

A mini project report on

A Review on Soil Stabilization using Microbial Bioenzymes



Submitted in Partial Fulfillment of the Requirement for the Award of the Degree of

MASTER OF TECHNOLOGY

In

CIVIL ENGINEERING

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ABSTRACT

Soil stabilization is a critical aspect of civil engineering and construction, aiming to enhance the engineering properties of soil to meet the demands of various infrastructure projects. Traditional stabilization methods often involve the use of chemical additives, which may pose environmental concerns and sustainability challenges. Currently, there is a growing interest to identify new and green technology to improve construction techniques and to expand the road network. Therefore, the search for new materials and improved techniques to process the local materials has received an increased focus. For developing countries, bioenzymes are now creating an opportunity to improve soil stability with tremendous effectiveness in the overall process of soil stabilization. This report reviews bio enzyme based soil stabilization techniques with an emphasis on bioenzymes production, types & mechanism of soil stabilization, future challenges, and opportunities of the sector. This report also discusses about the advantages and disadvantages of bio-enzymes. Soil stabilization using bioenzymes emerges as a sustainable solution, due to its low production cost, easily and widely applicable, and environmentally friendly enzymatic formulations from locally available raw materials, offering improved soil properties for construction while minimizing the negative impact on ecosystems. This research contributes to the growing body of knowledge on eco-friendly geotechnical engineering practices and encourages further exploration of bio-enzymes as a viable alternative in the realm of soil stabilization.

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CHAPTER 1

Introduction

1.1 Soil Stabilization

Soil stabilization is the permanent physical and chemical alteration of soils to enhance their physical properties. In its broadest sense, it includes compaction, pre consolidation, drainage, and many such processes. However, the term stabilization is generally restricted to the process which alters the soil material itself for improvement of its properties. It is the collective term for any physical, chemical, or biological method, or combination of such methods employed to improve certain properties of natural soil to make it serve for intended engineering purposes. Improvements include increasing the dry unit weight, bearing capabilities, volume changes, the performance of in situ subsoils, sands, and other waste materials in order to strengthen road surfaces, and other geotechnical applications. It is required when the soil available for construction is not suitable for the intended purpose and mainly aimed at increasing resistance to softening by water through bonding the soil particles together, water proofing the particles, or combination of the two.

Soil stabilization is a technique employed to enhance the engineering properties of soil, making it more durable and suitable for construction purposes. This process involves the application of various materials or methods to improve the soil's strength, durability, and load-bearing capacity. Common stabilizing agents include lime, cement, fly ash, and chemical additives, which are mixed with the soil to create a stable and compacted foundation. The goal is to mitigate issues such as soil erosion, settling, and subsidence, ultimately ensuring the integrity of structures built on the stabilized soil. Soil stabilization is crucial in civil engineering and construction projects, playing a vital role in the development of resilient and long-lasting infrastructures.

Soil stabilization holds paramount importance in the realm of civil engineering and construction for a multitude of reasons. One of the primary concerns addressed by soil stabilization is the enhancement of soil strength and load-bearing capacity. Unstable or weak soils can pose significant challenges during construction, leading to issues such as settling, subsidence, and structural failures. By stabilizing the soil, engineers can ensure a solid foundation that can withstand the pressures exerted by structures, ultimately enhancing the longevity and safety of buildings and infrastructure.

Soil stabilization is generally costly and requires huge investments. In some cases, construction has been hindered due to the high cost of soil stabilization techniques and depletion of stabilizing materials. The development of cost-effective materials and processes has been a crucial part of any construction for years. Hence, cost-effective road construction techniques are vital for economical growth in any country. As a result, there is an urgent need to identify new and cost-effective materials to improve construction techniques and expand road networks. Recently, the search for new materials and improved techniques for processing the local materials has received an increased interest. However, more attention has recently been given to the use of bioenzymes as soil stabilizers.

CHAPTER 2

Literature Review

2.1 Review of past studies on Bio-enzymatic Stabilization

Miburn & parsons (2003) A series of tests were conducted on soils which are stabilized with cement, lime, terrazyme and class c fly ash. Durability testing procedures included freeze–thaw, wet–dry, and leach testing. Atterberg limits and strength tests also were conducted before and after selected durability tests. Lime- and cement-stabilized soils showed the most improvement in soil performance for multiple soils, with fly ash-treated soils showing substantial improvement. The results showed that for many soils, more than one stabilization option may be effective for the construction of durable subgrades. The enzymatic stabilizer did not perform as well as the other stabilization alternatives.

Shukla et al. (2003) Made experiments on an expansive soil treated with an organic, non-toxic, ecofriendly bioenzyme stabilizer in order to assess its suitability in reducing the swelling in expansive soils. The experimental results indicate that the bioenzyme stabilizer used in the present investigation is effective and the swelling of an expansive soil reduces on wet side of OMC.

