may be due to the well-graded nature and lesser 2% clay content.

3.18. CBR for ML-Soil

ML-Soil recorded a 451.79% increase in un-socketed CBR value for a dose of 200 ml / 2 m³ of soil. At the end of the 2nd week. ML-Soil registers a maximum percentage increase of 200.42% for 200 ml / 2 m³ of soil in soaked CBR for 2 weeks of curing.

3.19. CBR for ML-Soil

SM-GM are poorly graded silty sands or poorly graded sand with silt and gravel.

3.19.1. Unconfined Compression Strength (UCS) Results for Stabilized SM-GM and SP-Soils

Table 14 shows the % variation in UCS in SM-GM and SP-soils. UCS-SM-GM soil shows a maximum gain of nearly 450% of UCS with an enzyme dose of 200 ml / 2 m³ of soil. All the doses show a consistent rising trend of UCS values.

3.20. California Bearing Ratio (CBR) Test Results for Stabilized SM-GM Soil

CBR SM-GM soil shows a linear percentage increase in values of CBR with a maximum percentage increase of 210.00% for a dose of 200 ml / 2 m³ of soil at the end of the 4th week. The sieve analysis indicates a lesser percentage of clay, and silt and the presence of sandy and gravelly content, SM-GM soil resister higher CBR values maybe because of the inherent strength of sand and gravel matrix bonded with enzyme-treated silt content.

3.21. Analysis of SP-Soil

SP-soils are poorly graded sands with little or no fines.

3.21.1. Unconfined Compression Strength (UCS) Results for Stabilized SP-Soil

UCS-SP-soil 1 shows a consistent increase in UCS values with the duration and doses, the highest rise of nearly 426.34% for intermediate enzyme dose of 200 ml / 2.5 m³ of soil at the end of the 4th week. The same UCS-SP-soil 1 at dry of OMC however gives much higher overall consistent values. The maximum increase in UCS is nearly 475% for a dose of 200 ml / 2 m³ of soil (Table 14). Also, enzyme treatment provides better stabilization at the moisture content of 2% to 3% lesser than the optimum (OMC). The same UCS-SP-soil 1 on the wet side of optimum gives the highest increase of nearly 339% for an enzyme dose of 200 ml / 2 m³ of soil the maximum percentage increase in UCS values for SP-soil 1 is obtained at the dry of optimum. However, UCS-SP-soil 2 shows a maximum percentage increase of 111.45% for a dose of 200 ml / 2 m³ of soil at the end of the first week. SP-soil 1 shows high UCS values as compared to SP-soil 2 despite both having similar clay content of 1% and 2.45%, respectively

SC-soils are clayey sands (sand-clay mixtures) with plasticity and may be normal to inactive based on their clay content. SP-SC soils are poorly graded sands with clay with slight plasticity. SM-soils are silty sands (sand-silt mixtures) with slight plasticity, generally inactive. The results for enzyme stabilization of these soils could be concluded due to insufficient data.

4. Factors Affecting Results

Laboratory tests for mapping improvements in index and engineering properties of stabilized soil are recommended before field applications. However, the results describing the efficacy of enzymes for soil stabilization vary greatly among available literature.

Table 13. Maximum % variation in Atterberg limits in ML-Soils and SM-GM soils

Soil	ML-soil				SM-GM				
	Doses 200 (ml/m ³)	3	2.5	2	1.5	3.5	3	2.5	2
% Variation (+ve increase, -ve decrease)	LL	NS	NS	-27.78, (2 days)	-3.70, (1 day)	-14.29, (1 day)	-14.29, (1 day)	NS	-21.14, (4 days)
	PL	-33.22, (2 days)	-24.6, (1 day)	NS	NS	NS	NS	-4, (4 days)	-4, (4 days)
	PI	NS	71.50 (1 day)	-7.65 (2 days)	NS	-50 (1 day)	-50 (1 day)	NS	-63.3, (4 days)

