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Comparative Studies on the Strength and Swell Characteristics of Cohesive Soils Using Lime and Modified Enzyme-Induced Calcite Precipitation Technique

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Abstract: Enzyme-induced calcite precipitation (EICP) emerges as a highly effective and well-established technique within bio-cementation approaches, offering notable advantages over traditional methods. Conversely, lime, known for its accessibility, cost-effectiveness, and efficacy, serves as a valuable material in enhancing the engineering properties of problematic soils. This study explores the application of EICP and lime treatments separately on two distinct soils (low-plastic and high-plastic soil) exhibiting different mineralogical and plasticity characteristics to assess their impact on strength and swell characteristics. Various combinations of treatments, including jack bean (JICP), soya bean (SICP), and bio-enhancer (BICP), were employed for EICP treatment. Bio-enhancer, rich in natural urea and urease enzyme, was particularly remarkable due to its compatibility with urea supplementation. Similarly, jack bean and soya bean exhibited high efficacy in natural urease enzyme content. The study has revealed that the unconfined compression strength (UCS) of red soil increased significantly by six times at the end of 21 days of the curing period with JICP treatment, while lime treatment was more effective for the black soil. Specifically, the UCS of black cotton soil increased by 11 and 17 times when treated with Enzyme-Induced Calcite Precipitation (EICP) and lime, respectively. Moreover, EICP with J2 solution (jack bean solution with 1M urea and 4 g/L non-fat milk powder) reduced swell pressure by 60% and 67.5% in low-plastic and high-plastic soil, respectively. Lime treatment, on the other hand, led to a swell pressure reduction of 47% and 70% in low-plastic and high-plastic soil, respectively. As a result, EICP proved efficient in mitigating swell pressure for red soil, whereas lime treatment performed exceptionally well for black soil, highlighting the soil-specific effectiveness of each method. Furthermore, a life cycle assessment revealed substantial carbon footprint emission savings with EICP treatment strategy. In brief, this paper contributes to understanding the phenomena and significance of these two treatment techniques on distinct mineralogical soils.

Keywords: soil stabilization; urease enzyme; soya bean; jack bean; bio-enhancer; lime; low-plastic soil; high-plastic soil

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1. Introduction

Owing to rapid urbanization and an increase in population, the expansion of cities has necessitated the utilization of sites that were previously deemed unfavorable for foundations. Enhancing the engineering properties of these adverse sites for geotechnical application is crucial. The conventional technique for ground improvement involves mechanical stabilization and grout injections. Since time immemorial, chemical amendments such as lime, cement, and their derivatives have served as soil admixtures due to their short-term and long-term benefits in soil amelioration [1]. The materials, including lime, fly ash, cement, etc., effectively improve the soil's engineering properties for different field applications [1–3]. However, other materials, such as nanomaterials, chitosan, gaur gum, biochar, etc., have proven their efficiency in stabilizing the soil [4]. However, they have disadvantages: they are costly, show less improvement, have unfavorable for severe climatic conditions, etc. [5–8]. Lime is one of the oldest products used as a construction material, and its significance in the geotechnical field has recently been observed. This lime is widely available in different forms, such as CaO and $Ca(OH)_2$. Quick lime is highly reactive when added to the water and evolves large heat with a hissing sound [9]. Due to the exothermic reactions, the hydrated lime, which is relatively less reactive, is formed [10]. Adding lime to the soil can affect the plasticity indices, which reduces the liquid limit and increases the plastic limit, thereby reducing the plasticity index. Soils dominated by clay minerals like montmorillonite exhibit a highly expansive nature with significant swelling characteristics [11]. The lime treatment can be very effective in reducing its plasticity and reducing its affinity to water adsorption. Then, probable swell pressure also decreases when lime is added to the highly expansive soil [12]. The lime is used in this study to observe the index and engineering properties of different mineralogical soils.

Bio-stabilization of soils through EICP and microbial-induced calcite precipitation (MICP) techniques holds the potential to achieve the escalating needs of establishing new infrastructure and remediating contaminated soils [13]. The bio-stabilization of soil represents an evolving trend characterized by comparatively easy in situ applications [14]. This technique involves the injection of various enzymes into the soil for soil improvement. The formation of precipitates within the soil voids proves beneficial in reducing hydraulic conductivity [15]. Likewise, the MICP technique resulted in the reduction of hydraulic conductivity effectively in sandy soil [16]. However, considerations regarding the application and the type of stabilizers on-site raise concerns.

So far, different techniques, including mechanical, bio-mediated, and electrical, have proven their sustainability, eco-friendliness, and effectiveness in soil treatment, resulting in enhancements in the engineering performance of soil. These improvements include reduced permeability, decreased soil mass porosity and dust, increased the control of soil erosion and stabilization of slopes, and enhanced bearing capacity, liquefaction mitigation, seepage control, and contaminant remediation [17–20]. From the previous studies, it is observed that the EICP treatment has proven its efficiency in increasing soil erosion resistance and soil stability against rainfall. The EICP enhanced with fibers increased the engineering properties of soil [21]. Similarly, the coupled effect of EICP with sodium alginate achieved higher strength values [22]. The size of the bacteria constrains its intrusion into soils for calcite precipitation. Pores present in the soil with dimensions less than 0.5 μm does not allow the microbes to accommodate in it and hinder the calcite precipitation process, as their sizes typically vary from 0.5 μm to 3 μm. Meanwhile, many enzyme particulates, with a size of about 12 nm, facilitate calcite precipitation even in clays containing fines [13]. Treating the soil using MICP/EICP may expose it to challenges such as groundwater contamination with chloride due to the $CaCO_3$ precipitates [23]. Applicability issues include soil type and cost, which further studies can address for improved bio-treatment methods. While bio-stabilization is environmentally friendly, excessive calcite formation may contaminate groundwater with chloride, and the release of ammonia ions and increased pH pose potential threats to air and water quality and corrosion risks [24].

EICP is favored over MICP among the two methods due to its advanced nature and lower monitoring requirements. Literature indicates that precipitates formed by MICP are susceptible to moisture and may dissolve [8,23,25]. Maintaining the proper environment for bacterial growth and urease enzyme production poses challenges [26]. In contrast, free urease is more promising for calcite precipitation in soil grain voids. The EICP method offers a convenient approach to soil treatment due to its ease of application and lower maintenance requirements than the MICP method [27].

The following gaps have been found from the earlier works: MICP and EICP are effective, but economy-wise, they are on a higher range and may not be feasible for general applications; in terms of life cycle analysis, the amount of greenhouse gases and carbon footprints are also on a higher range. Hence, the use of naturally available ingredients in soya bean, jack bean, and bio-enhancers has been evaluated in this study to act as a replacement for EICP synthesis.

Calcite precipitation occurs due to hydrolysis of urea in the soil environment in the presence of the urease enzyme, which acts as a catalyst [28]. This happens through the breakdown of urea, leading to ammonia (NH_3) and carbon dioxide (CO_2) formation as expressed in the below Equation (1). Thereby, the ions of bicarbonates and hydrogen will be generated with the dissolution of formed CO_2. NH_3 is dissolved in water to form NH_4 and hydroxide, as shown in Equation (2). The increase in the pH value takes place due to the formation of these OH^- ions.

$$H_2N - CO - NH_2 + 2H_2O \xrightarrow{\text{Urease}} H_2CO_3 + 2NH_3 \quad (1)$$

$$HN_3 + H_2O \rightleftarrows NH_4^+ + OH^- \quad (2)$$

$$HCO_3^- + OH^- \rightleftarrows CO_3^{2-} + H_2O \quad (3)$$

$$CO(NH_2)_{2(s)} + 2H_2O \xrightarrow{\text{urease enzyme}} 2NH_4^+{}_{(aq)} + CO_3^{2-}{}_{aq} \quad (4)$$