Marasteanu et al. (2005) They conducted tri-axial tests on two soils which were stabilized with two different enzymes. Soil-I has 96% of fines (75% of clay) a specific gravity of 2.73 and plasticity index of 52%. Soil-II has 60% of fines (14.5% of clay) and plasticity index of 9.4%. Tri-axial shear tests were carried out on two confining pressures (4psi and 8psi). On average Enzyme-A increased the shear strength of soil-I by 9%, and by 23% the shear strength of soil II. On the other hand enzyme B increased the strength by 31% for soil I and 39% for soil II. It was also concluded that resilient modulus for all combinations of soil (I and II) and enzyme type (A, B) increases with curing time. They recommended that more mixtures of soils and enzymes be tested and laboratory data should be compared with field data. They also recommended 4 months curing time to achieve improvement in shear strength.

Shankar et al. (2009) They studied the effect of different dosages of Bioenzymes on Lateritic soil of Dakshina Kannada (district of India), having liquid limit and Plasticity Index more than 25%

and 6% respectively. Tests were conducted on lateritic soil by adding different percentages of sand as well. They concluded that there is medium improvement in physical properties of lateritic soil. Therefore it was suggested that effect of Bioenzyme on soil should be examined in laboratory before actual field application. Higher dosage (200ml/2m³ of soil) produced 300% increase in CBR, 450% in unconfined compressive strength and permeability was reduced by 42% after four weeks of curing. It was also observed that enzyme is not effective for cohesion less soil.

Peng et al. (2011) They conducted unconfined compression tests on three soils; fine-grained, silty loam and coarse grained textures named as Soil I, Soil II and Soil III respectively. Three soils were stabilized with quicklime and an enzyme (Permazyme). The samples were cured up-to 60 days in two different conditions; air-dry and in sealed container. In air-dry curing the samples were allowed to dry at room temperature where as in sealed container the moisture was preserved in the samples during the curing time. The enzyme was found more effective in air-dry curing for Soil I and Soil II than quicklime where as it was not effective for Soil III in air-dry curing and for three soils in sealed curing too. In sealed containers, the quicklime was found more effective than the enzyme as the water in the specimens was not allowed to evaporate which promoted the further hydration of quicklime.

Naagesh & Gangadhara (2011) This paper presents the effectiveness of a non –traditional liquid organic stabilizer mixed to an expansive soil in different proportions in reducing the swell characteristics of an expansive soil. Based on the experimental results, it is found that bio-enzyme is effective in reducing the swelling potential and swell pressure of an expansive soil when compacted at or above OMC. A reduction of about 50 % in swell potential and swell pressure is observed when soil specimens are treated with 2% bio-enzyme at OMC and reduction of 80% is observed when soil specimens are treated with 2% bio-enzyme slightly wet of OMC. The differential free swell of treated specimens also indicate a reduction of about 40% when treated with 2% bio enzyme. The reduction in swell is due to change in the fabric of the soil from flocculant structure to dispersed structure.

Subramanian & Dhinakaran (2011) They conducted tests on three soils with varied properties and different dosages of Bio-Enzyme. Three soils had liquid limits of 28, 30 and 46% and plasticity index of 6, 5 and 6%. Increase in unconfined compressive strength and CBR after 4 weeks of curing was reported as 152 to 200% and 157 to 673% respectively.

Agarwal, P. and Kaur (2014) The study reveals effect of Terrazyme on the Unconfined Compressive strength of the Black Cotton soil. Based on the tests, they found that stabilization of the soil using Terrazyme resulted in significant increase in the compressive strength of the Black Cotton Soil upto 200%. The soil treated with Terrazyme for 7 days gives higher strength. The optimum dosage of Terrazyme for improvement of UCS of Black Cotton soil is 1ml/per 5kg of soil.

Sharma et al. (2017) The study provides an effective technique of ground improvement using bio-enzyme. In this study a bio-enzyme named terrazyme is used for improving the California bearing ratio (CBR) value in road construction. Terrazyme can be used as soil stabilizer and also it can improve the CBR value in road construction. The dosage of terrazyme are taken as 500ml/m³, 700ml/m³, 900ml/m³ and 1000ml/m³ in the soil sample and result is analyzed. A significant increase is found in CBR value of the soil sample as the dosage of terrazyme has been increased.

Manu & Mahendra (2018) The paper studies the effects of varying dosages of Terrazyme on strength properties of Black Cotton Soil for different curing periods. Tests were carried out to determine the Atterbergs limits, Unconfined Compressive Strength and California Bearing Ratio of the soil specimens with and without Terrazyme subjecting to desiccator curing. Addition of various dosages of Terrazyme decreases the liquid limit and plastic limit of black cotton soil. Treating black cotton soil with Terrazyme, the strength increases for all dosages of Terrazyme, also an increase in unsoaked CBR value is observed for optimum dosage of Terrazyme.