Table 14. Maximum % variation in UCS for SM-GM and SP-Soils

Soil doses 200 (ml/m ³)	SM-GM		SP-soil 1				SP-soil 2			
	3.5	2	OMC		Dry of OMC		Wet of OMC			
(+ve increase, -ve decrease)	(1 week)	(4 weeks)	3	2	3	2	3	2	3	2
%UCS	44.37,	450.7,	101.88,	440.8,	216.64,	475.85,	63.60,	339.37,	-42.97,	111.45,
	(1 week)	(4 weeks)	(0 weeks)	(4 weeks)	(0 weeks)	(2 weeks)	(0 weeks)	(2 weeks)	(4 weeks)	(1 week)

Though the analysis of laboratory test results is generalized for various soil groups, the study indicates the inconsistency of results even within and obviously across the soil groups without any conclusive reasons. This variation of the results also necessitates considering the factors that may affect test results. Hence the possible reasons affecting the effectivity of enzyme stabilization and laboratory test results are also elaborated. The factors influencing the laboratory results are enzyme application rates, sample preparation, sample curing conditions test procedure etc.

- Enzyme application rates: The literature test results show the variation in properties of treated soil with enzyme doses within and across the soil groups. The method of enzyme application and calculation of enzyme doses are also variable in research studies. Most of the studies have preferred enzyme doses calculation based on the bulk unit weight of soil whereas few have preferred dose calculation based on dry unit weight. Though the difference is small, still it can affect the results.
- Sample preparation and tests: During specimen preparation, initial moisture content, enzyme doses, method of enzyme application, method of curing controlled conditions during curing, temperature control, etc. will have a pronounced effect on test results. Hence

needs to be monitored with utmost care. However, only a few authors have categorically mentioned these standards.

- Curing condition: The curing may be; air-dry at normal room temperature or in a sealed container under controlled conditions for preserving moisture content during the curing time. However, most of the studies discussed, have not specified details about the curing conditions. The improper curing conditions may have led to variations in test results. The controlled untreated samples also need to be tested for any thixotropic or ageing/curing strength gain specifically during UCS and CBR tests. Part of this significant improvement could be due to moisture loss as the moisture content variation during the sample preparation and testing stages was not categorically checked for many studies.
- Other factors: The enzymes are pH-sensitive too and work well around pH value 7 suggested that soil suitable for bio-enzyme stabilization should have some organic content. However, the laboratory studies have not specified organic contents and pH for the untreated soil. These parameters may have led to inconsistent, variable results among and within the soil groups. It is also a better idea to check and standardize the enzyme constituents to assured their consistent performance

which was also lacking in many of the studies. According to Aswar et al. (2022) along with the parameters of soil such as clay content, activity number (A), free swell index and mineralogical and organic content, the characterization of enzyme constituents' properties also need to be analyzed to understand the degree of the enzymatic reaction and the final stabilization.