Subsequently, with an increase in pH, the bicarbonates and hydrogen ions will undergo a reaction, forming carbonates as expressed in Equations (3) and (4). The increased pH helps to merge dissolved Ca^{2+} ions with carbonates to form calcium carbonate precipitation. This process results in the formation of calcite with the generated carbonates. The presence of water leads to the dissolution of ammonia and hydroxide ions. This work studied the utilization of soya bean, jack bean, and bio-enhancers in the preparation of urease solution. Soya beans are grown on a large scale, are rich in many nutrients, and have a protein content of 3.5%, including the soy urease enzyme [29]. In this case, soya was used as a natural replacement for Jack bean-based urease enzyme [30]. Bio-enhancer is a rich source of nutrients to soil microorganisms, and it contains an excess of 95% water, 2.5% urea and minerals, 24 types of salts, hormones, and 2.5% enzymes, and it stimulates urea hydrolysis in the soil, leading to CO_2 emissions [31]. Hence, it can be inferred that bioenhancer is a better source of urea than urease enzymes. Therefore, in this study, it was decided to replace urea in our samples and maintain the supply of urease enzyme from soya and jack bean. Life cycle assessment (LCA) on all three bio-stabilizers was conducted using data from literature and by applying openLCA 2.0, an open-source software, to conduct LCA analysis. The results obtained are discussed in detail [3]. In brief, this paper deals with the strength and swell characteristics of two different soils treated with EICP and lime material.

2. Materials

2.1. Soils

The soils were collected from 13°2′14.1396″ N, 77°37′11.928″ E, red soil (Soil 1), Bengaluru District, India, and 16°45′20.556″ N, 77°9′4.5072″ E, black cotton soil (Soil 2), Yadgir district, India. In accordance with ASTM D2487-17e1 [32], soil 1 and soil 2 were classified as low-plastic (CL) and high-plastic soils (CH), exhibiting a liquid limit of 30 and 54%, with a plastic limit of 17 and 27%, respectively. The optimum moisture content (OMC) of 16.8 and 15.2% with specific gravity of 2.6 and 2.5 were observed in soils 1 and 2, respectively. As per XRD analysis, the soils were identified as kaolinite and montmorillonite based on the mineral phase observed in the respective soils. These basic properties of the soils are illustrated in Table 1.

Table 1. Physical properties of soils.

Properties	Soil 1	Soil 2	Standard
Specific gravity	2.6	2.5	ASTM D854-23 [33]
Liquid limit, W_L (%)	30	54	ASTM D4318-17e1 [34]
Plastic limit, W_P (%)	17	27	ASTM D4318-17e1 [34]
Plasticity index, I_p (%)	13	27	ASTM D4318-17e1 [34]
Soil classification	CL	CH	ASTM D2487-17e1 [32]
Maximum dry density (g/cm^3)	1.81	1.76	ASTM D698-12 [35]
Optimum moisture content (%)	16.8	15.2	ASTM D698-12 [35]

2.2. Enzymes and Other Ingredients

Three different components representing urease enzymes were taken: jack bean urease, bio-enhancer, and soya bean-based urease. Jack bean was obtained from type III powder, 15,000–50,000 units/g solid (Urease Enzyme), and was supplied by Sigma-Aldrich. Similarly, a fresh bovine fluid was obtained from a dairy farm having a pH of 8. It was used immediately, and the bovine fluid was filtered with a stainer to remove any grit. Bio-enhancer was obtained by diluting the filtrate in a ratio of 1:10, with u/w ratio representing one part of the bovine fluid to 10 parts of double distilled water; the mixture was kept in containers under low temperatures. The soya bean was ground into fine powder and then soaked in double distilled water in a solid-to-liquid ratio of 1:10 for 120 min in a refrigerator. This mixture was filtered in gauze to obtain soya bean powder filtrate, which was directly used for all the experiments. Additional ingredients were prepared, and their concentrations and combinations are given in Table 2. It includes calcium chloride dihydrate ($CaCl_2$ $2H_2O$), chosen for its effectiveness in $CaCO_3$ precipitation, as mentioned in Equations (5) and (6). Urea (NH_2-CO-NH_2) and non-fat milk powder are also integral components of the mixture.

$$CaCl_{2(s)} \xleftrightarrow{H_2O} Ca^{2+}_{(aq)} + 2Cl^-_{aq} \tag{5}$$

$$Ca^{2+}_{(aq)} + CO^{2-}_{3(aq)} \xleftrightarrow{\text{precipitation}} CaCO_{3(s)} \tag{6}$$

Table 2. Stoichiometry of enzyme combinations.

Jack Bean Solution	**Ingredient-1 Urea (CH_4N_2O) (M)**	**Ingredient-2 Calcium Chloride ($CaCl_2$ $2H_2O$) (M)**	**Ingredient-3 Urease Enzyme (Jack Bean) (g/L)**	**Ingredient-4 Non-Fat Milk Powder (g/L)**
J1	1	0.67	3	0
J2	1	0.67	3	4
J3	0.37	0.25	0.85	4
Bio-enhancer Solution	Ingredient-1 Bio-enhancer-based Urea (CH_4N_2O) (in 1:10 u/w ratio)	Ingredient-2 Calcium Chloride ($CaCl_2$ $2H_2O$) (M)	Ingredient-3 Urease enzyme (Bio-enhancer) (g/L)	Ingredient-4 Non-fat milk powder (g/L)
B1	1:10	0.67	2	0
B2	1:10	0.67	3	0
B3	1:10	0.25	0.85	0
Soya Bean Solution	Ingredient-1 Urea (CH_4N_2O) (M)	Ingredient-2 Calcium Chloride ($CaCl_2$ $2H_2O$) (M)	Ingredient-3 Urease enzyme (Soya bean) (g/L)	Ingredient-4 Non-fat milk powder (g/L)
S1	1	0.67	2	0
S2	1	0.67	3	0
S3	0.37	0.25	0.85	0

2.3. Lime

The commercially available hydrated lime was adopted for this study. The presence of $Ca(OH)_2$ in the material was confirmed by the results of XRD analysis. The peaks of the hydrated calcium hydroxide were observed in the XRD analysis of lime, as shown in Figure 1.

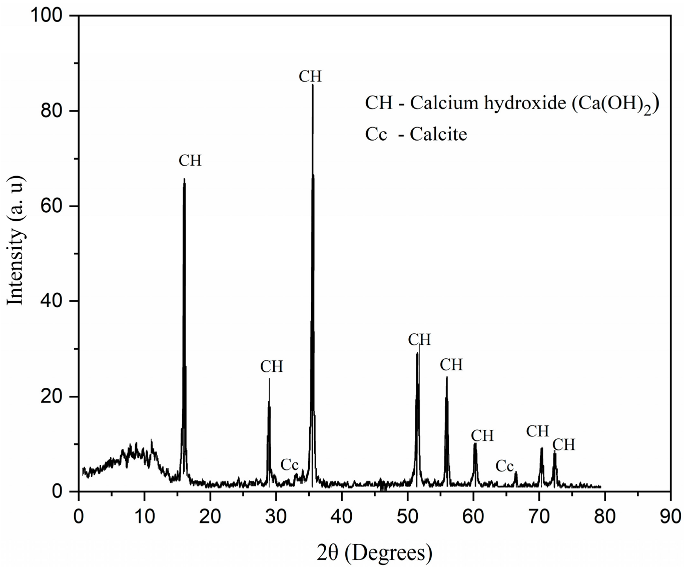

Figure 1. XRD of hydrated lime showing the peaks of calcium hydroxide.

3. Experimental Methodology

3.1. Sample Preparation

The urease solutions were prepared using different urease enzymes, including jack bean and soya bean mixed with urea, bio-enhancer, non-fat milk powder, and calcium chloride as shown in Figure 2. The possible set of combinations with their concentrations are mentioned in Table 2. The urease solutions were injected into the two soils individually. Adding the urease solution using injections can achieve uniform distribution and deeper penetration into the soil. Lime was added to the two soils at 3%, 6%, 9%, and 12% (W/W) dosages. W/W indicates weight/weight. For example, 3% W/W represents 3 g of lime for 100 g of dried soil. The compacted cylindrical samples of 38mm diameter and 76mm height were sealed in air-tight polyethylene bags and cured for a period of 0, 7, 14, and 21 days. UCS tests were conducted on the prepared specimens following ASTM D2166/D2166M-16 [36]. Furthermore, specimens of the specified size were prepared to evaluate the soil swell pressure using the oedometer cell.