Joshi & Solanki (2019) they studied the effect on two different soils stabilized with bio-enzymes along with lime. The tests are carried out to determine the consistency limits, compaction characteristics, unconfined compressive strength, California bearing ratio. The laboratory test results show promising results in terms of strength of the stabilized soil. Strength characteristics like UCS and CBR value were improved by 14 times and 8 times to its initial value, respectively, and the soil which was highly expansive became low expansive after treatment.

Mittal (2020) The study reviews the available knowledge on use of bio-enzymes in particular terrazyme for soil stabilization purpose. It was found that use of bio-enzyme in soil leads to formation of calcium silicate hydrate (CSH) compound having cementitious properties. Application of terrazyme in expansive soil reduces its plasticity characteristics and provides volumetric stability thus reducing susceptibility to crack formation. The strength of un stabilized earthen

construction and adobe blocks can be increased effectively. Being a natural product (extracted from vegetables and fruits) it is eco-friendly. Its application in subgrade soil significantly reduces the thickness requirement of pavement thus saving costly base and sub-base aggregate materials.

CHAPTER 3

Bioenzymes

3.1 General

Bioenzymes are organic molecules that are derived from natural sources and can act as catalysts in chemical and biological processes. They are also known as fruit enzymes or garbage enzymes. Bioenzymes are made from organic waste, plant and animal tissues, and microorganisms. They are biodegradable and have a lower carbon footprint than traditional chemicals. Bioenzyme is a natural, non-toxic, non-corrosive and non-flammable liquid enzyme formulation fermented from vegetable extracts that improves the engineering property of soil and increases the stability by accelerating the reactions between the clay and the organic cations and accelerates the cationic exchange process to reduce the diffused double layer thickness (Manu A.S, S.P Mahendra, 2018). They are water-soluble, dark in color, and have an odor resembling molasses. Organic enzymes are usually in a liquid state. They are utilized in commercial agriculture applications and have been discovered to have the ability to increase the stability of soils when used with specific soil types. The bioenzyme is supposed to change the engineering qualities of soil. Before being used, they must be diluted in water depending on the type of soil and enzyme dose; adding bioenzyme to water and mixing it with soil changes the engineering characteristics. These enzymes are liquid additives that react on the soil to decrease adsorbed water and remove voids between soil particles for optimum compaction (Naik et al. 2020).

3.2 Bioenzymes as stabilizers for soil

Enzymes are the catalysts of biological systems that not only control the rate of reactions but also can lower the activation energy for the formation of one product from another by favoring certain geometries in the transition state (D. E. Scholen, 1995). Bioenzymes are protein molecules that catalyze chemical reactions in the soil to form a cementing bond that stabilizes the soil structure and reduces the soil's affinity for water (S. Tingle, 2007). The idea of using enzyme stabilization for soil pavement was developed from the application of enzyme products used to treat soil in order to improve horticultural applications (M. Taha, 2013).

Bioenzymes work on a variety of soils as long as the minimum amount of clay particles is present. Enzymes may work suitably for soils containing 12–24% clay fraction with a plasticity index between 8 and 35 (Khan &Taha, 2015). When applied at low application rates to the surface of the unbound road surface, enzymatic emulsions perform well for dust suppression (S.M Lim, 2014). At higher application rates, enzymatic emulsions can be used to stabilize unpaved and paved roads, paths and shoulders, access roads, unpaved and paved parking lots, orchards and crop roads, mining haul roads, access roads, parking areas, airfields, minor rural roads, property driveways, and where you need to improve the engineering properties of road bed materials (ALBC, 2014). When applied and compacted properly, the treated soils can be stabilized to form a dense, firm-to hard, water-resistant bound layer that can be used as a road surfacing.

3.3 Comparison between Bioenzymes and Traditional Stabilizers

Traditional stabilizers such as cement and lime are relatively expensive and in some areas the cost would be up to three times the cost of bioenzymes and become even more so when they have to be transported long distances to low-volume road construction sites, because they are bulky (A. T. Visser, 2007). On the other hand, bioenzymes are usually sold as concentrated liquids, diluted with water at the construction site and then either spread on the soil before compaction or pressure injected to treat deeper soil layers (Rauch et. al 2001). Due to this fact, it is possible to transport with relatively reduced price. Because of the lower transportation costs, concentrated bioenzymes can be an attractive alternative for stabilization projects. As a consequence, unlike the traditional soil stabilization techniques, bio-enzymes are the cheapest, nontoxic, environmentally friendly, and organic technology.

As a consequence, recently more attention has been given to the use of bioenzymes as soil stabilizers. This is due to the expansion in manufacturing capacity, low cost, and relatively wide applicability of the enzymes as compared to standard stabilizers requiring large amounts of stabilizers to stabilize soils which in turn increases manufacturing cost.

