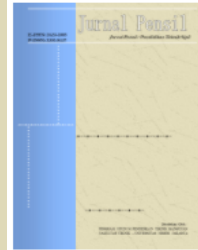


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IMPROVING THE ELASTIC MODULUS OF CLAY USING BIOSTIMULATED MICROBIALY INDUCED CALCITE PRECIPITATION (MICP)

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Abstract

Low-plasticity clay (CL) exhibits low stiffness and is easily deformable, thus requiring an efficient and environmentally friendly improvement method. Prior studies on Microbially Induced Calcite Precipitation (MICP) have predominantly emphasized improvements in soil compressive strength, whereas investigations addressing its effects on soil stiffness and elastic modulus remain relatively limited. Accordingly, the present study aims to evaluate the effectiveness of biostimulated MICP in enhancing the elastic modulus parameters (E_0 and E_t) of low-plasticity clay (CL). To achieve this objective, laboratory tests were conducted on soil specimens under two conditions, namely untreated samples and samples treated with an MICP solution, with incubation durations of 7 and 14 days. The Unconfined Compression Test (UCS) was conducted to obtain stress-strain relationships, which were then analyzed to determine the elastic modulus values. The results show that at 7 days of incubation, the MICP-treated soil exhibited increases of approximately 92% in E_0 and 90% in E_t compared to the untreated condition. With extended incubation to 14 days, the improvements became more pronounced, with E_0 increasing by approximately 104% and E_t by about 125% relative to untreated soil. The ANOVA results indicated that the differences between the untreated specimens and those treated with MICP were statistically significant, as evidenced by p-values below the 0.05 significance threshold. These findings demonstrate that the biostimulation process effectively enhances soil stiffness through interparticle calcite formation, providing a foundation for developing more efficient and eco-friendly soil stabilization methods applicable to infrastructure projects in tropical regions.

Keywords: Microbially Induced Calcite Precipitation (MICP), Biostimulation, Clay Soil (CL), Elastic Modulus, Unconfined Compressive Strength (UCS)

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Introduction

Low-plasticity clay (CL) is a common type of subgrade soil found throughout Indonesia, generally dominated by kaolinite minerals as its primary constituent (Lehmann et al., 2021). This soil exhibits low stiffness, with an average elastic modulus ranging from 0.5 to 3 MPa, and is highly susceptible to deformation under loading, which can lead to settlement in the subgrade layer of structures (Obrzud & Truty, 2018). The low stiffness is primarily attributed to its microstructure, which is composed of fine particles smaller than 2 μm with weak interparticle bonding (Won et al., 2021). These characteristics result in a relatively low and unstable elastic modulus for CL soil, thereby necessitating an improvement method capable of enhancing soil stiffness without altering its natural characteristics.

Among various soil improvement methods, Microbially Induced Calcite Precipitation (MICP) is widely acknowledged for its effectiveness in increasing soil stiffness. This mechanism involves urease-producing bacteria that catalyze urea hydrolysis, leading to the production of carbonate (CO_3^{2-}) and ammonium (NH_4^+) ions. Subsequently, the carbonate ions react with calcium ions (Ca^{2+}), resulting in the precipitation of calcium carbonate (CaCO_3) within the soil matrix (DeJong et al., 2010). The precipitates serve to bond soil particles together, resulting in an increase in the elastic modulus (Song et al., 2022). In practice, MICP is typically implemented through two approaches: bioaugmentation and biostimulation (Kannan et al., 2020). The bioaugmentation approach involves the addition of external bacterial cultures; however, it presents challenges such as high cost, the need for controlled laboratory conditions, and limited effectiveness under field applications (Graddy et al., 2021). In contrast, the biostimulation approach relies on the activation of indigenous bacteria inherently existing within the soil by enhancing their metabolic activity through the supply of appropriate nutrient sources. (Fan et al., 2024). Biostimulation is considered more practical and cost-effective for field applications, as it eliminates the need for external bacterial inoculation while achieving ureolytic activity comparable to that of bioaugmentation (Rahman et al., 2020).

