

PERFORMANCE PARAMETERS FOR BIO-ENZYMES SOIL STABILIZATION

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Abstract

Literature published case studies are analyzed rationally to conclude the performance parameters of bio-enzymes for soil stabilization. The various aspects of enzyme stabilization such as stabilization mechanism, soil types suitable for enzyme stabilization, enzyme application rates and stabilization durability are evaluated. The improvements in index and engineering properties of enzyme-stabilized soil are used as a measure of the degree of stabilization achieved. The parameters affecting the results of laboratory tests on stabilized soils and also the changes observed in the index and engineering properties of enzyme-stabilized soils are worked out for the study. The study highlights that, though there are claims by the manufacturer and researchers, bio-enzymes catalyze very specific chemical reactions under conducive conditions thus bio-enzymes have very soil specific performance only. The performance of the enzyme stabilization cannot be trusted without independent testing. There are no particular laboratory tests for anticipating the field efficacy of Enzyme stabilizers. The actual field tests for intended performance under actual working conditions are recommended for the proposed/selected enzyme and available soil type before wide-scale stabilization on site. It is concluded that further study is required to establish the performance of bio-enzyme stabilization with various soil types. The appropriate performance evaluation laboratory tests for enzyme stabilizers need to be developed.

Keywords: Bio-enzyme, stabilization mechanism, affecting parameters, soil properties.

1. INTRODUCTION

In the stabilization process, the soil is treated chemically or mechanically to improve its desired engineering properties. Enzyme treatment is the non-traditional soil stabilization method.[1] The enzymes are proprietary, concentrated, nonbacterial, biodegradable, preparations. They are supposed to minimize compaction efforts, increase the soil density, and shear strength and also lower the soil permeability [2]. Enzymes act as the catalysts of biological systems that control the reactions rate and reduce the activation energy required for the development of the new product.[3] The enzymes are more efficient compared to inorganic catalysts. They 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. The enzyme stabilization is affected by the soil pH. The enzymes performance is efficient around a pH of 7 [4].

The major advantages of the enzymes compared to other conventional stabilizing additives are cost-effectiveness, environmental sustainability and their convenience in 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.

2. 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 is chosen. The variation in a characteristic of soils needs systematic study to determine the enzyme's stabilization performance. Presently the enzyme stabilization is mostly based on empirical guidelines from past experiences and not much subjected to technical evolutions.[2] The conducive conditions and exact stabilizing mechanism of enzymes are still uncertain. The experimental and field studies conducted for evaluation of the enzyme stabilization have shown unlike results. Therefore, it is attempted to study the various aspects of enzyme stabilization such as stabilization mechanism, favourable conditions for stabilization, parameters affecting the results of laboratory studies and also the measures of the degree of stabilization induced by the enzyme. These important facets of enzyme stabilization from the literature are analyzed for this study.

A detailed study of published literature was carried out to represent various aspects of enzyme stabilization. The various aspects of enzyme stabilization from case studies are discussed. The performance evaluation of the stabilized soils marked by the improvements in index and engineering properties are also discussed and elaborated. The following sections highlight and conclude the various aspects of enzyme stabilization such as the stabilization mechanisms, enzyme application rates, soil suitability for stabilization, index and engineering properties of enzyme-stabilized soils, stabilization durability etc.

3. ENZYME STABILIZATION MECHANISMS

According to researchers the mechanisms of enzyme stabilization is, with a certain level of ambiguity as the enzymes may act in several manners due to their formulations. According to Scholen D.E. [3] and Petry, T. M. [5] The pH levels, protein content, enzyme activity with soil constituent and surfactant characteristics of the enzyme products may lead to soil stabilization. According to Scholen enzyme products stabilizes the soils through various mechanisms, like clay mineral encapsulation, break up of clay minerals with the exclusion of the electric double layer water, and subsequent interlayer expansion due to the entrapment of moisture or exchange of interlayer cations. [6]

Following are the different philosophies involved in the enzyme stabilization process,