3.4 Mechanism of Bioenzyme Soil Stabilization

The major problems for soil stability during any construction is the nature of the clay constituting the soil mass. Clayey soils have a high affinity for water because of their small particle size and high surface activity. Thus, the particles are almost always hydrated, that is, surrounded by layers

of water molecules adsorbed onto the clay particles. This affinity for water can be attributed to hydrogen bonding (oxygen or hydroxyl molecules attract the hydrogen of water), Van der Waals attractions, and charged surface dipole attractions (Fig 1). It is this water layer that affects all soil properties including plasticity, compaction, strength, and water movement in the soil. Among these different types of bonding, the hydrogen bonding is the strongest and is considered to be the primary reason behind the swelling of expansive soils due to water absorption. Due to this, montmorillonite clays suffer volume changes due to moisture content changes which results in swelling and shrinkage. This phenomenon is influenced by many clay properties including specific surface area, cation exchange capacity, organic matter content, and availability of soil stabilizing agents. Soil stabilizers bond soil minerals together and lead to suppression of swelling by increasing strength to the soil material.

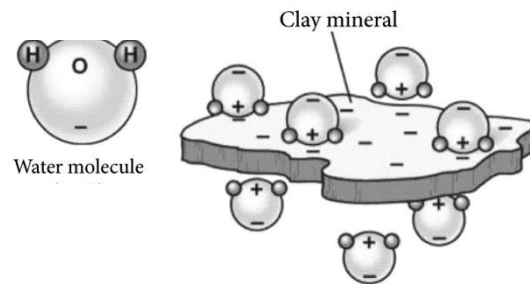


Fig. 1 Clay particle and surface charge display (Mekonnen 2020)

The clay particles hold a high concentration of cations to balance the negative surface charges attributed to the presence of broken bonds and isomorphous substitution. These cations are termed as “adsorbed cations” and are strongly held by the negatively charged clay particles. The cations tend to diffuse away from the clay surface in order to balance the low cation concentration within the absorbed water. However, this kind of diffusion is offset by the electrostatic attraction between the positively charged cations to the negatively charged clay surface, which is more dominant close to the clay particles. The negatively charged clay surface, along with the strongly held cations (close to the clay particle) and the relatively loosely held diffused cations (further away from the clay particle), form the diffuse double layer. This diffuse double layer governs the clay-water interaction and affects the engineering properties of clay, including swelling and plasticity (Fig 2).

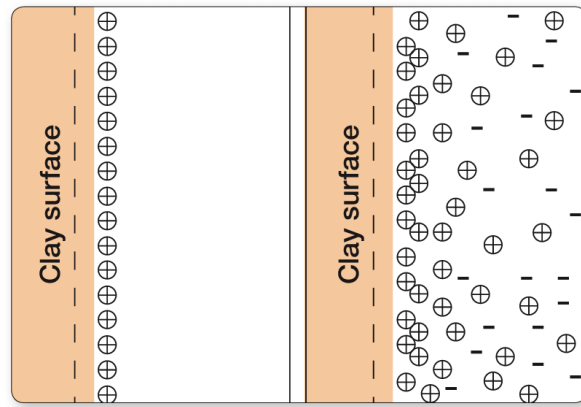


Fig. 2 Representation of Diffuse double layer at the clay surface to soil-water interface (Source: <https://images.app.goo.gl/twKKmxm5HRrWWNU68>)

Unlike traditional stabilizers, the attempts done to define the stabilization mechanisms of nontraditional stabilizers including bioenzymes have been limited. Based on the various perspectives on the mechanisms of bio-enzymes, it can be summarized that there are primarily three mechanisms: cation exchange, specific binding, and surfactant. Two mechanisms of bioenzyme soil stabilization were proposed by researchers. The first proposed mechanism of stabilization explained that the enzymes that are present in treated soil are adsorbed by the clay lattice, and in turn cations are released as an exchange, a process similar to cation exchange. For instance, Pooni et. al 2021 proposed that bioenzyme is adsorbed onto clay minerals, inducing relaxation of the mineral lattice. This relaxation leads to inner-layer expansion and subsequent moisture retention. Consequently, there is a reduction in the absorption of moisture within the soil structure, lowering its affinity for water and mitigating the impact of moisture on the soil.