Numerous studies have demonstrated that biostimulated Microbially Induced Calcite Precipitation (MICP) effectively improves the unconfined compressive strength (UCS) of clayey soils, with the magnitude of strength gain being strongly influenced by the incubation or curing period. Tiwari and Satyam (2021) reported that in clayey soils, UCS increased progressively with incubation periods of 1–4 days, rising from values 96 kPa to 225 kPa at the longest incubation period, corresponding to an improvement of about 134%. Islam et al. (2019) demonstrated that biostimulated MICP produced substantial UCS enhancement in clayey soils under controlled testing conditions, with strength gains reaching up to approximately 300% after treatment durations of 7 and 14 days. Chittoori et al. (2021) further showed that UCS development was highly sensitive to incubation time, where UCS increased from 155.5 kPa at 0 days, and reached 232.8 kPa after 7 days, beyond which additional incubation did not lead to further strength improvement. Kannan et al. (2020) observed that the implementation of biostimulation-based MICP in clayey soils resulted in a measurable increase in unconfined compressive strength (UCS). Nevertheless, although existing studies consistently demonstrate the effectiveness of biostimulated MICP in enhancing UCS, the majority of these investigations predominantly emphasize peak stress responses, whereas stiffness-related parameters have received comparatively limited attention. In particular, the elastic modulus parameters E_0 , and E_t , which represent soil stiffness at the initial loading stage, and at the onset of plastic behavior, have not been systematically evaluated for low-plasticity clay (CL) treated using the biostimulation approach. As these parameters describe the evolution of soil stiffness across different loading stages and cannot be derived directly from UCS values, their assessment is necessary to improve the understanding of the mechanical response of biostimulated CL soils.

This research evaluates the effectiveness of biostimulated Microbially Induced Calcite Precipitation (MICP) in improving the elastic modulus of low-plasticity clay (CL). The analysis

focuses on elastic modulus parameters, namely the initial tangent modulus (E_0) and the tangent modulus (E_t), which collectively represent the soil's elastic response at different stages of loading (Gupta, 2000). The analysis was conducted through Unconfined Compression Tests (UCS) to obtain the relationship between load and deformation, which was then used to determine the modulus values. This research provides an evaluation of soil stiffness enhancement through biostimulation and offers a foundation for developing more efficient and environmentally friendly stabilization methods suitable for infrastructure projects in tropical regions.

Research Methods

This study utilized soil obtained from the Kadiri University area at a depth of approximately 80 cm to obtain undisturbed soil conditions (Chrysanthopoulos & Kallioras, 2025). The specimens were air-dried under ambient laboratory conditions for a period of two weeks, with exposure to direct sunlight avoided, until the moisture content decreased to approximately 10%, in order to maintain the viability of the indigenous bacterial community (Wang et al., 2024). The soil was then pulverized using a mechanical sieve with a 0.3 mm aperture to achieve particle uniformity (Consoli et al., 2012). Soil characterization was performed in accordance with the procedures outlined in ASTM D2487-17, ASTM D4318-17, and ASTM D7928 standards. According to the Unified Soil Classification System (USCS), the soil was identified as low-plasticity clay (CL), comprising 31,45% sand, 52,39% silt, and 16,17% clay. Results from Atterberg limit testing using the Casagrande apparatus indicated a liquid limit (LL) of 24,99%, a plastic limit (PL) of 10,93%, and a corresponding plasticity index (PI) of 14,06%.

The MICP solution was formulated from a mixture of 0.333 mol/L urea (N. Tiwari et al., 2021), 0.3 mol/L calcium chloride (CaCl_2), 0.1 mol/L sodium acetate (CH_3COONa), 0.001 mol/L sodium hydroxide (NaOH) (Islam et al., 2019), and 3 g/L sugarcane molasses as an organic carbon source (Pakbaz et al., 2020). All components were measured according to the specified concentrations, dissolved in distilled water, and stirred until a homogeneous solution was obtained. The prepared solution was used immediately after mixing to prevent any decrease in the chemical activity of its components (J. Huang et al., 2025; Kariminia et al., 2025).

Two treatments were used in the UCS testing: an untreated sample (using plain water) and a sample treated with the MICP solution (Arpajirakul et al., 2021). Cylindrical specimens with a diameter of 3.65 cm and a height of 7.4 cm were prepared in accordance with ASTM D2166, maintaining a wet unit weight of 1.61 g/cm³, while either water or MICP solution was added at a proportion equivalent to 40% of the soil mass. Paraffin paper was applied to the inner mold walls to prevent soil sticking, and the air pluviation technique was employed to ensure homogeneous particle distribution (Hariprasad et al., 2016). After the molds were filled, the MICP solution was added in the specified dosage and allowed to stand for approximately 20 minutes to ensure uniform absorption. The specimens were then removed from the molds, wrapped with plastic film, and incubated under controlled room-temperature conditions for 0, 7, and 14 days to prevent moisture loss, without direct exposure to sunlight (Chen et al., 2023; Purba et al., 2025). These incubation periods were selected to observe CaCO_3 formation, as ureolytic bacterial activity typically increases after 7 days and reaches its peak around 14 days (Pakbaz et al., 2020), while the 0-day incubation served as the initial control condition (Gowthaman et al., 2025).