- According to Scholen [6], the enzymes react with organic molecules and surround the clay minerals, counterbalancing negatively charged clay particles and thus reducing their affinity for water.
- Velasquez, R.A. [7] and Tingle, J.S. [8] confirm that the enzymatic reaction produces organic molecules to act as a reactant intermediary/mediator, neutralizing the clay negative charge. These exchanges ions with clay, thus breaking its lattice structure.
- The enzymes stabilize the soil by cation exchange process, the clay-water effect, and interaction with the electrical double layer thus effectively dissipating the pore water.[9] The soil-stabilizing enzymes are similar to protein compounds and act as catalysts in chemical reactions with soil bearing organic content. [10]
- During the enzymatic reaction, the specific metabolite fitting on the enzyme surface is transformed into a substrate. The enzyme catalytic efficiency is measured as the number of substrate moles produced per unit time. The polypeptide chain folding of enzyme act as an active site for the substrate-binding during catalysis. This substrate is kept in place for other molecules to react in the correct orientation [4]. Both, catalytic behaviour and the enzyme-soil interaction bring down the affinity of clay for water.
- Surfactants in the enzymatic compounds reduce the surface tension and enable closer packing of the particles. The increased wetting capacity of treated soil allows the soil particles to get more densely packed. Chemical bonding due to enzymatic reaction fuses the soil particles, forming a permanent soil structure capable of withstanding weathering, wear and water penetration. [11].
- According to Tolleson, [12], the enzymes provide a surface for the microbe's adsorption. The microbes in closer proximity bond together through covalent bonds with the reduction in surface tension of soil water. The soil particles agglomerate, get cemented, thus, reducing the voids and resulting in the formation of a dense stratum.
- According to some thoughts [6] [3], the enzyme stabilization is by expedited shale formation process. Enzymes cover the clay particles with organic molecules at a higher reaction rate. The accompanied compaction results in the formation of the enzyme-stabilized soil matrix within a short time. This process reduces the tendency of the clays to absorb water. 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 required for gel crystallization. The fine organic matter and clay-sized particles are essential for stabilization and bond formation.

4. IMPORTANT FACTORS AFFECTING ENZYME STABILIZATION

Laboratory tests for mapping improvements in index and engineering properties of stabilized soil are recommended before field applications; however, the variation of test results observed in the literature also necessitates considering the factors affecting the results. The factors that affect laboratory results are the type of enzyme, enzyme application rates, soil suitability sample preparation and test procedure, sample curing conditions etc.

4.1. Type of Enzyme

A study by Tanveer Ahmed Khan shows the degree of effectiveness of stabilization is different for various enzymes. Therefore, it is required to ensure enzyme suitability earlier for larger-scale applications. [13]. Also, it's a better idea to check and standardize the enzyme constituents to assured its consistent performance.

4.2. Enzyme application rates

Enzymes are applied in low concentrations as they are not depleted by the soil reactions. The pore fluid in the soil offers mobility to the bio-enzyme molecules for diffusion into the soil mass with time. Thus, leading to bio-enzyme reaction with the particular soil chemistry. This enzymatic action would continue till there are no further reactions to catalyze. Hence enzymes are needed in small doses. The stabilizing reaction of an enzyme is considered likely to be very soil specific [14]. However, the application rate or enzyme doses for the required performance may depend on the specific soil characteristics, such as the clay content and the constituent clay mineral. The enzymatic reaction would be very soil specific. If suitable to the soil type and used in an appropriate

dose, enzymes can add to the structural integrity of the soil matrix. Hence, before the application of these enzymatic products, an appropriate enzyme dose should be determined.[15]

Bio enzyme stabilizers are supplied as concentrated liquids, with the dilution range around 1:150 to 1:300 (stabilizer: water ratio). The guidelines about application rates given by the manufacturers are related to the upper and lower limit only. Dosages recommended by the manufacturer can be 200 ml for a bulk volume of 3.5 m³ to 2 m³ of soil even up to 1 Liter per 25 m³ based on soil properties. The literature records several case studies aimed at investigating the optimum enzyme doses. The process also recorded the variation in properties of treated stabilized soil with changes in enzyme doses. However, results defining the performance of enzymes for soil stabilization vary greatly in the literature. The experimental results on soil samples treated with different doses of enzymes showed no differences, indicating that both the low and high dosages of the enzyme are ineffective [7]. On the contrary, literature also described that the varying enzyme dosages have shown a different degree of stabilization on the same soil. The insufficient enzyme quantity would not result in required stabilization, whereas higher enzyme doses may adversely affect the stabilization.[16]