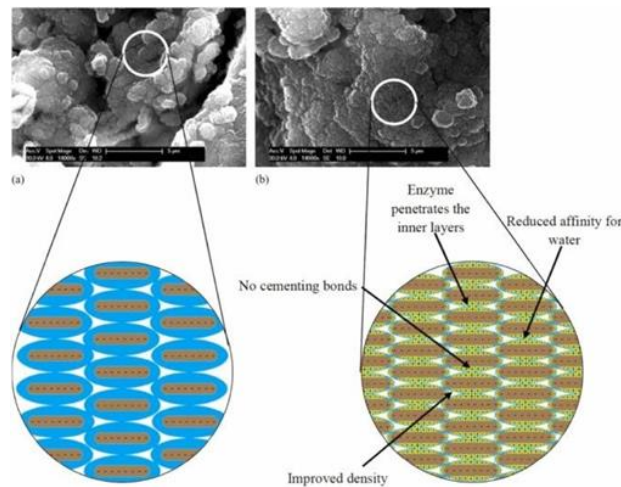


Fig. 3 Schematic illustration of the enzyme stabilization mechanism on expansive soil (Pooni et al., 2021):(a) untreated soil; and (b) stabilization soil.

The other widely accepted hypothesis of bioenzyme soil stabilization mechanism is specific binding, proposed by (Scholen 1995), when bioenzyme formulations are mixed with soil, enzymes combine with big organic molecules in the soil solution to generate a reactant mediator. The large organic molecules have large flat structures that approach the size of small clay particles which can blanket the clay minerals, neutralizing the negative charge and reducing the clay's affinity for moisture. As a result, this produces a covering effect, which blocks further absorption of water and loss in density.

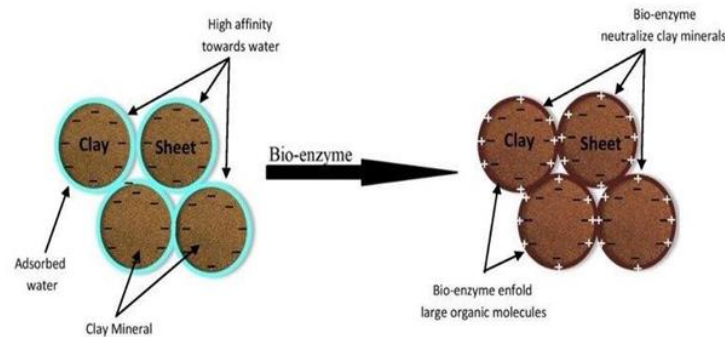


Fig. 4 Enzyme stabilization mechanism (Renjith et al., 2020): (a) natural clay particle with high affinity for water; (b) organic encapsulation decreasing the double layer of water; and (c) stable clay particles.

The third mechanism, surfactant action, reduces surface tension of soil particles, thereby lowering capillary suction and the thickness of the water absorption layer. All the mechanisms collectively decrease the affinity of soil for water, reduce the thickness of the electric double layer, and facilitate soil compaction, ultimately achieving soil stabilization. Overall, although these mechanisms are functionally similar, there are differences in the way they perform their roles: they function through the adsorption of the enzyme by the clay lattice, the specific binding of the enzyme to organic molecules surrounding clay particles, and the reduction of surface tension by surfactants, respectively.

Several researchers showed the formation of stable clay lattice structure and a reduced affinity for moisture after treatment with various bioenzymatic formulations. Rauch et al. 2003, through different chemical and physical tests, endorsed the hypothesis proposed by Scholen stating that enzymes unite with the large organic molecules and adhere to clay surfaces, thus jamming potential cation exchange sites and preventing absorption of moisture and subsequent swelling. In

addition, in their separate studies, Santoni et al. 2003 & Tingle et. al 2007 reported a series of laboratory tests with various bioenzymatic stabilizers evaluating the performance effects in terms of increased strength improvement in both granular and fine-grained subgrade materials. However, these experiments only categorized the proposed stabilization mechanisms as either a mechanical bonding or a chemical reaction mechanism with no details of the proposed physicochemical changes.

However, Rauch et al. (2003), Lindembaum (2008), C. Stanet. al (2012) suggested that soil suitable for bioenzyme stabilization should have chemical substances like clay minerals that may react with other chemicals. They indicated that enzymes are appropriate only for use with clay materials that have an affinity for water, particularly high plasticity clays with some organic content. Thus, materials such as silts and granular soils would not possess a significant affinity for water and would be unsuitable for stabilization with enzyme products. In addition, literatures suggested that the use of enzymes will also be critically dependent on the environmental conditions and may take considerable time to occur. Lindenbaum in his patent publication also explained a mechanism that bioenzyme during soil stabilization breaks down the electric double layer between the clay and adsorbed water. By this, the clay particles lose its inherent charge and loose the adhered static water layer. In this mechanism, the clay particles segregate and are so fixed crystallographically that it prevents any further volume changes on exposure to water. He also added that organic cations generated by the growth of vegetation and microorganisms will have the capability to exchange position with other ions attracted to the clay platelet in the soil. In contrast to metal cations, the organic cations have large flat structures that approach the size of small clay particles. These organic cations can blanket the clay particle and effectively neutralize its negative charge in a short distance, thus greatly reducing the double layer thickness.