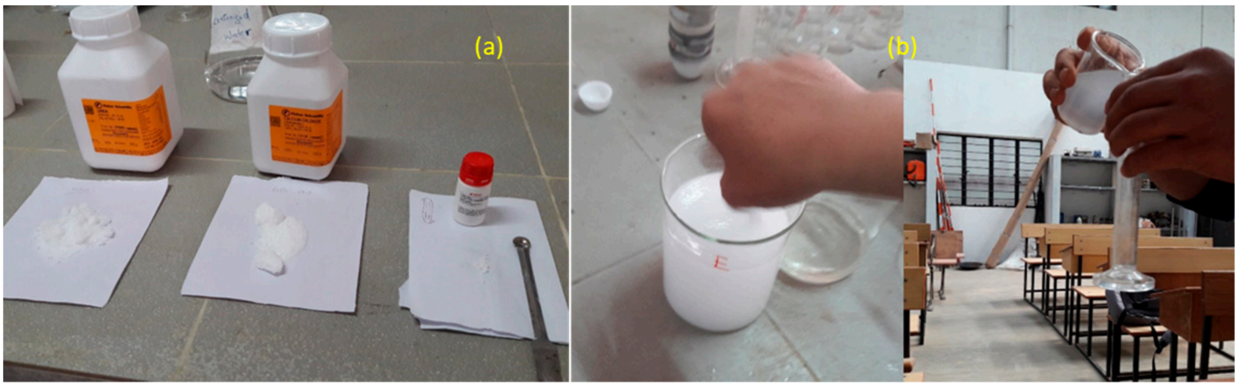

Figure 2. (**a**) Urea, calcium chloride, and urease enzyme. (**b**) Urease solution preparation.

3.2. Characterization of Treated Soil

Physical properties of the soils treated with urease solutions and lime at different proportions were determined. In accordance with ASTM D4318-17e1 [34], the treated soils were tested to determine the liquid and plastic limits, respectively. The optimum moisture content (OMC) and maximum dry density (MDD) of the treated soils were determined in accordance with ASTM D698-12 [35].

3.3. Unconfined Compressive Strength Test

Unconfined compressive strength (UCS) of soils, treated with urease and lime methods, was determined in accordance with ASTM D2166/D2166M-16 [36]. Cylindrical samples were compacted at their MDD and OMC and tested under a universal testing machine. A set of triplicate samples was prepared for each combination, with UCS determination based on the average peak stress, ensuring the variations remained below 10%. Testing was carried out at the end of each specified curing period, maintaining a controlled loading rate of 1.0 mm/min. The test was terminated once the specimen offered the highest resistance to the load.

3.4. One-Dimensional Consolidation (Oedometer Test)

According to the ASTM D4546-21 [37], the swell pressure was determined using an oedometer. The samples cured at different periods were placed in the oedometer cell by maintaining the maximum dry density. These specimens were subjected to an initial seating load of 7 kPa and flooded with water to swell. The swell readings were recorded until they reached the equilibrium. The incremental loading was applied with each increment for 24 h till the specimen reached its initial void ratio. The swell pressure was calculated from the deformations noted during the oedometer test.

4. Results and Discussion

4.1. Plasticity Characteristics

4.1.1. EICP Treated

The soil was treated with three urease enzyme solutions, and the plasticity characteristics were determined at different curing periods, as depicted in Figure 3. Initially, the liquid limit of soil 1 was recorded as 30%. A reduction in the liquid limit was observed upon treatment with the enzyme solutions, with the highest reduction achieved for the J2-treated soil. Specifically, when the soil was treated with the jack bean urease solution (J1 and J2), the liquid limit decreased to 21% and 19% at the 21-day curing period, respectively. However, the liquid limit increased when the soil was treated with the J3 solution, attributed to the absence of non-fat milk powder in the enzyme solution.

The formation of $CaCO_3$ precipitation in the pores of the treated soils results in Ca^+ ions replacing the ions on the soil surface, facilitating the reduction of the diffuse double layer. Consequently, the liquid limit can decline due to increased ionic concentration and diffuse double-layer compression. In bio-enhancer-treated soil, a significant reduction in the liquid limit was achieved with B2, followed by B1. Conversely, the B3-treated soil exhibited less variation in the liquid limit, possibly due to the lower urease enzyme concentration of 0.85 g/L, resulting in the formation of less precipitation.

Likewise, in soil 2, a significant reduction in the liquid limit was observed in the J2-treated soil as shown in Figure 4. The least improvement in the plasticity characteristics in soil 2 was found in the soya bean-treated soil with a lower enzyme concentration of 0.85 g/L. However, S2 and S3-treated soils exhibited relatively better liquid limit values after a 21-day curing period. When comparing soils 1 and 2, the trend diverged beyond the 14-day curing period due to variations in the urease rate in these two distinct mineralogical soils [38].

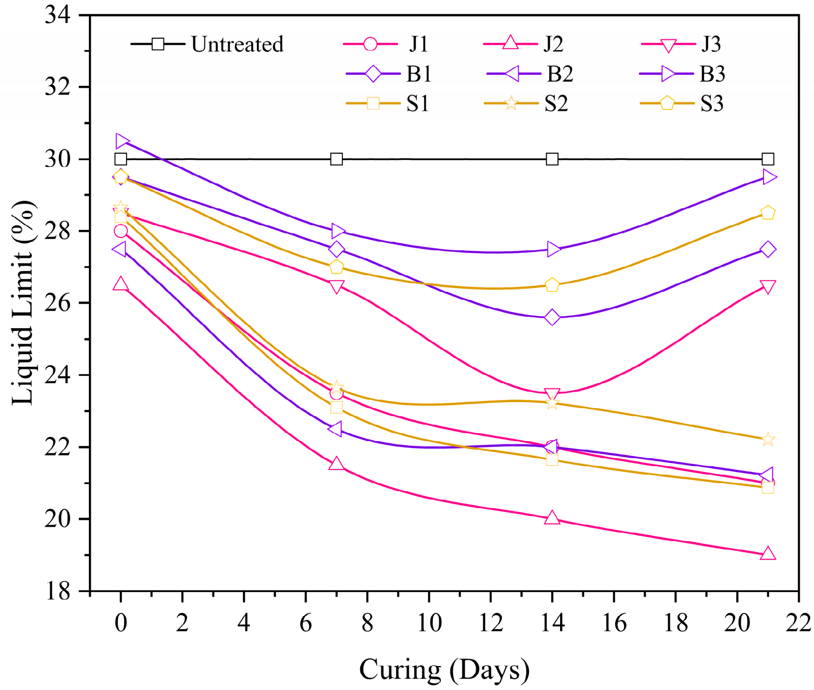

Figure 3. Liquid limit variation of soil 1 treated with enzyme solutions.

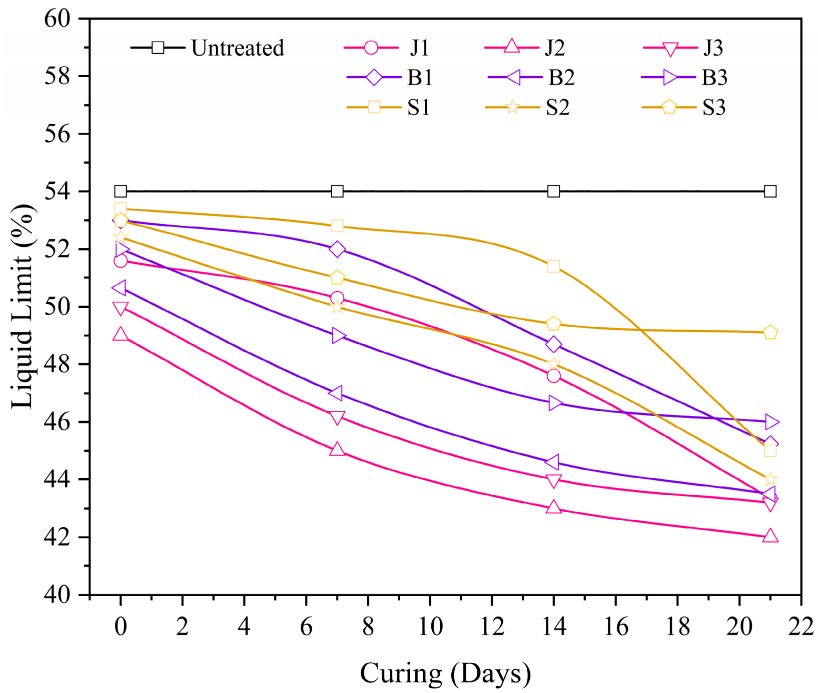

Figure 4. Liquid limit variation of soil 2 treated with enzyme solutions.