4.3. Soil Types Suitable for Enzyme Stabilization

Another perception is that soil type and specifically the clay content in the soil greatly affects enzyme performance. The percentage of clay size contents, the chemical and mineral constituents of soil affect the stabilization mechanism. Hence precaution should be practised in the selection of enzyme treatment for specific soils. For an effective enzymatic reaction, the soil must have the necessary clay contents. Rauch et al. reported that for effective bio enzyme stabilization, the soil should have clay minerals for reaction with other chemical constituents. Enzymes are suitable for the treatment of soils with high plasticity clays having an affinity for water and have some organic content for the enzymatic reaction. [17] The researchers suggested that in several field applications that enzyme may have performed poorly due to the deficiency of requisite clay content in the treated soil.[18] For bio-enzyme stabilization, the soil needs to have some clay content. As per Bergmann, a minimum of 2% clay is needed for enzyme stabilization. The clay content of 10% to 15% delivers effective stabilization. [19]. According to some manufacturers, enzyme products are most efficient for the treatment of soils containing 18% to 30% clay content/cohesive fines and soils having a maximum liquid limit (WL) of 30% and a plasticity index (PI) between 2% and 10%.[10] For the soil to be stabilized, the minimum clay content has to be at least 10% and with a PI of more than 8%. Maximum stabilization results are obtained for soils having 12% to 24% clay content (even up to 30% for some enzymes), with a plasticity index (PI) between 8 and 35%. Also, enzymes give the best stabilizing performance at moisture content on the 2-3% drier side of optimum moisture content (OMC).[20]

4.4. Laboratory Sample Preparation and Tests

Proper sample preparation is a must for standardizing the findings of laboratory investigations. The literature laboratory studies barely discuss the precaution taken to control the affecting parameters and standards followed during the testing. Though it is understood that laboratory experimentation complies with standard procedure, small factors such as sample preparations, moisture content, curing conditions, temperature control, enzyme doses, etc. induced large variation in results either way. The soil experiences irreversible structural changes at greater heating temperatures (in general greater than 40°C) modifying its physical and mechanical behaviour. If required a suitable drying temperature needs to be selected during sample preparation.

According to Rauch, Katz [15] enzyme-treated samples performed unsatisfactorily due to the faulty sample preparation methods. Also, the inefficiency of stabilization can be due to defective sample preparation, improper and insufficient curing, and selection of unsuitable soil types for stabilization. Proper soil moisture content during stabilization should be ensured considering the moisture provided by enzymatic emulsion itself. The suppliers specify the recommended enzyme application rates using various terminology (units). The recommended doses are very low, hence for homogeneous mixing, the enzymes can be diluted in water for their application. Rauch et al. [17] developed a revised protocol for soil test specimen preparation, counting the change in compaction characteristics introduced by the enzyme stabilizers. The enzyme-treated soil samples were prepared at the OMC obtained for the controlled specimen of untreated/virgin soil. It is recommended to follow the Dilution mass ratio (DMR) i.e. the stabilizer dilution before application. Once the test soil is pre-moistened with initial moisture content (IWC) the calculated quantity of diluted stabilizer is added to attain the required Application mass ratio (AMR) for sample preparation. [17]. During the process, the sample needs to mature for 16 hours in a sealed container for uniform moisture absorption. The final moisture content of the treated sample must be obtained within the allowable range of OMC. The thoroughly mixed soil sample is then mellow for 1 hour in a sealed container and then compacted in a specified manner, extruded stored and cured in a sealed container at room temperature for a duration of 07 days. The sample is then trimmed to the appropriate test size. If the determined moisture content is not within the required limits for compaction, new specimens need to be prepared using an adjusted IWC [15]. The

moisture content of samples should be determined at both times of compaction and testing, the variation should be within the minimum permissible limit (say 1% of targeted water content.) [17].

Untreated control specimens of virgin soil without the enzyme stabilizer should also be prepared in the same manner. During specimen preparation, doses of stabilizer, moisture control and curing conditions will have a pronounced effect on soil properties. However, only a few authors have categorically mentioned the standards followed during sample preparation and testing procedure. It's hard to ensure compliance with the rational protocol for specimens' preparation and standards of laboratory conduction based on literature-based data.

4.5. Curing conditions

As per the manufacturers, the enzyme stabilization needs of 03 to 07 days period of curing after treatment with soil. The literature shows samples curing for the various duration up to 60 days under controlled conditions to check the effectivity of enzymatic reaction with time. The curing may be; air-dry at room temperature or in a sealed container for preserving moisture content over the curing duration.