5. Results

- All CH-soils show a reduction in PI and an increase in shrinkage limit with curing duration. CI-soils also show a reduction in PI values.
- The most prominent variation (negative sign indicates decrease) in consistency limits of untreated and treated soils (LL = -68.82%, PL = -50%, PI = -59.26%, Ws = +25.38%) was observed for CI-soils (average % clay content 32.25%). Also, among clayey and silty soils, the CI-soils have shown the maximum increase in dry density (+68.71%) and maximum decrease in OMC (-23.70%).
- There is substantial variation in optimal doses and curing period to obtain the maximum average % MMD for various soils (CH-Soils: 200 ml / 1.5 m³ curing duration 0 weeks, CI-soils: 200 ml / 0.6 m³ curing duration 6 weeks). CH-soils need higher doses to obtain the maximum average % MDD. The most marked variation in MDD and OMC was witnessed by the CI-soils, which show the best improvement in MDD value (+68.71%).
- The improved performance of CI soil may be because of lower the average clay/fine content among the clayey soils (Average % clay content CH = 36.82%, CI = 32.25%, CL = 39.40%, CL-ML = 59.00%, MH = 54.95%) and thus lesser PI (average PI values CH = 35.59%, CI = 19.76%, CL = 15.35%, CL-ML = 6.64%, MH = 23.13 %) of CI soils. Also, the comparatively well-graded nature may have resulted in improved compaction characteristics.
- TerraZyme stabilized CI, CL-ML, and CH and witnessed maximum betterment in UCS with curing periods. These clayey soil groups with significant clay/fine content, however, need a more curing period to achieve the maximum UCS. (CH = +942.9%, 8 weeks, CI = +827.78%, 8 weeks, CL-ML +375%, 4 weeks). However, as clay contents go on decreasing the time (curing duration) to reach maximum UCS also reduces.
- The contribution to UCS improvement may be contributed to enzymatic reaction with clay/fines or other factors such as thixotropic properties and/or reduction in water content with curing or the combination of all these factors which could not be confirmed. The causes are inconclusive as sample moisture content during testing or the effect of thixotropic properties is not mentioned. The UCS results for SP-soils are found least affected by clay content showing similar improvement in UCS values (+475.85%) with enzyme doses (200 ml / 2 m³) and curing durations (2 weeks).
- For UCS and CBR, the performance of CH-soil for air dry curing (UCS = +942.9%, CBR unsoaked = +480%, soaked = +400%) is found better than desiccator curing (UCS = +244.9, CBR = NS). Similarly, improved performance for UCS values is observed for CI-soil with air dry curing (UCS = +827.78%) compared to desiccator curing (UCS = +158.94%).
- For the efficacy of enzymatic reaction, some initial moisture content should be available during the stabilization process however, the underperformance of desiccator-cured samples compared to air-dried specimens cannot be justified only by curing conditions. Further study on the effect of curing conditions on enzymatic reaction is required.
- The clay and silt-enriched soils have witnessed significant betterment in unsoaked and soaked CBR. However,

when compared among CL (Average % clay content = 39.40%) and CL-ML (59%) soils, CL soil having less clay content shows a greater percentage improvement in both Un-soaked CBR values (CL = +218.18, 4 weeks, CL-ML = 185.32, 4 weeks) and soaked CBR (CL = 333.33, 4 weeks, CL-ML = NS) values.

- For CH soils, higher clay content (Average % clay content = 36.82%) shows better performance for both unsoaked and soaked CBR however, for UCS performance the higher clay size fraction affects adversely. A similar reduction in UCS values with higher clay content was also found for CI-soils (Average % clay content = 32.25%).
- For CH-soils (Activity number A = 1.4) more enzyme doses (CH-soil 3 200 ml / 0.25 m³) are needed for the maximum CBR value (+480%). The MH-soils (A = 0.49) attained the maximum value (423.08%) at a slightly lower dose (200 ml / 2 m³). The ML and SM-GM soils with lower average clay/fine content (ML 15.65%, SM-GM 2%) attend the highest average CBR at moderate enzyme dose (un-soaked CBR ML-soil +451.79% for 200 ml / 2 m³, SM-GM +210.00% for 200 ml / 2 m³ of soil). Normally curing period reaches the maximum average percentage, and CBR decreases with clay/fine contents (CH = +480%, 3 weeks, MH = 423.08%, 3 weeks, ML = 451.79, 2 weeks).
- In general, for MH-soils, the enzyme stabilization effect on consistency indices is inconclusive as the result shows an inconsistent variation in values with enzyme doses and duration. The ML-soils with the average PI (3.79%) and least average clay/fine content (15.65%) compared to other soil groups records higher betterment in optimum moisture content at a lower enzyme dose of 200 ml / 3 m³.
- ML-soil with low clay/fine content and plasticity index witnessed higher improvement in optimum moisture

content (-41.18%, 1 week) at a lesser enzyme dose (200 ml / 3 m³). Thus clay/fine content and PI are prominent factors controlling the optimum dose. Similar results were recorded for MH-soils (A = 0.49) reaching maximum average % CBR (+423.08%) at a relatively lesser dose (200 ml / 2 m³).