4.1.2. Lime Treated

The plasticity of the soil is predominantly linked to the liquid limit. The liquid limit for untreated and lime-treated soils was determined at different curing periods, as shown in Figure 5. In the case of soil 1, adding lime up to 3% resulted in a reduction in the liquid limit, followed by a subsequent 40% increase at 6% lime content. Beyond this point, the liquid limit remained constant, with up to 12% lime content. This decline in liquid limit is attributed to the reduction in diffuse double layer bounded on the clay particles, induced by the release of Ca^{+} ions from lime into the soil water [39]. This phenomenon elevates ionic concentration, thereby decreasing the diffuse double layer and subsequently reducing the soil's water-holding capacity. The increased liquid limit values at higher lime contents and long curing period of 21 days may indicate the formation of an increased quantity of calcium silicate hydrate (CSH) gel, influenced by the presence of silica in the red earth [40]. Notably, no significant variations in liquid limit were observed beyond 6% lime content.

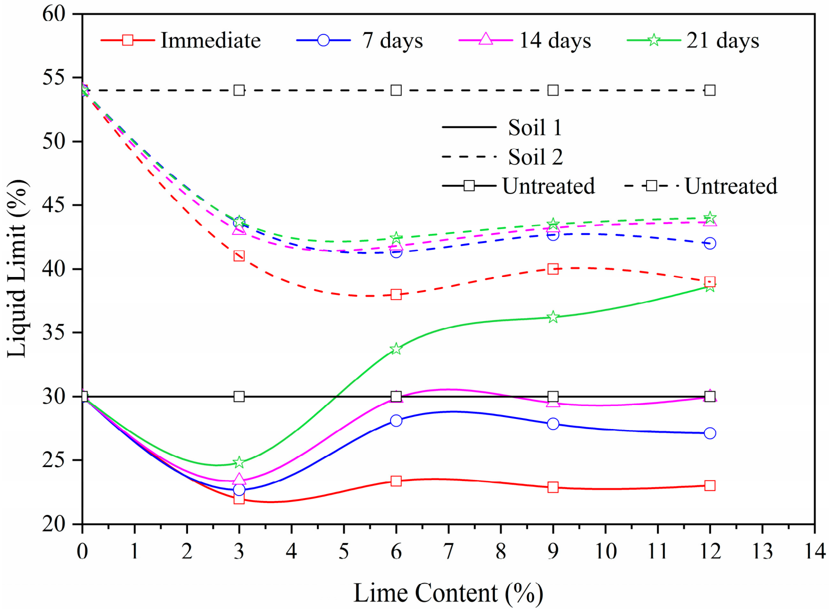

Figure 5. Liquid limit variation with lime content at different curing periods for soil 1 and soil 2.

In the context of soil 2, a decline in liquid limit was observed with lime addition up to 6%, maintaining a consistent trend with soil 1. This trend persisted across various curing periods at different lime dosages. The reduction in liquid limit is attributed to cation exchange capacity, which adsorbs calcium ions onto the soil surface from the lime-dissolved soil water. Due to the complete saturation of the calcium, the liquid limit might remain approximately constant at 9 and 12% lime content. Additionally, liquid limit values increased for both soils over curing periods, potentially owing to long-term physicochemical reactions between lime and the soil. One possible mechanism for this behavior is pozzolanic activity, which may lock water within the soil, contributing to an elevation in the liquid limit.

4.2. Effect on Unconfined Compression Strength

4.2.1. EICP Treated

UCS tests were conducted on soil samples treated with enzymes following the guidelines of ASTM D2166/D2166M-16 [36]. These tests aimed to provide insights into the impact of Enzyme Solutions on the strength properties of soils. Figures 6 and 7 present the UCS results for soil 1 and 2 samples, respectively, treated with enzyme solutions and cured for 7, 14, and 21 days. The observations revealed that the UCS values of the soil samples reached their maximum levels when treated with J2 and cured for 21 days. Non-fat milk powder in J2 played a key role as the primary strength enhancer, which improved the strength to 465 kPa by creating nucleation sites in the soil, thereby improving the precipitation of $CaCO_3$. However, non-fat milk powder was also present in J3, where the strength gain was less than that of J2 due to the lower amount of urease enzyme used in J3 (0.85 g/L compared to J2) [41].

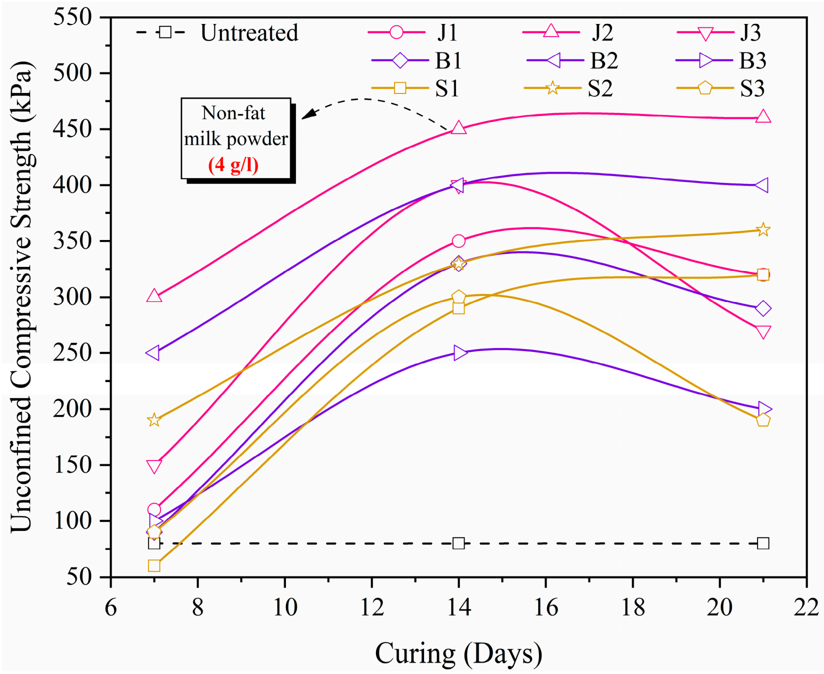

Figure 6. UCS values for red soil treated with enzyme solutions.