As per Peng, the enzyme was more effective in 60 days of air-dry curing for fine-grained soil (Soil-I) and silty loam (Soil- II) compared to ground quicklime. However, the enzyme stabilization was ineffective for coarse-grained Soil-III in air-dry curing as well as in sealed curing for these soils. Peng et al. also conclude that soil samples cured in a sealed condition depict no improvement while soil samples in an air-dry cured condition show a maximum of 10% gain in strength.[21]

Ramesh H. & Sagar S. also conclude that air dry curing is better than desiccator curing.[22] However, whether the gain in strength is because of enzyme treatment or simply because of loss of moisture due to drying is not confirmed in most of the study cases. According to Muguda S [23], sealed curing of the enzymes treated soil was more effective in gaining strength than wet curing, therefore selection of a suitable curing method close to the actual field conditions is recommended.

For Unconfined Compression Strength (UCS) and California bearing ratio (CBR), performance air dry curing is found better than desiccator curing. A similar better performance for UCS values is observed for CI-soil with air dry curing. CH-Soil1 gives a maximum percentage increase in UCS of 942.86 % for an enzyme dose of 200ml/1.5m³ of soil at the air-dry curing of 8 weeks. Whereas for the same soil, the desiccator cured specimen gives the maximum increase of 244.90% for the same enzyme dose and curing duration. CBR-CH-Soil1 air dry cured sample shows CBR value increase of 400% as compared to 266.67% of increase for desiccator cured sample with the same enzyme dose of 200ml/2m³ of soil at the end of 4th week. For numerous literature case studies, the distinction between curing type was not specifically mentioned making it difficult to conclude with the effect of the curing method on soil properties. There is a need for initial moisture content for initiation and effectiveness of enzymatic action, however, lower values of desiccator cured laboratory specimen- compared to air dry curing cannot be explained based on curing conditions only [24].

5. LABORATORY TEST ON ENZYME-TREATED SOILS

Bio-Enzyme stabilization of expansive soil has shown a greater improvement in the engineering properties attributed to the higher reactivity of soil constituents with the Bio-Enzyme. For ensuring the expected field stabilization results, it is highly advocated to check the laboratory performance of bio-enzyme stabilization.[25]

In general, the performance of stabilized soil in the index and engineering properties test is considered an evaluation parameter for the effectiveness of the degree of stabilization achieved. Consistency test, Proctor Test, UCS, Soaked/Unsoaked CBR tests, Free Swell Test, Swell Pressure tests, consolidation test, permeability test are also can be used for verifying the performance of stabilized soils. The dynamic cone penetration test (DCP), results on stabilized soils can also be correlated with the field engineering properties of the soil. However, there are not any parametric ranges of these improvements specified and standardized for various engineering applications. There are several mixed examples from literature where laboratory testing shows highly beneficial or no substantial improvement in properties of enzyme stabilized soils.

5.1. Atterberg Limits

The index properties such as liquid limit, and the plastic limit is related to various properties of soil such as cohesion, capillarity and also form the basis for soil classification and its specification as embankment material. Atterberg Limits Test for untreated and treated soil samples with various enzyme doses and curing periods have shown no substantial improvement. The plasticity indices for two doses showed a marginal change from the untreated samples. However, the variation in plasticity indices (PI) may be due to laboratory inconsistencies [13]. Rauch et al. [26] also reported no reduction in PI for the tested five different soils.

Literature studied all CH, CI, MH, ML and SM-GM soils shows reduction LL, PL, PI (increase in shrinkage limit) with enzyme treatment. The CI-Soils witness the maximum average percentage reduction of 68.82% in liquid limit (LL) (200ml/0.6m³, 3 weeks), reduction of 50% in Plastic Limit (PL) (200ml/0.6m³, 3 weeks), reduction of 59.26% in Plasticity Index (PI) (200ml/6.6m³, 4 weeks), increase of 25.38% in Shrinkage Limit (Ws)

(200ml/1.5m³, 0 weeks). Though the most prominent improvement in all consistency limits was shown by CI-Soils, however, to achieve these improvements there is significant variation in optimum enzyme doses and duration of curing required. Lesser average clay content (and thus lesser PI) of CI soils may be the reason for this improvement. The increase in shrinkage limit for treated Soil shows a higher degree of volume stability after treatment [24].