CHAPTER 4

Types of Bioenzymes

4.1 General

Different types of bio-enzymes have found their way into soil stabilization and have proven to be extremely efficient and cost-effective (Vedula et al. 2002), for its unique properties and applications. Some common types of bio-enzymes used for soil stabilization :

- **Terrazyme**

TerraZyme is a natural, non-toxic liquid, formulated using vegetable extracts and accepted all over the world as a sound and resourceful road building practice, which completely replaces the conventional granular base and the granular sub base, it emphasizes on strength, performance and higher resistance towards deformation. Terrazyme is specially formulated to modify the engineering properties of soil. They require dilution in water before application. The use of Terrazyme enhances weather resistance and also increases load bearing capacity of soils. These features are particularly evident in fine-grained soils such as clay in which the formulation affects the swelling and shrinking behavior. This formulation has the ability to change the matrix of the soil so that after compaction the soil loses its ability to reabsorb water and the mechanical benefits of compaction are not lost even after water is reapplied to the compacted soil. Once the enzyme reacts with the soil, the change is permanent and the product is biodegradable.

Advantages of Terrazyme (Rajoria and Kaur, 2014)

- a) Terrazyme improves the pavement's resilience and minimizes the soil's swelling qualities.
- b) Due to the reduction in material transportation and reuse of onsite resources, subgrade costs are reduced by around 20–40%.
- c) The usage of Terrazyme enhances the resistance of soil to weathering and load-bearing capability.

- **Permazyme**

Permazyme is a compaction enzyme, when it is added to a soil and aggregate mixture; it causes the compaction of clays and silts with a much faster rate than that occurs in nature. According to the manufacturer, this enzyme is a natural organic compound, similar to proteins, which acts as a catalyst. Their large molecular structures contain active sites that assist molecular bonding and interaction. The organic formulation is designed to maximize compaction and increase the natural properties of soil to optimal conditions. This enzymatic stabilizer increases the wetting action of water to help achieve a higher density during compaction and the formulation accelerates cohesive bonding of soil particles, creating a tight permanent stratum (Agarwal and Kaur 2014).

Advantages of Permazyme (Rajoria and Kaur 2014)

- a) Permazyme enhances the lubricity of soil particles, facilitating less compaction effort to achieve the desired soil density.
- b) It increases quick saturation and prevents surface evaporation, lowering the water need by up to 25% of the optimum moisture content.
- c) Permazyme is a biodegradable and eco-friendly product.

- **Fujibeton**

The Fujibeton material, developed in Japan, is climatically stable material and suitable for stabilization of all types of soils. Basically, the product is an inorganic polymer that chemically binds with all compounds, when blended with ordinary Portland cement in 1 to 3% by weight of OPC. The blended mix is called Fujibeton Mix, which is used for soil stabilization to improve the engineering properties of soil.

Advantages of Fujibeton (Rajoria and Kaur 2014)

- a) Fujibeton enhances the CBR of the subgrade while preventing shrinkage cracks, making it ideal for clayey/ soils.
- b) For embankments and subgrade and sub-base course construction, this approach is both efficient and cost effective.
- c) Fujibeton achieves a high dry density with minimal compaction.

- **Renolith**

Renolith is polymer based chemical, which is environmental friendly and which facilitates the bonding of soil particles. A mixture of Renolith and water in the right proportion can be utilized in cement-based aggregates or other soil types. When Renolith is blended with soil, it produces an exothermic reaction. The reaction results in a highly dense layer, which stabilizes the soil. When the water evaporates from the blended soil, the Renolith stabilizer covers the soil particles and forms a physical link between them, resulting in a soil–polymer. The tensile strength and elasticity of this soil polymer are both high. These characteristics can reduce the risk of any breaking of the asphalt layer due to shrinkage of the concrete or roadway foundation during compaction, as well as improve porosity Jayalekshmi & Reddy (2012), Singh & Garg (2015).

Advantages of Renolith (Rajoria and Kaur 2014)

- a) The usage of Renolith can reduce the cost of pavement construction by 20 to 40%.
- b) Since an aggregate of requisite specifications needs not to be imported, locally available material can be used.
- c) Renolith gives the pavement sufficient flexibility and resilience to prevent fractures from forming.