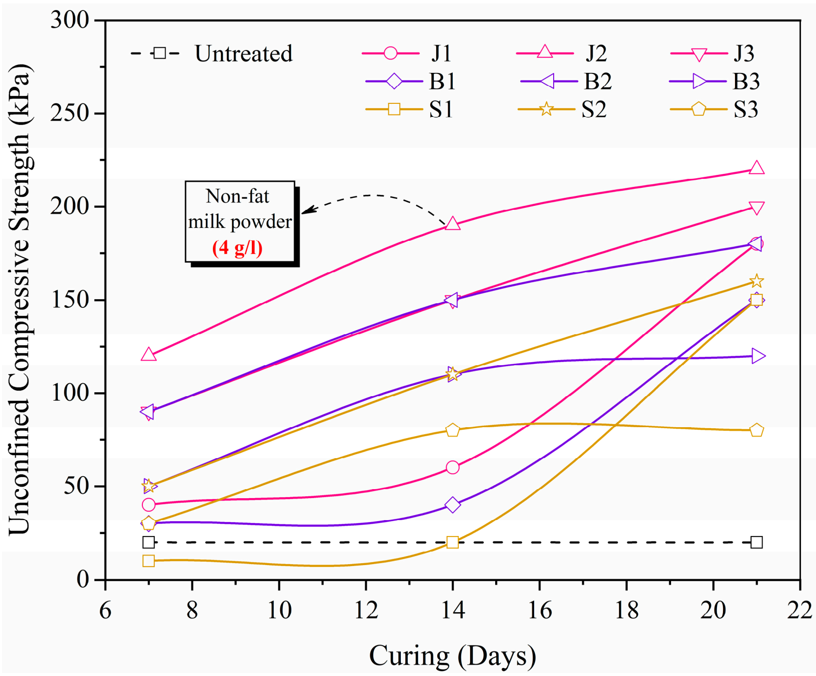

Figure 7. UCS values for black cotton soil treated with enzyme solutions.

It was observed that J2 depicted better performance in improving the strength characteristics when UCS tests were performed, and a gain in strength for soil 1 (Figure 6) and for soil 2 (Figure 7) can be observed. The increase in strength of the UCS value for soil 1 treated with J2 and cured for 14 days when compared to raw soil 1 (Figure 8) was found to increase from 70 kPa to 440 kPa. Moreover, the soil 2 sample, when treated with J2 and cured for 21 days, exhibited a notable increase in UCS value to 220 kPa compared to the raw soil 2, which had a UCS value of 13 kPa. The SEM images of raw and EICP-treated soils revealed the formation of calcite precipitation after 21 days of the curing period, as shown in Figure 9. Urea plays a crucial role in enhancing urease activity in the soil until it reaches a saturation point for the enzyme [42]. Additionally, as depicted in Figure 10, the incorporation of non-fat milk powder was observed to enhance the UCS values by 8.45 and 11 times for soils 1 and 2, respectively, by fostering nucleation sites in the soil mass, leading to the precipitation of $CaCO_3$ [13].

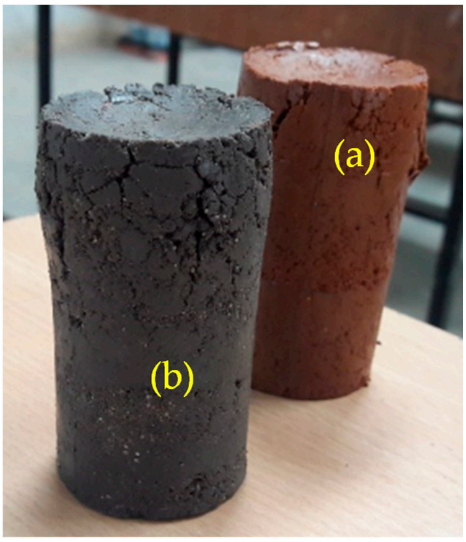

Figure 8. Failure patterns observed in the UCS tested: (**a**) red soil and (**b**) black cotton soil.

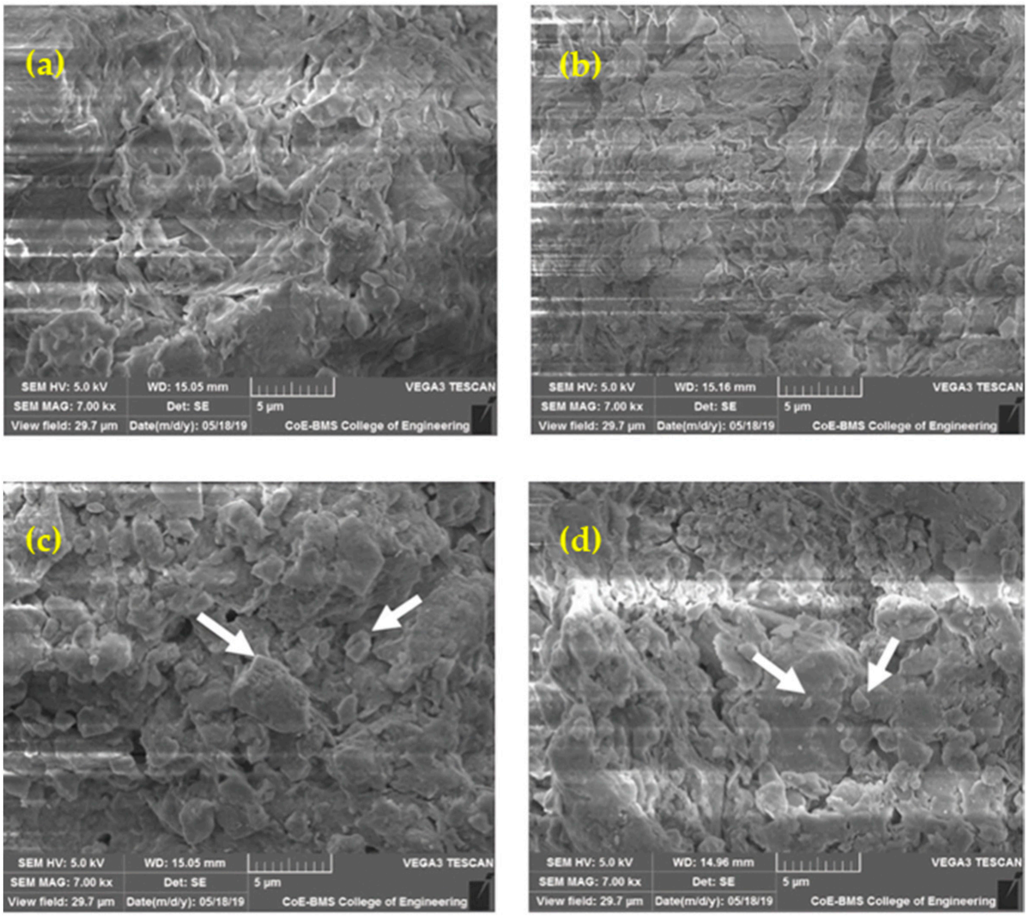

Figure 9. SEM images of (**a**) raw black cotton soil, (**b**) 21-day cured EICP-treated black cotton soil, (**c**) raw red soil, and (**d**) 21-day cured EICP-treated red soil.

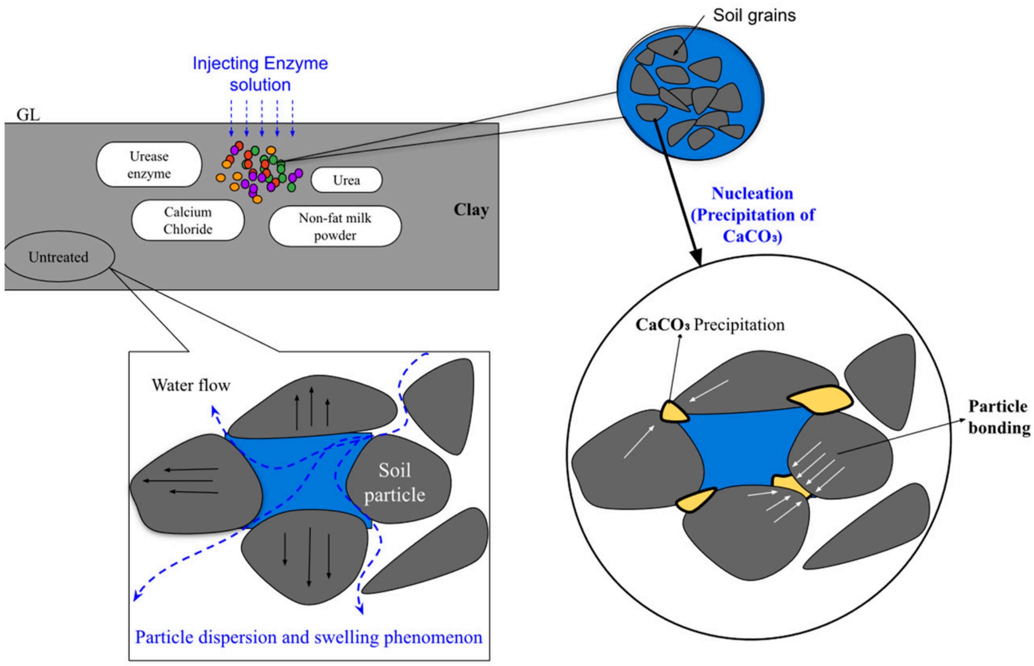

Figure 10. Schematic representation of EICP phenomenon in the soil pores.