- Normally curing period of 4 weeks for CL-ML soils and CI soils, 3 weeks for MH soils, and 2 weeks for ML soils is needed to attain the maximum average%, CBR. Thus, indicating the need for a lower curing period with a decrease in clay/fine contents. The soaked CBR requires a greater curing period of 3 weeks to 8 weeks to attain maximum CBR value. The higher duration may be due to the leaching of the enzyme during soaked CBR.

6. Conclusion

The CI-soils are found comparatively more suitable TerraZyme stabilization. In general, the improvement in soil properties depends on average clay content, particle size gradation, type and duration of curing. However, these parameters affect the individual test results for various soils differently as witnessed in the result discussion. There is substantial variation in optimum enzyme dose and curing period requirements for different soils based on the parameters being analyzed. The maximum individual parametric improvements for various soil types are at different enzyme doses. Hence optimal enzyme dose cannot be fixed. However, in general, optimized enzyme doses for various soil types are CH-soils 200 ml / 1.5-2 m³, CI-soils and CL-soils 200 ml / 6.6 m³, CL-ML soils 200 ml / 0.5 m³, MH-soils 200 ml / 2-2.5 m³, ML-soils, SP-soils and SM-GM soils 200 ml / 2 m³. Similar variation is observed in the curing duration required to attain the maximum parametric variation hence it is necessary to ensure enough curing duration before use.

The enzyme stabilizer evaluation studies

have yielded unlike performances witnessing inconsistent improvement in properties even among the soils belonging to the same group. One of the causes for such an unforeseen result may be due to deviation from the standard test procedures. The limited specimens tested, under the non-uniform procedure and test conditions can also contribute to such unexpected results.

There are not any parametric ranges of improvements in index and engineering properties of stabilized soils specified and standardized for various engineering applications.

There is a need for specific laboratory tests for the assessment of the field performance of enzyme stabilizers. The enzyme stabilizers without significant improvement under controlled laboratory conditions are less likely to attain desired performance in less favourable field conditions. Hence both laboratory and field tests are recommended before large-scale application. The research emphasizes the necessity of an elaborate organized study on the enzyme and soil characterization, their suitability, and optimization of doses, curing type and duration to have a better knowhow of the effectiveness of enzymatic stabilization.

6. Declarations

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6.2. Conflict of Interest

The authors declared that they have no conflicts of interest in this work. We declare that we do not have any commercial or associative interest that represents a conflict of interest in connection with the work submitted.

6.3. Availability of Data and Material

Not applicable

6.4. Code Availability

Not applicable

6.5. Author Contributions

The corresponding author claims the major contribution of the paper including formulation, analysis and editing. The co-author provides guidance to verify the analysis result and manuscript editing.

6.6. Compliance with Ethical Standards

This article is a completely original work of its authors. It has not been published before and will not be sent to other publications until the journal's editorial board decides not to accept it for publication.

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6. Appendix

Details and characterization of the soil types used in the study.