CHAPTER 5

Effect of bioenzyme on properties of soil

5.1 Consistency of Soil

Consistency refers to the relative ease with which the soil can be deformed. The addition of bio enzyme alters the consistency, ultimately helping in the stabilization of soil. Consistency of soil can change with the type of bio enzyme and variation of dosage of bio enzyme. With adequate curing, the addition of bio enzyme to the natural soil resulted in a reduction in both liquid and plastic limits. However, the reduction in liquid limit was higher, leading to an overall reduction in the plasticity index as well. This could be attributed to the fact that, at the liquid limit, the water-holding capacity of the soil is more compared to that at the plastic limit, leading to better electrolyte movement in the soil. This enhanced electrolyte movement would have enabled better interaction of bio enzymes with soil particles. On the other hand, the plastic limit has shown a decrease with the curing period. This may probably be due to the aggregation of soil particles as a result of the stabilizing effect of bio enzyme. This reduction of the liquid limit and plastic limit of bio enzyme-treated soil with the curing period indicates that the addition of bio enzyme to the soil reduces its plasticity (Ravi Shankar et al. 2009; Chitrakar et al. 2019). The reduction in plasticity index, when the bioenzyme was added to an expansive soil, was found to be maximum when the dosage was 150 ml/m³ of the soil. When the dosage was increased beyond this level, the test results were adversely affected (Vinay Kumar et al. 2020).

A majority of experimental studies indicated that the dosage of the bio enzyme required to be added for optimum improvement in the plasticity characteristics was around 150–200 ml/m³ of soil. The plasticity index of soil was reduced by 30% for a 150 ml/m³ dosage and 15% for a 200 ml/m³ dosage (Vinay Kumar et al. 2020). The Liquid Limit and Plasticity index of Shedi soil decreased by 30.20% and 86.86%, respectively, compared to their original values after 30 days of curing for the dosage of 200 ml/m³ of soil (Naik et al. 2020).

5.2 Permeability of soil

Permeability, also termed hydraulic conductivity, is the property of soil by which it allows the flow of fluid through it. In the laboratory, the coefficient of permeability is determined by allowing water to pass through compacted specimens and measuring the rate of flow of water through the soil samples. Generally, the addition of bio enzymes lead to a reduction in soil permeability (Singh and Garg 2015, Ravi Shankar et al. 2009) . For a 14-day curing period, the permeability with the silty sand samples, amended with 2–10% cement by weight of soil, reduced drastically with the addition of increasing Renolith dosage. Bio enzymes were not equally effective in all types of soils. For example, in laterite soils, the influence of Terrazyme was found to be minimal with only about 25% reduction in the permeability of soil treated with 100 ml/m³ of Terrazyme after four weeks of curing (Ravi Shankar et al. 2009).

5.3 Compaction characteristics of soil

Compaction characteristics define the packed state of soil and can be improved by compaction of soil, which is the process of artificially rearranging the soil solids by applying external pressure. Compaction reduces the void ratio, permeability, and compressibility and increases the degree of denseness, stability, shear strength, and bearing capacity. Experimental studies prove that with a proper curing period, the optimum moisture content (OMC) of soil increased marginally, coupled with a corresponding decrease in the maximum dry density (MDD) with increasing dosage of bio enzyme (Ganapathy et al. 2017). Certain types of soil responded extremely well to the bio enzyme addition. The OMC decreased by about 14.8%, and the MDD increased by 61.59% after 28 days of curing upon adding Terrazyme to locally available soil in Kerala used for road construction (Athira et al. 2017). At a dosage of 200 ml/m³ of soil and after 30 days of curing, the MDD of bio enzyme-treated Shedi soil increased by 19.42%, whereas the OMC of soil reduced by 38.62% (Naik et al. 2020). As the water is disseminated, a cementing action takes place, where the smaller soil particles come close to each other and fill the voids within the soil, creating densely packed soil. This reduction in OMC is due to the effective cation exchange process which generally takes more time in the absence of such bio enzyme (Ganapathy et al. 2017). The addition of 5% Renolith to clays amended with 10% cement resulted in a reduction of 10% OMC and almost an equal increase in MDD (Singh and Garg 2015).

5.4 Unconfined compressive strength test

The addition of bioenzyme to clayey soil, silty clay, and sandy soil increased the UCS of soil. The UCS of fine grained soils improved by almost 25% when Terrazyme was added at a dosage close to 100 ml/m³ and cured for 7 days. When the dosage was increased to about 200 ml/ m³, this improvement was close to 45% (Sahoo and Sridevi 2018). Similar test results were obtained by Rohit and Thakur (2020), wherein an increase of 37% in the UCS of Terrazyme-stabilized soil was observed. Samples of black cotton soil showed a drastic improvement in the UCS of about 232% under an optimal Terrazyme dosage of 100 ml/ m³, whereas the improvement in red soil under the same dosage was about 150% after curing in a desiccator. For both these soils, curing by air drying gave better results (Ramesh and Sagar 2015). The use of Terrazyme in Black cotton soil, at a dosage of about 400 ml/m³ of soil, resulted in a 200% increase in UCS values (Agarwal and Kaur 2014). The UCS value of lateritic soil, at a Terrazyme dosage of 100 ml/m³ of soil, improved the UCS to about 450% after four weeks of curing (Ravi Shankar et al. 2009). The addition of about 8% lime to Terrazyme-stabilized soil further improved the UCS values. Similar test results were obtained for fly ash–Terrazyme mixes as well (Aswathy et al. 2021).