In the bio-enhancer, it was found that the urease enzyme of 2, 3, and 0.85 g/L were maintained in preparing B1, B2, and B3 solutions, respectively. The presence of natural urea from the bio-enhancer was facilitated to understand its efficacy in increasing the strength of UCS samples. When soil 1 was treated with a B2 combination, the UCS value was increased from 55 to 400 kPa at the 14-day curing period, as shown in Figure 6. Moreover, all combinations of the B series were observed to have high UCS values at a curing period of 14 days. However, this increased strength was not more than the values reported with J series samples. Among all combinations in the B series, B2 has recorded high strength, followed by B1 and B3. It may be noted that B2 samples have given good increased results in strength due to higher concentrations of urease enzyme, whereas B1 and B3 had urease concentrations of 2 and 0.85 g/L, respectively. Similarly, as observed in Figure 8, the same trend was observed in soil 2, i.e., B2 > B3 > B1. However, the UCS values of soil 2 were increased beyond the 14-day curing period, unlike the trend observed in soil 1. This reverse trend might be due to the rate of urease activity being different in two different mineralogical soils [38].

It shows that a higher concentration of urease enzyme is a prime factor in strength gains. The same is the case with soya combinations. Even here, the S2 combination gained maximum strength, followed by S1 and S3. The order of strength gains is J2 > B2 > S2 (450 > 400 > 330 kPa); urea hydrolysis in soils occurs rapidly. Hence, there was a strength gain of 400 and 325 kPa in the short term for bio-enhancer and soya bean-based urease as the enzyme increased the penetration resistance in soil interstices. However, it is active only during the short term, which leads to degradation due to natural erosion in the long run. Therefore, it is recommended to conduct these experiments to understand their long-term behavior. The order of strength gains for both soil combinations treated with enzyme solutions is found to be enzyme-treated soil 1 > enzyme-treated soil 2 and is attributed to the difference in the mineralogical compositions (i.e., kaolinite in soil 1 and montmorillonite in soil 2).

4.2.2. Lime Treated

The UCS of soils 1 and 2 treated with lime at 3, 6, 9, and 12% dosages was determined. The initial strength of soil 1 was 80 kPa, and it was increased to 265 kPa immediately after adding 3% lime. The highest strength was observed as 385 kPa at 9% dosage, as depicted in Figure 11. The ionic concentration increased due to the transfer of Ca^+ ions from lime to the soil surface resulted in the thinning of diffuse double layer, followed by the reduction of the liquid limit. This phenomenon might be responsible for the strength improvement at zero-day curing. Gradually, the strength was increased with the curing period. The higher value of 420 kPa was achieved at 9% dosage when cured for 21 days. This improvement is attributed to the pozzolanic activity that forms the cementitious gel when calcium hydroxide reacts with the silica and alumina present in the soil. The strength in soil 1 was improved by 425% with the lime addition of 9%. However, the decline in the strength was shown after 9% lime. The particles of hydrated lime are finer and do not possess any considerable cohesion and friction. In higher quantities, lime can act as a lubricant that reduces the bonding between the particles [43]. Comparing this technique with EICP, soil 1 achieved relatively better strength with the treatment of urease solution. It can be noted that kaolinitic soil can be treated best with the EICP technique rather than lime.

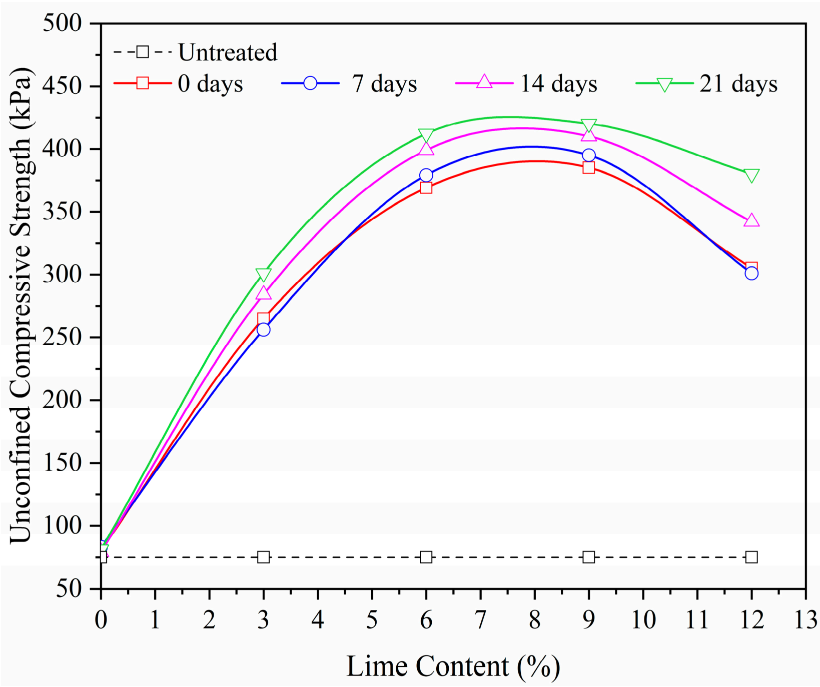

Figure 11. Strength variation in lime-treated red soil at different curing periods.

Similarly, soil 2 was treated with lime to determine its behavior on strength characteristics. The UCS of the untreated soil was observed as 20 kPa. As depicted in Figure 12, a drastic strength improvement of 130 kPa was observed at 3% lime, and later, the highest value was observed as 300 kPa at 9% lime content. As mentioned earlier, this initial improvement might be due to the drop in the liquid limit, resulting in the moisture reduction in the treated soil. The curing for an extended period increased the UCS values due to the calcium aluminosilicate (CAS) gel, which binds the soil particles [44]. Strength was approximately reduced by 35% with the lime addition of 12%. This can be due to the increased gel formation that increases pores, forming the porous matrix. The EICP treatment with the J2 combination achieved the highest strength of 250 kPa after curing for 21 days. Meanwhile, the soil treated with lime at 9% reached a UCS value of 340 kPa after 21 days of curing. Hence, it is worth noting that lime treatment is more effective in increasing the strength in montmorillonite soils.

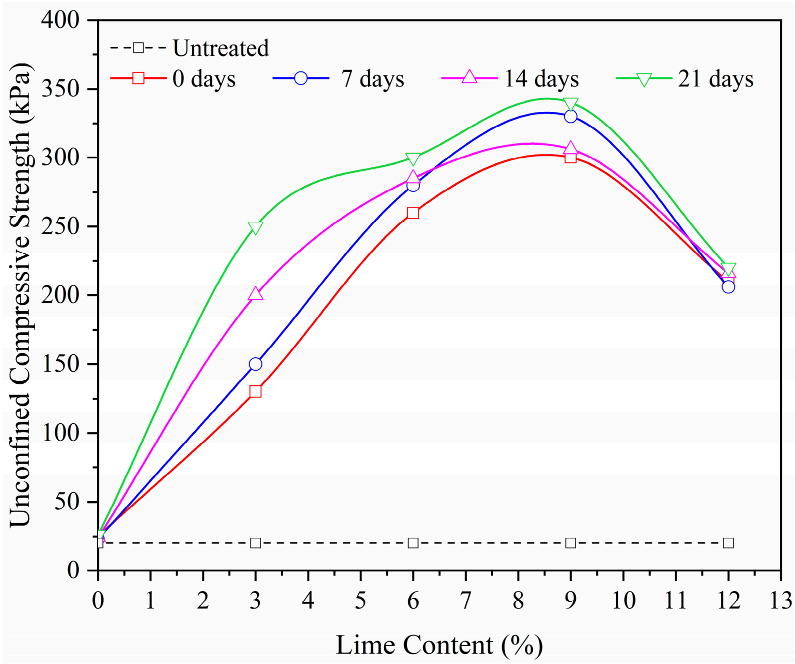

Figure 12. Strength variation in lime-treated black soil at different curing periods.