5.2. Compaction Characteristics

A compaction 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 achieved at OMC. The water content and the density relation from the test can be used for field compaction control. Rauch et al. [26] investigated the effect of enzyme stabilization on compaction characteristics using the OMC of untreated soils. However, observed no improvement in the maximum dry density (MDD) for montmorillonite, illite kaolinite, and the other two native Texas clays. It is recommended that OMC and MDD for enzyme stabilized soils should be determined separately [26]. It was also observed that enzyme stabilization results in a higher increase in the compaction on the dry side of optimum moisture content, giving more compaction for lower water contents.

A decrease in OMC may be because of the effective cation exchange process due to enzymatic reaction. The most prominent variation in compaction characteristics was shown by CI-Soils, thus soils with intermediate clay contents show the best improvement in MDD value. The maximum average percentage reduction in OMC of 68.71% (200ml/0.6m³, 6 weeks) and increase of 68.71% (200ml/0.6m³, 6 weeks) in MMD was witness by CI-Soils. CH-Soils require higher enzyme doses to achieve the maximum average percentage MMD value. However, there is significant variation in optimum enzyme doses and curing duration required to acquire the maximum average percentage MMD for different soils [24].

5.3. Unconfined compressive strength (UCS)

UCS test is used for determining the compressive and shearing strength of undrained clayey soils without confining pressure. It is evident from the literature that the increase in strength was moderate and applicable to individual cases, without any persistent consistency trend. UCS results even for the long 84-day curing did not show much significant improvement [13]. The experimental results of UCS on soil samples with different doses of enzymes have shown no differences [7].

The controlled untreated soil samples also need to be tested to check the strength gain due to thixotropic characteristics or ageing of soil. Part of this significant improvement could be due to the loss of moisture. The moisture content at the sample preparation and testing at the end of curing duration was not specified for several studies.

The clayey soils (CH, CI, CL-ML) have shown maximum improvement in UCS values with ageing. CH (increase of 493.43% 200ml/2m³, 8 weeks), CI soils (increase of 493.36%, 200ml/2.5m³, 8 weeks) give the highest improvements in UCS values over almost similar enzyme doses and curing duration are with 36.82% and 32.25% clay size particles respectively. For CH, MH and SP-Soils reach maximum average percentage UCS value at moderate enzyme dose of 200ml/2m³ however, clayey soil requires a higher curing duration to reach maximum value. As clay contents go on decreasing time required to attend maximum value also decreases. The CL-ML Soils requires a higher enzyme dose of 200ml/0.5m³ and moderate duration to attend the maximum average percentage UCS value [24].

5.4. California Bearing Ratio (CBR) test

CBR value measures the soil strength based on the condition of the material during the testing. The CBR value can be correlated with plasticity index, modulus of subgrade reaction and modulus of resilience. As per Isaac et al. (2003), TerraZyme stabilized lateritic soil and clayey soil cured for a duration of 8 weeks have reported a 136 to 1800 times increase in CBR value over untreated soil [27]. Sedimentary residual soil was treated and cured for four months with the recommended dose and also with 2, 5 and 10 times the recommended enzyme dose observed no improvement for both soaked and unsoaked CBR values. T. A. Khan [28] also reported no significant improvement in, Atterberg/consistency limits, compaction characteristics and UCS of the soil treated with the three enzymes. Some improvement, for a few individual cases, could be explained by the speculation that the enzymes did not produce any significant chemical change leading to stabilization, on the contrary, they only prevented moisture absorption to bring the particles to a closer state [13]. Overall, the laboratory results of enzyme stabilization are quite variable and inconclusive.

Both clayey and silty soils have shown maximum improvement in both soaked and Unsoaked CBR values. CH-Soils witnessed maximum average % increase in unsoaked CBR 480% (200ml/0.25m³, 3 weeks) and 329.85% in soaked CBR (200ml/0.75m³, 8 weeks) where as for MH-Soils maximum average % increase in unsoaked CBR 435% (200ml/6.6m³, 4 weeks and 380% in soaked CBR (200ml/6.6m³, 4 weeks). For CH-Soils (Activity no.=1.4) higher doses are required to reach the maximum average percentage CBR value whereas for MH-Soils (Activity

no.=0.49) reaches to maximum average percentage CBR value at comparatively smaller enzyme doses. Whereas ML, SM-GM soils with lesser clay content (15.65% and 2% respectively) reach to highest average performance at moderate enzyme dose. In general curing duration required to reach the maximum average percentage, CBR value decreases with clay contents [24].