5.5 California Bearing Ratio Test

The CBR test is performed to determine the mechanical strength of the base course and subgrade underlying the roadway. CBR values for both unsoaked and soaked samples increased significantly with the addition of bioenzyme to kaolinite, red soil, and lateritic soil. The optimum stabilizer percentage for maximum improvements in CBR values in red soil, laterite, and kaolinite was observed as 4%, 2% and 6%, respectively (Mamta and Honna 2014). The CBR values for unsoaked native soil samples increased by almost 40% after 4 days of curing when the Terrazyme dosage was 0.05%. Soaked CBR values showed sufficient improvement in Terrazyme addition, although less than those shown by soil in the dry state (Agarwal and Kaur 2014). The CBR value of Black cotton soil improved from 3 to 15% upon adding Terrazyme at a dosage of 100 ml/m³. For red earth, the same dosage showed an improvement in CBR from 7 to 32% (Ramesh and Sagar 2015). When comparing the CBR value of treated soil to untreated blended soil, larger curing durations resulted in a higher CBR value. After one week of curing on 10% blended soils, the CBR increased by 238%, which further increased to 300% after four weeks of curing. Similarly, after curing for one to four weeks, the CBR of 20% blended soil improved from 200 to 288%, 30% blended soil

improved from 175 to 275%, and 40% blended soil improved from 163 to 250%. The CBR values of lateritic soil mixes were found to increase with an increase in the duration of the curing period (Ravi Shankar et al. 2009).

CHAPTER 6

Advantages and Disadvantages of Bioenzymes

• Advantages

Bioenzymes offer several benefits when utilized for soil stabilization, making them an attractive alternative to traditional stabilizers. It enhances the physical properties of soils, such as reducing the liquid-plastic limit and plasticity index, which leads to improved strength and stability in engineering applications. This improvement is particularly valuable for construction projects, as it increases the durability of the infrastructure in which the stabilized soil is used. Moreover, they are cost-effective compared to conventional stabilizers like cement and lime, which can be significantly more expensive, particularly when transportation costs are considered. Bioenzymes are typically sold as concentrated liquids that can be diluted on-site, making logistics simpler and less costly for projects. Bioenzymes are also environmentally friendly and non-toxic, contributing to a reduction in environmental pollution associated with traditional stabilization methods. Their use promotes sustainable construction practices by minimizing carbon emissions often associated with conventional stabilizers. Additionally, the ability to produce bioenzymes from local raw materials allows for the development of low-cost and widely applicable solutions tailored to specific regional needs.

• Disadvantages

Despite their advantages, there are also several disadvantages associated with the use of bioenzymes for soil stabilization. One major concern is the variability in soil types and conditions. Effectiveness can be highly dependent on the characteristics of the soil, such as clay content and moisture levels. Enzymes may work best in soils with specific clay fractions and plasticity indexes, and their performance can diminish in soils lacking these characteristics.

Additionally, there is limited research available on the long-term effects and performance of bioenzymes in various environmental conditions. Many studies focus primarily on performance evaluations, neglecting the mechanisms by which bioenzyme stabilization actually occurs. This gap in knowledge can hinder widespread adoption, as construction engineers may prefer

established technologies with extensive proven records. Furthermore, the high initial cost of bioenzyme formulations may pose a barrier, particularly for large-scale projects. Although they can be cheaper in the long run, upfront investment may discourage some stakeholders from choosing this option. Additionally, production procedures and specific formulations are often proprietary, which can limit access to detailed information on their use and effectiveness.

CHAPTER 7

Conclusion

Bioenzymes present a promising and environmentally friendly alternative for soil stabilization, offering numerous benefits such as biodegradability, renewable sourcing, and improved soil health. The use of bioenzymes in soil stabilization contributes to sustainable practices, aligning with global efforts towards eco-friendly solutions and reducing dependence on traditional stabilizers derived from non-renewable resources. Due to its huge economic impact and nontoxicity on the environment, bio-enzyme holds the most promising key for developing countries. Various more researches are currently being done on enzymes like Terrazyme, Permazyme, Fujibeton and Renolith to further know about their benefits. While bioenzymes demonstrate significant potential, it is essential to acknowledge certain limitations. These include variability in performance based on environmental conditions, concerns about long-term stability, and challenges related to ease of use and production costs. Researchers and industry practitioners continue to address these limitations through ongoing advancements in bioenzyme formulations, production processes, and application techniques. As the field of bioenzyme research evolves, it holds promise for contributing to sustainable soil management practices, erosion control, and ecological restoration. Further interdisciplinary collaboration, robust scientific inquiry, and practical field applications will play crucial roles in realizing the full potential of bioenzymes as effective and sustainable soil stabilizers.

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