4.3. Soil Swell Behaviour

4.3.1. EICP Treated

Untreated soils and soils injected with enzyme solutions were tested for their swell behavior by performing a soil swell test (using an oedometer) as per ASTM D4546-21 [37]. Figures 13 and 14 depict the swell pressure of controlled and EICP-treated samples obtained from the present study. It was found that with the treatment of enzyme solutions and the specimen aging effect, the swell pressure values were dropped. The effectiveness of J2 is notable in both soils, demonstrating superior results. The initial swell pressure for untreated soil 1 was 119.83 kPa, and for untreated soil 2, it was 169.1 kPa. After enzyme treatment, the swell pressure significantly decreased to 37.68 kPa for soil 1 and 47.2 kPa for soil 2, particularly when treated with J2 and cured for 21 days. Similarly, bio-enhancer- and soya bean-amended soil samples also reduced the swell pressure. The order of reduction in swell pressure was J2 < B2 < S2, which strongly agreed with the previous studies [45]. The management of swell characteristics in expansive soils can be obtained through the precipitation of $CaCO_3$ [46]. This enhancement in swell characteristics may result from the adhesion of soil grains to each other following $CaCO_3$ precipitation. Moreover, the EICP technique, known for its effectiveness in improving UCS values, indicates that externally applied loads become less effective on the soil due to $CaCO_3$ precipitates. This same mechanism is attributed to the reduction in swell pressure in the soil, where capillary water appears to detach the soil grains [41].

A notable decrease in swell pressure, from 120 to 50 kPa, was observed in soil 1 upon treatment with the jack bean urease solution (J2) after a 21-day curing period. Likewise, soil 2 exhibited the highest reduction in swell pressure, dropping from 175 to 55 kPa when treated with the J2 solution. This decrease in swell pressure indicates enhanced resistance to volume change when the soil comes into contact with water. It is likely attributable to the formation of calcite precipitation, which fosters inter-particle bonding, subsequently mitigating the pressure exerted during soil swelling [47]. Moreover, the soil voids filled with generated calcite precipitation can enhance resistance against swelling [48]. Even if $CaCO_3$ becomes detached from soil grains due to excess loads, it still provides a cushioning effect to adjacent soil grains, forming an effective chain of forces in the soil mass that transfers loads to the nearest hard surface [49]. Therefore, the adhesion of soil grains by precipitated $CaCO_3$ contributes to the reduction of swell pressure in the soil.

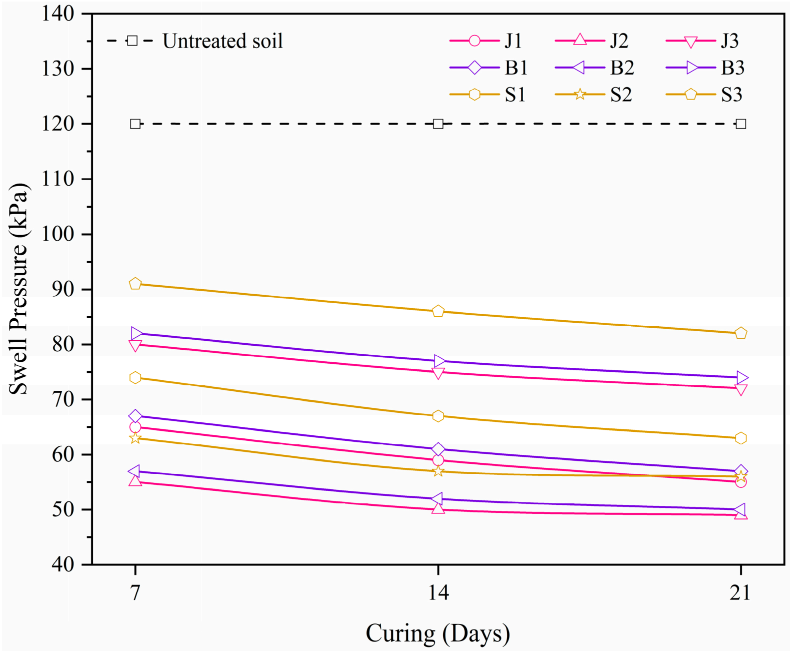

Figure 13. Swell test results of red soil treated with enzyme solutions.

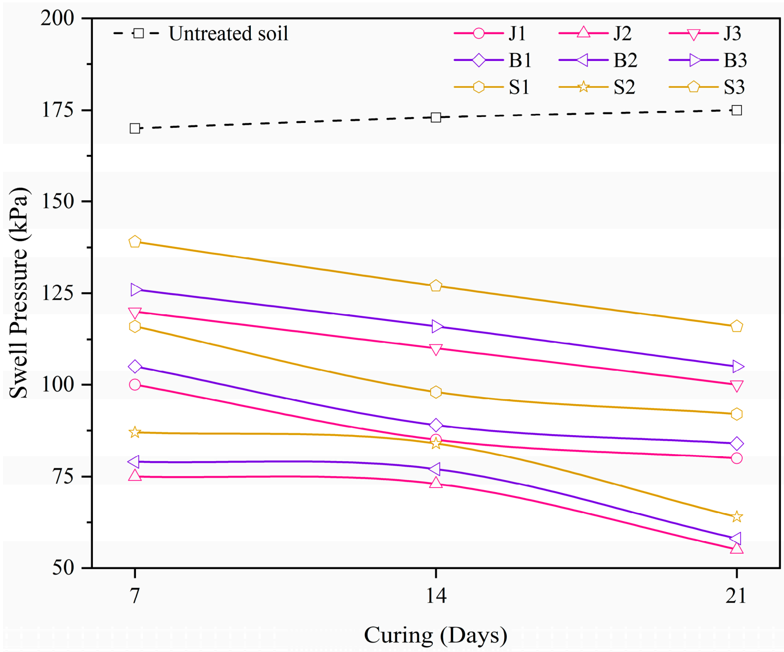

Figure 14. Swell test results of black cotton soil treated with enzyme solutions.

4.3.2. Lime Treated

Using an oedometer, the change in the soil volume was determined to study the swell pressure. It is the pressure at which the soil's volume can be restrained from swelling. The swell pressure of the untreated soil 1 was observed as 119.83 kPa, and it had reduced gradually to 88, 76, 65, and 64 kPa when lime content of 3, 6, 9, and 12% was added to the soil 1 after curing for 21 days as shown in Figure 15. This decline in the swell pressure represents its resistance to the volume change when soil gets in contact with water. As shown in Figure 15, the swell pressure reduced its value until the soil was added with lime up to 9%. The soil's swell pressure remained the same, with up to 12% lime content. The decreased swell pressure is associated with the reduced diffuse double layer arising from the increased ionic concentration at higher lime content. Moreover, the soil mitigated more swell with an increased curing period. This phenomenon can be attributed to the enhancement of soil bonding through the pozzolanic reaction, which develops gradually over extended periods.