6. MICROSCOPIC CHARACTERIZATION OF TREATED SOILS.

Bio-Enzyme stabilization of expansive soil has shown a high improvement in the engineering properties. This improvement may be due to the chemical reaction of soil constituents with Bio-Enzymes [25]. Though the laboratory tests results are with variable trends the stabilization of soil is brought about by chemical reaction with soil constituents. One of the advanced parameters to check the degree of chemical reaction of the enzyme with soil is to check changes in microscopic characters of stabilized soils. Zhang et al. [29] and Khan and Taha [13] have discussed the changes in the microstructure of untreated and enzyme-treated soils with curing. Enzyme stabilization leads to denser agglomeration of particles compared to untreated soil. There are several examples in literature discussing the microstructural changes observed with Enzyme stabilization.

- As per BET surface area analysis, the enzyme stabilizer results in a significant agglomeration of the soil particles irrespective of the type of the soil. Environmental scanning electron microscopy (ESEM) images also confirmed the greater aggregated nature and less clayey features of enzyme-treated soils compared to corresponding untreated material.
- X-ray diffraction (XRD) and X-ray fluorescence (XRF) of treated and untreated soils after two months of curing did not reveal any appreciable changes in chemical composition and mineralogy. Thus, indicating no evidence of any chemical reaction for the treated soil. However, the field emission scanning electron microscopy (FESEM) images display a denser close packing of soil particles treated with some bio enzymes [13].
- Rauch et al. [15] confirm minor changes in X-ray diffraction results, specific surface area, and alumina/silica ratios for high enzyme doses of 50% by dry weight of soil.
- The scanning electron microscopy (SEM) micrographs show the microstructure of the TerraZyme stabilized soil is denser than that of untreated soil as the particles of stabilized/ treated soil samples are coarser and blockier than those untreated with TerraZyme. The TerraZyme treated soil show lesser pores compared to untreated soil. This compact microstructure should be the basis of the higher UCS value of TerraZyme stabilized soil [29].

An Analytical X-ray diffraction (XRD), test, was carried out to explore chemical changes, due to enzyme treatment. The XRD diffractogram patterns for treated samples are consistent with the respective untreated patterns. No chemical changes occurred leading to changes in the chemical composition of the enzyme-treated soils. FESEM images of treated samples with a certain enzyme dosage are more accumulated than the untreated sample and are with fewer observable clay features. However, no change in the material composition was reported [30].

The significant improvement in soil properties cannot be justified without a chemical reaction accompanied by alteration in the chemical composition of enzyme-treated soils. The studies reporting a high degree of enzyme stabilization are without any validation of the occurrence of any such chemical reactions. Thus, there are no conclusive evidence and the literature results are contradictory about the efficacy of enzyme stabilization. The micro-level observations seem to be too inconclusive to confirm the changes induced by enzymatic reactions. These changes are again variable with soil mineralogical contents.

7. THE DURABILITY OF ENZYME STABILIZATION.

Life expectancy and performance of enzyme stabilized soil can be ensured with optimum dosage, proper application conditions and adequate maintenance.[20] The durability of the stabilized road materials is also affected by traffic and climatic conditions. Usually, life expectancy is of 5 years to 7 years or even 12 or more years in exceptional applications cases.[6] Further grading and additional enzymatic emulsion may be needed for preserving the quality of stabilized layers [20]. However, for the stabilizers, additional studies are needed to assess the long-term effectiveness and permanence of the enzyme products [15].

The literature witnesses the improvements in enzyme stabilized soils, however, there are also several conditions where enzymatic stabilization has underperformed. Under serve field conditions the stabilized soil may likely to subjected to alternate dry and wet cycles and also to leaching out in the saturated condition. Although enzyme stabilization is effective for the compaction of clays at lesser moisture content. These stabilized clayey soils perform very poorly under alternate wet-dry cycles and also with leaching tests. The enzyme stabilized soils exhibited the nearly same strengths as that of untreated materials [31].

8. CONCLUDING REMARKS

Considering the study, the authors recommend precaution in enzyme application practice for soil stabilization. Both applicable laboratory tests accompanied by actual field testing are advocated before on-site large-scale enzymes stabilization.