Figure 1 illustrates the small-scale unconfined compressive strength (UCS) testing apparatus developed by AIC Innovation. The system consists of a stepper motor coupled with a planetary gearbox and ball screw mechanism to apply controlled axial displacement to the specimen. Axial load is measured using a calibrated load cell, while vertical deformation is continuously monitored by a linear potentiometer. All mechanical and sensor components are integrated with a digital output data logger to ensure synchronized and accurate acquisition of load–deformation data during testing.

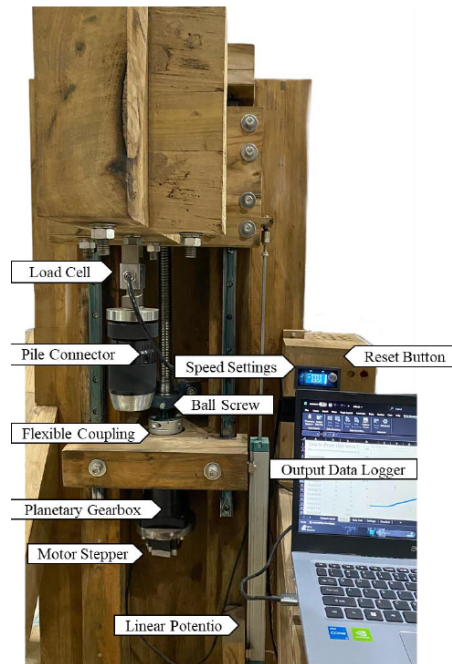


Figure 1. Loading Test Equipment (AIC-Innovation)

The unconfined compressive strength (UCS) test was performed using a small-scale testing apparatus developed by AIC Innovation, in which axial load was applied through a spherical bearing to reduce loading eccentricity. The test was conducted under displacement-controlled conditions at a loading rate of 0.005 mm/s, in accordance with ASTM D2166 (Safi’i et al., 2025). The test was terminated when the specimen reached failure, as indicated by the attainment of the maximum load and the subsequent loss of load-carrying capacity. Load data were recorded using a calibrated load cell, while vertical deformation was measured using a linear potentiometer automatically synchronized with Excel Data Streamer (Al-Dahiree et al., 2022; Ozcep, 2010; Solaiman et al., 2025). The measured load–deformation data were transformed into stress–strain relationships by determining the axial stress (σ) as the ratio of the applied load (P), to the specimen’s cross-sectional area (A) (Lv et al., 2025).

$$\sigma = \frac{P}{A} \quad (1)$$

Meanwhile, the axial strain (ϵ) was determined by dividing the axial deformation (ΔH) by the initial height of the specimen (H_0).

$$\epsilon = \frac{\Delta H}{H_0} \quad (2)$$

The elastic modulus was subsequently obtained from the stress–strain curves as the slope of the corresponding stress–strain relationship.

$$E = \frac{\sigma}{\epsilon} \quad (3)$$

The stress–strain curves obtained were further utilized to assess soil stiffness parameters, including E_0 and E_t , at various stages of loading (Urmi et al., 2023). The values of E_0 were obtained from the initial slope of the curve (H. Huang et al., 2018), and E_t at the point where the soil began to exhibit plastic behavior (H. Huang et al., 2018). A one-way ANOVA was applied to compare the mean values of E_0 and E_t between the untreated and MICP-treated specimens at each incubation period. Statistical significance was evaluated using the p-value criterion at a 95% confidence level ($\alpha = 0.05$) (Thoriya et al., 2025).