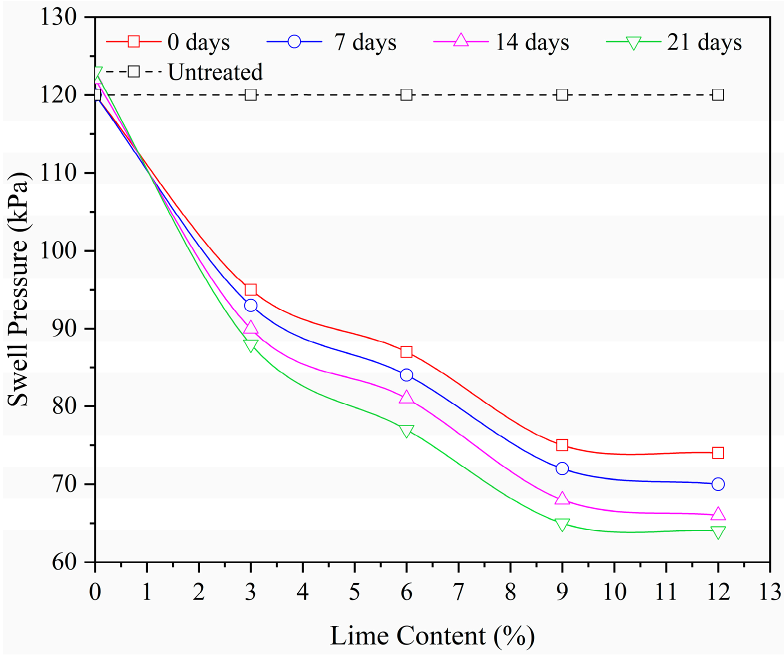

Figure 15. Effect of lime dosage on the swell pressure of red soil.

Figure 16 represents the swell pressure for soil 2 when lime was added at different dosages. The untreated soil 2 exhibited a swell pressure of approximately 170 kPa at different curing periods. The trend in reduction of swell pressure was observed to be similar to soil 1. The exchange of the cations on the clay surface can change the soil mineralogy, which reduces its affinity to adsorb water and swell [50]. This phenomenon can make the expansive soil more stable against swelling behavior. Here, the lime particles, which were replaced with expansive soil particles, reduced the swelling nature. An increase in lime content within the soil promotes enhanced cementation among soil particles, resulting in a reduction in swell pressure. Following a 21-day curing period, soil 1 experienced a notable 47% reduction in swell pressure upon the addition of 12% lime. Likewise, soil 2 showed a more substantial reduction, with a 70% decrease in swell pressure following the inclusion of 12% lime during the identical curing period.

EICP treatment with jack bean urease solution significantly reduced swell pressure in both soil 1 (from 119.83 kPa to 47.2 kPa) and soil 2 (from 169.1 kPa to 55 kPa). Among various urease solutions, J2 demonstrated notable effectiveness, decreasing swell pressure by 59% and 67.5% in soils 1 and 2, respectively. Compared with lime treatment, it exhibited a significant swell reduction in soil 1. Conversely, a more notable swell reduction was observed in soil 2 with lime treatment.

This study serves as a preliminary guide, showcasing the marginal improvements in strength and swell characteristics for two distinct soils. It highlights the necessity for further research to investigate the long-term stability and effectiveness of the treatments. The results discussed in the study are based on short curing periods of up to 21 days. It is advisable to interpret these findings with caution when considering the application of these techniques under field conditions.

Future research should consider extending the curing periods to assess the long-term effectiveness of these treatment methods. Additionally, studying with different concentrations of enzyme solutions and incorporating a variety of soil types would provide a better understanding of the factors influencing the durability of these treatments over time.

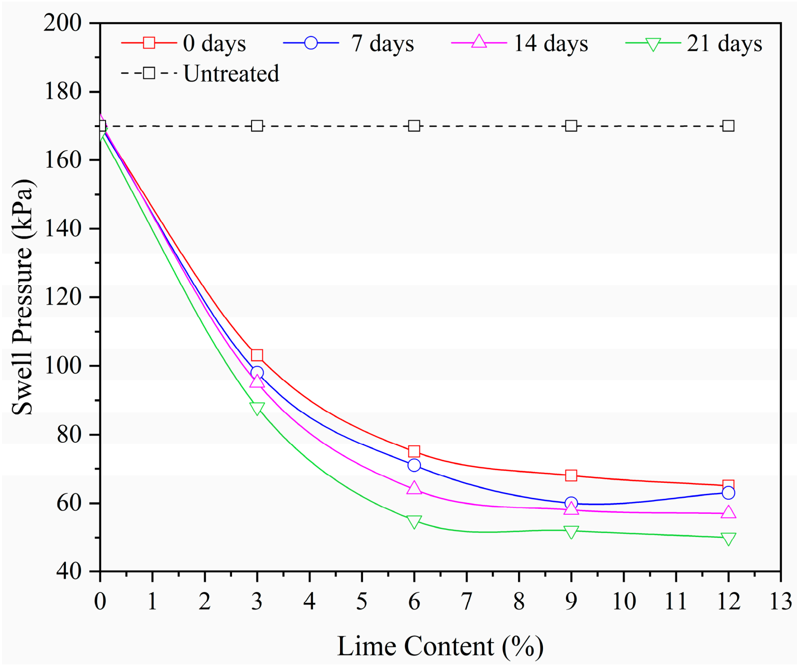

Figure 16. Effect of lime dosage on the swell pressure of black soil.

4.4. Life Cycle Assessment (LCA)

LCA studies were conducted on all the enzyme-treated samples, and due to brevity, data from literature have also been used. It has been reported by [19] that EICP in soil stabilization has higher acidification and eutrophication potential due to the use of urea and the formation of ammonia and nitrogen. Under global warming potential (GWP), JICP-based CO_2 contribution stands at 175 tons CO_2 eq, followed by methane with 20 tons CO_2 eq and nitrogen oxides at 50 tons CO_2 eq. Similarly, a bio-enhancer is found to have a GWP at 7.6 tons CO_2 eq and urea at 217 tons CO_2 eq. Also, applying soya bean as a substitute to urease enzyme has a GWP of 6.44 Kg CO_2 eq. By comparing all three prospective enzyme solutions, it can be inferred that all of them would perform well due to various means of calculating the LCA in some respects, such as GWP. Still, based explicitly on acidification and eutrophication potential, possibly sustainable material may have a higher value. Hence, to conclude, the LCA of each material has to be based on site-specific conditions, and the type of process involved in producing it must be considered. By comparison, it can be observed that for this type of soil stabilization, soya bean is a potentially sustainable material compared to jack bean urease and bio-enhancers.

5. Conclusions

In this study, the use of non-conventional enzyme solutions was adopted as a soil amendment for strength enhancement and reduction in swell pressure. These tests were even followed by a life cycle analysis to obtain a potentially sustainable enzyme mixture. Furthermore, a conventional material, lime, was also used as an admixture to enhance the engineering properties of the two different soils. The following are the outcomes from this study that could help in choosing the right combination of enzyme solutions for soils exhibiting different plasticity.

- Soil 1 achieved the relatively highest UCS value at 450 kPa with jack bean-based urease solution compared to the lime-treated soil. Meanwhile, the lime treatment resulted in a better UCS value of 340 kPa in soil 2 compared to the enzyme treatment.
- The swell pressures were controlled and reduced by approximately 60 and 67.5% for soils 1 and 2, respectively, when subjected to EICP treatment, particularly using J2 solution. Consequently, lime-treated soils 1 and 2 exhibited a reduction in swell pressure values by 47 and 70% following 21 days of curing period, respectively.

- Jack bean-based urease enzyme and low-fat milk powder mixture resulted in higher UCS values. However, a bio-enhancer with jack bean urease enzyme, which acted as a substitute for urea, resulted in relatively better UCS values than the soya bean-based urease treatment. Still, its efficacy was found to under-perform compared to the jack bean-based urease.
- Therefore, it is worth noting that the urease enzyme injection into the kaolinite soil has shown better improvement, whereas lime treatment is an appropriate method for enhancing montmorillonite-based soil.
- The study has revealed that soya bean acts as a sustainable urease enzyme producer with less GWP than jack bean urease and bio-enhancer.

This study can assist in the selection of suitable methods, whether biochemical or chemical, for soil stabilization. Bioinspired projects, engaging both engineers and biologists in large-scale field applications, have the potential to overcome challenges such as the production of urease solutions, maintaining a stable environment for enzymes, understanding soil–enzyme interactions, and addressing other factors on a larger scale. Hence, acquiring essential knowledge and understanding the fundamentals and mechanisms associated with geochemistry and biochemistry is imperative.

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