No.	References	IS Soil Classification	Clay size (%)	Soil Description	Soil Plasticity	Activity	FSI (%)	Soil nature based on FSI (North Dakota DOT, 2014)	
1	Ramesh and Sagar (2015)	CH-Soil1	21.6	Clay of high plasticity	Highly Plastic	Active	NS	-	
2	Sweta and Maheswar (2017)	CH-Soil2	42	Clay of high plasticity	Highly Plastic	Normal	100	Montmorilloniti, Swelling, Very high expansivity	
3	Patel Usha et al. (2018)	CH-Soil3	30	Clay of high plasticity	Highly Plastic	Normal	80	Montmorilloniti, Swelling, Very high expansivity	
4	Vinay Kumar et al. (2020)	CH-Soil4	66.8	Clay of high plasticity	Highly Plastic	Inactive	NS	-	
5.1		CH	CH-Soil5	33	Lean clay	Highly Plastic	Normal	170	Montmorilloniti, Swelling, Very high expansivity
5.2	Myint et al. (2014)	CH-Soil6	20.2	Fat clay with gravel	Highly Plastic	Active	163	Montmorilloniti, Swelling, Very high expansivity	
5.3		CH-Soil7	12.3	Sandy fat clay	Highly Plastic	Active	150	Montmorilloniti, Swelling, Very high expansivity	
6	Marasteanu et al. (2005)	CH-Soil8	30	Clay of high plasticity	Highly Plastic	Active	78	Montmorilloniti, Swelling, Very high expansivity	
7	Joydeep et al. (2015)	CH-Soil9	68.7	Clay of high plasticity	Highly Plastic	Active	72.8	Montmorilloniti, Swelling, Very high expansivity	
8	Ramesh and Sagar (2015)	CI-Soil1	18.2	Intermediate plasticity	Highly Plastic	Normal	NS	-	
9	Dhanesh and Mohandas (2016)	CI	CI-Soil2	53	Intermediate plasticity	Highly Plastic	Inactive	NS	-
10	Shah and Shah (2016)	CI-Soil3	25.5	Intermediate plasticity	Highly Plastic	Normal	88	Montmorilloniti, Swelling, Very high expansivity	
11	Nandini et al. (2015)	CL-Soil1	78	Clay of low plasticity	Highly Plastic	Inactive	NS	-	
12	Myint et al. (2014)	CL	CL-Soil2	7.1	Sandy lean clay	Moderately Plastic	Active	170	Montmorillonitic, Swelling, Very high expansivity
13	Sodhi and Ocean (2018)	CL-ML	CL-ML	59	Silty clay	Slightly Plastic	Inactive	NS	-
14	Nandini et al. (2019)	MH-Soil1	53	Silt of high plasticity	Highly Plastic	Inactive	NS	-	
15.1	Kumar and Kumar Khan (2018)	MH-Soil2	40	Silt of high plasticity	Highly Plastic	Inactive	55	-	
15.2		MH-Soil3	39.4	Silt of high plasticity	Highly Plastic	Inactive	48	-	
16.1		MH-Soil4	78	Silt of high plasticity	Highly Plastic	Inactive	75	-	
16.2	Priyanka et al. (2016)	MH	MH-Soil5	84	Silt with high plasticity	Highly Plastic	81.82	Montmorillonitic Swelling, Very high expansivity	
16.3		MH-Soil6	92	Silt of high plasticity	Highly Plastic	Inactive	90.5	-	
17	Farooq and Sukhdeep (2020)	MH-Soil7	20.4	Silt of high plasticity	Moderately Plastic	Normal	NS	-	
18	Daigavane and Ansari (2021)	MH-Soil8	68.7	Silt of high plasticity	Highly Plastic	Inactive	NS	-	
19	Jenith and Parthiban (2017)	MH-Soil9	20.2	Silt of high plasticity	Highly Plastic	Normal	NS	-	
20	Myint et al. (2014)	MH-Soil10	53.8	Elastic silt	Highly Plastic	Inactive	93	Montmorillonitic, Swelling, Very high expansivity	
21	Venika et al. (2015)	ML	ML-Soil	15.7	Silt of low plasticity	Slightly Plastic	Inactive	NS	-
22	Muguda and Nagaraj (2019)	SC	SC-Soil1	21	Clayey sand	Highly Plastic	Normal	120	Montmorillonitic, Swelling, Very high expansivity
23	Priyanka et al. (2016)	SC-Soil2	00	Clayey sand	Slightly Plastic	Inactive	25	-	
24.1		SP-SC	SP-SC	8	Poorly graded sand with clay	Slightly Plastic	Normal	NS	-
24.2	Venkatasubramanian and Dhinakaran (2011)	SM-Soil1	20	Silty sand	Slightly Plastic	Inactive	NS	-	
24.3		SM-Soil2	12.5	Silty sand	Slightly Plastic	Inactive	NS	-	
25	Shankar et al. (2009)	SM-GM	SM-GM	2	Silty sand	Moderate Plastic	Active	NS	-
26.1	Nandini et al., (2020a)	SP-Soil1	1	Poorly graded sand	Moderate Plastic	Active	NS	-	
26.2	Nandini et al. (2020b)	SP-Soil2	2.45	Poorly graded sand	Highly Plastic	Active	NS	-	