- i Under conducive conditions only, enzymes react and catalyze only specific chemical reactions. Thus, it is hard to distinguish a general stabilization mechanism and the degree of its effectivity due to variations in the soil characteristics and reactions.
- ii Enzymes are suitable for stabilizing high plasticity, water affine soils with some organic content for the enzymatic reaction. The silty and granular soils with lesser clay contents would be unsuitable for enzyme stabilization.
- iii Enzyme stabilizers might only be effective to treat specific soils. Poorly graded soils should be modified by blending with the soil of suitable particle ranges. Although some manufacturers claim that materials containing no fines/no clay contents can be stabilized no such practice or evidence from case studies was observed.
- iv It is recommended to obtain truly representative soil samples from the field. The enzyme-treated soil specimens for laboratory testing should be prepared strictly using standard methods and as per the manufacturer guidelines. Any deviation from standard test procedures may also be led to an inconclusive result.
- v Though there are published studies and results, the stabilization performance of the enzyme cannot be ensured without unbiased laboratory and field testing. The enzymes which do not show substantially improved stabilization in the controlled laboratory conditions are less likely to attain desired performance in less favourable field conditions.
- vi There are no specific laboratory tests for predicting the field performance of enzyme stabilizers. Hence field tests are recommended for the selected enzyme and applicable soil type before wide-scale application on-site. It has been recommended that different types of soils and enzymes need to be tested to quantify the benefits and effectiveness in actual field behaviour. If the laboratory results indicate significant improvement for the treated soils, then only the enzyme product can be tested and used for field operations as per recommendations by the manufacturer.
- vii Though in general the laboratory tests are not recommended for performance evaluation of the enzyme stabilization as these standard tests do not reflect actual field conditions and therefore can give misleading results. Further research is required for developing appropriate laboratory tests.

REFERENCES.

- [1] Mekonnen E, Kebede A, Tafesse T and Tafesse M 2020 Application of microbial bio enzymes in soil stabilization International Journal of Microbiology, vol. 2020, pp. 1–8, DOI: 10.1155/2020/1725482
- [2] Taha Mohd Khan, Tanveer Jawad, Ibtehaj and Firoozi Ali. 2013 Recent experimental studies in soil stabilization with the bio-enzymes-a review. Electronic Journal of Geotechnical Engineering. 18. 3881-3894
- [3] Scholen D E 1995 Stabilizer mechanisms in nonstandard stabilizers Proc., 6th International Conference on Low-Volume Roads, Vol. 2, Minneapolis, Minn., TRB, National Research Council, Washington, D.C. pp. 252–260
- [4] Norris R, Ryan L and Acaster D 2011 Cambridge International AS and A Level Chemistry Coursebook. Cambridge University Press, UK, pp. 413-414
- [5] Petry T M 1997 Performance based testing of chemical stabilizers. In Transportation Research Record 1589, TRB, National Research Council, Washington, D.C. pp. 36–41
- [6] Scholen D E 1992 Non-Standard stabilizers. Report FHWA-FLP-92-011. FHWA, U.S. Department of Transportation, Washington D.C.
- [7] Velasquez R A , Marasteanu M and Hozalski R 2006 Investigation of the effectiveness and mechanisms of enzyme products for subgrade stabilization International Journal of Pavement Engineering, Vol. 7, No. 3, pp. 213–220
- [8] Tingle J S and Santoni R L 2003 Stabilization of clay soils with nontraditional additives In Transportation Research Record: Journal of the Transportation Research Board, No. 1819, Transportation Research Board of the National Academies, Washington, D.C. pp. 72–84 doi: 10.3141/1819b-10
- [9] How Terrazyme Works. Terrazyme, Inc. Accessed Dec 2021 <https://www.nature-plus.com/terrazyme-how-it-works>
- [10] North Dakota DOT 2014 Materials and research division experimental study ND 2014-01, Evaluation of Permazyme 11x™ Soil Stabilization Work Plan, SS-4-053(015)030
- [11] Marasteanu M O, Hozalski R, Clyne T R and Velasquez R 2005 Preliminary laboratory investigation of