Research Results and Discussion

Elastic Modulus of Untreated Clay at 7 Days Incubation

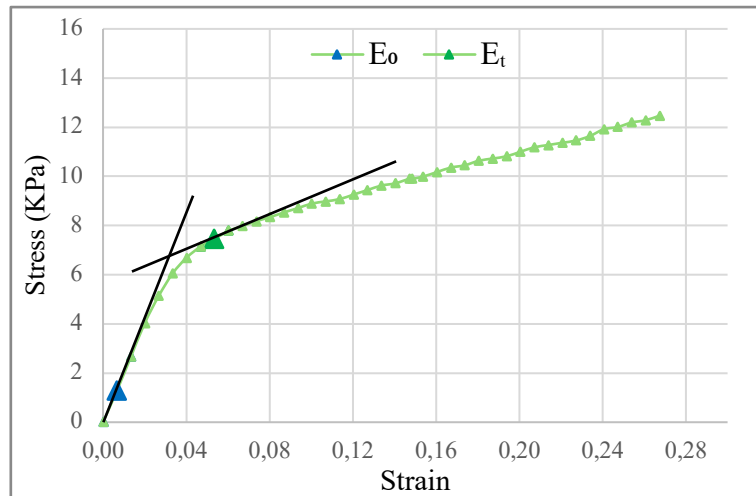


Figure 2. Stress–Strain Curve of Untreated Clay with 7-Day Incubation

Figure 2 illustrates the stress–strain relationship of untreated low-plasticity clay (CL) at a 7-day incubation period. The stress–strain curve shows that the increase in stress occurs more slowly than the increase in strain, indicating that the soil exhibits low stiffness and undergoes relatively large deformation during loading. The initial elastic modulus (E_0) of 192,07 kPa represents the soil stiffness at the early stage of loading. The tangent elastic modulus (E_t) of 139,47 kPa, which is lower than E_0 , indicates a reduction in stiffness as strain increases. This behavior suggests that the elastic limit is reached at a relatively small strain, followed by plastic deformation. The difference between E_0 and E_t indicates that the mechanical response of the CL soil is still governed by weak interparticle bonding and has not undergone structural strengthening. The 7-day incubation without biological treatment does not alter the fundamental elastic response of the soil. Therefore, this condition is used as a mechanical reference for comparing soil responses under different treatments and incubation periods.

Elastic Modulus of Untreated Clay at 14 Days Incubation

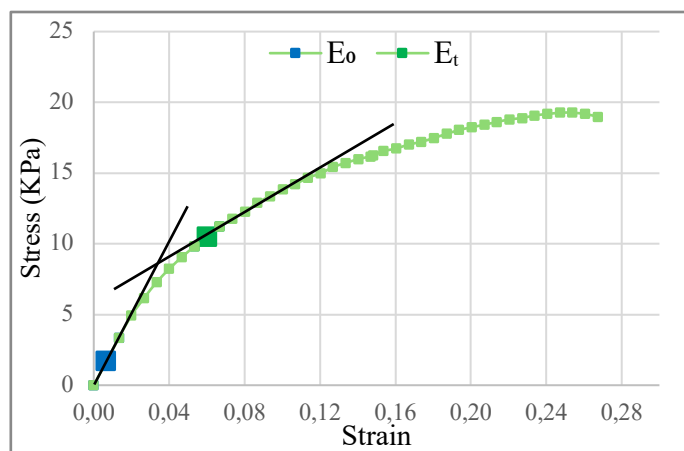


Figure 3. Stress–Strain Curve of Untreated Clay with 14-Day Incubation

Figure 3 illustrates the stress–strain relationship of untreated low-plasticity clay (CL) at a 14-

day incubation period. The stress–strain curve exhibits a more stable response compared to the 7-day incubation, although relatively large deformation is still observed during loading. The initial elastic modulus (E_0) is recorded as 251,81 kPa, representing the soil stiffness at the early stage of loading. The tangent elastic modulus (E_t) of 174,09 kPa, which is lower than E_0 , indicates a reduction in stiffness as strain increases and approaches the plastic condition. This behavior suggests that the elastic limit is still reached at a relatively small strain. The difference between E_0 and E_t indicates that the mechanical response of the soil is primarily governed by the natural structure of the CL soil, without any interparticle strengthening mechanism. Incubation for up to 14 days without biological treatment does not alter the fundamental stiffness characteristics of the soil. Therefore, this condition is used as a reference for evaluating the effect of biostimulated MICP at subsequent incubation stages.