- enzyme solutions as a soil stabilizer, MN/RC- 2005-25, Minnesota Department of Transportation, Research Services Section, St. Paul, Minnesota 55155
- [12] Tolleson A R , Shatnawi F M, Harman N E and Mahdavian E 2003 An evaluation of strength change on subgrade soils stabilized with anenzyme catalyst solution using CBR and SSG comparisons. Final report. South Carolina State University, Orangeburg
- [13] Khan T A and Taha M R 2015 Effect of three bioenzymes on compaction, consistency limits, and strength characteristics of a sedimentary residual soil *Advances in Materials Science and Engineering*, vol. 2015, pp 1-9, doi: 10.1155/2015/798965
- [14] Tingle J S et al 2007 Stabilization mechanisms of non-traditional stabilizers. In *Transportation Research Record: Journal of the Transportation Research Board*, No. 1989, Vol.2, Transportation Research Board of the National Academies, Washington, D.C., pp. 59-67
- [15] Rauch A F, Katz L E and Liljestrand H M 1993 An analysis of the mechanisms and efficacy of three liquid chemical soil stabilizers: Volume 1 Report No. 1993-1 FHWA/TX-03/1993-1 (Volume 1), Texas Department of Transportation Research and Technology Implementation
- [16] Shankar A R, Rai H K, Mithanthaya R I 2009 Bio-enzyme stabilised laterite soil as a highway material. *J Indian Roads Congr* 70:143–151
- [17] Rauch A F, Katz L E and Liljestrand H M 2003 An analysis of the mechanisms and efficacy of three liquid chemical soil stabilizers A report, Center for Transportation Research 'University of Texas, Austin, TX, USA
- [18] Woll J H, Surdahl R W, Everett R and Andersen R 2008 Road stabilizer product performance: seedskae national wildlife reserve FHWA/CFL/TD-08-005. Central Federal Lands Highway Division, FHWA, U.S. Department of Transportation, Lakewood, Colo.
- [19] Bergmann and Roger 2000 Soil stabilizers on universally accessible trails Technical Report 0023-1202-DTDC, San Dimas, Ca: U.S. Department of Agriculture, Forest Service, San Dimas Technology and Development Center, pp 10
- [20] Kestler M A 2009 Stabilization selection guide for aggregate and native surfaced low-volume roads U.S. Department of Agriculture, Forest Service, National Technology and Development Program
- [21] Peng H T, Su H T, Zhang X P and Wang J 2011 An experimental comparison of compressive strengths of soils stabilized with enzyme and ground quicklime *Advanced Materials Research* 280: pp 9-12
- [22] Ramesh H & Sagar S 2015 Effect of drying on the strength properties of TerraZyme treated expansive and non-expansive soils 50th Indian Geotechnical Conference
- [23] Muguda S and Nagaraj H B 2019 Effect of enzymes on plasticity and strength characteristics of an earthen construction material *Geo-Engineering*, vol. 10, no. 1, p. 2, doi: 10.1186/s40703-019-0098-2
- [24] Aswar D S, Bajad M N, Ambadkar S D 2022 Performance of TerraZyme for soil stabilization of various soil groups. *International Journal of Engineering Trends and Technology*, 70(4), 258-271. <https://doi.org/10.14445/22315381/IJETT-V70I4P222>
- [25] Guthrie W S, Simmons D O, and Eggett D L 2015 Enzyme stabilization of low-volume gravel roads *Transportation Research Record*, vol. 2511, no. 1, pp. 112–120, doi: 10.3141/2511-13
- [26] Rauch A F, Harmon J S, Katz L E, and H. M. Liljestrand H M 2002 Measured effects of liquid soil stabilizers on engineering properties of clay, *Transportation Research Record*, vol. 1787, no. 1, pp. 33–41, doi: 10.3141/1787-04
- [27] Isaac K P Biju and Veeraragavan 2003 Soil Stabilization Using bio-enzyme for rural roads presented at the IRC Seminar Integrated Development of Rural and Arterial Road Networks for Socio-Economic Development New Delhi
- [28] Tanveer Ahmed Khan, Mohd Raihan Taha et al. 2017 California Bearing Ratio tests of enzyme-treated sedimentary residual soil, *Sains Malaysiana* 46(8): 1259–1267, <http://dx.doi.org/10.17576/jsm-2017-4608-11>
- [29] Zhang X, Zhang X P, Peng H T, Xia Q and Wang J 2013 Relation of microstructure and unconfined compression strength of soil stabilized with TerraZyme *AMR*, vol. 664, pp. 760-763, , doi: 10.4028/www.scientific.net/AMR.664.760
- [30] Khan T A et al 2020 Strength and volume change characteristics of clayey soils: performance evaluation of enzymes *Minerals* vol. 10 no. 1 pp 52 doi: 10.3390/min10010052.
- [31] Parsons R L and Milburn J P 2003 Engineering behavior of stabilized soils *Transportation Research Record* vol. 1837 no. 1 pp. 20–29, doi: 10.3141/1837-03