Elastic Modulus of MICP-Treated Clay at 7 Days Incubation

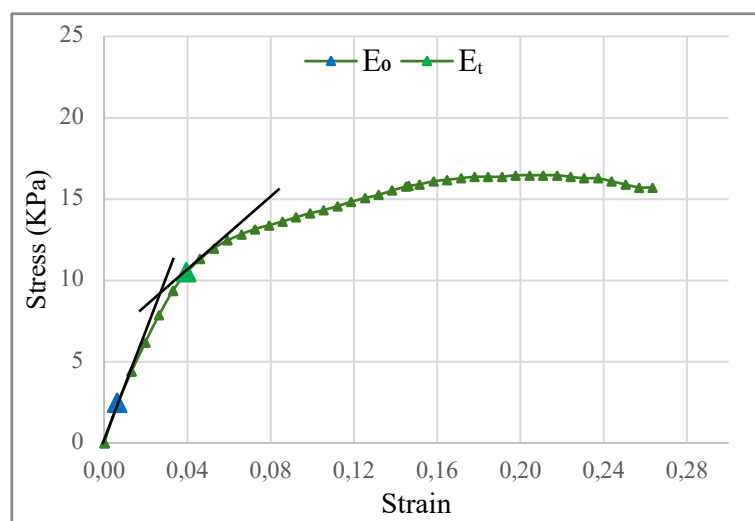


Figure 4. Stress–Strain Curve of MICP-Treated Clay with 7-Day Incubation

Figure 4 illustrates the stress–strain relationship of low-plasticity clay (CL) treated with biostimulated MICP at a 7-day incubation period. The stress–strain curve exhibits a stiffer response compared to the untreated soil, particularly at small to intermediate strain levels. The initial elastic modulus (E_0) of 369,08 kPa indicates improved soil stiffness at the early stage of loading. The tangent elastic modulus (E_t) of 264,98 kPa, which is lower than E_0 , indicates a reduction in stiffness as strain increases; however, the soil is still able to maintain an elastic response up to near the plastic condition. This behavior suggests that the transition toward plastic behavior occurs in a more controlled manner. These results indicate that at a 7-day incubation period, biostimulated MICP has begun to modify the soil structure through the initial formation of calcium carbonate (CaCO_3) precipitates that strengthen interparticle contacts. This observation is consistent with the findings reported by Chittoori et al. (2021), who observed that improvements in the mechanical response of clay soils can be detected at the early stages of MICP treatment, even though the cementation process has not yet fully developed.

Elastic Modulus of MICP-Treated Clay at 14 Days Incubation

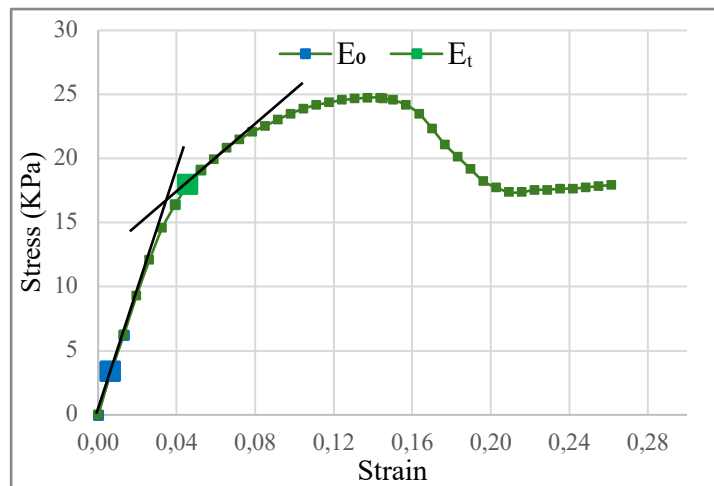


Figure 5. Stress–Strain Curve of MICP-Treated Clay with 14-Day Incubation

Figure 5 illustrates the stress–strain relationship of low-plasticity clay (CL) treated with biostimulated MICP at a 14-day incubation period. The stress–strain curve exhibits a stiffer and more stable response compared to the 7-day incubation, particularly during the early to intermediate stages of loading. The initial elastic modulus (E_0) of 513,59 kPa indicates high soil stiffness at the early stage of loading. The tangent elastic modulus (E_t) of 392 kPa, which is slightly lower than E_0 , indicates that soil stiffness can be maintained up to near the plastic condition. The relatively small difference between E_0 and E_t suggests a more stable elastic–plastic transition. This behavior indicates that at a 14-day incubation period, biostimulated MICP has produced a more consolidated soil structure through a more uniform precipitation of calcium carbonate (CaCO_3), thereby strengthening interparticle bonding and enhancing resistance to deformation. A similar response was reported by Islam et al. (2019), who showed that increasing the incubation duration of MICP-treated clay results in a more stable elastic response. The high values of E_0 and E_t indicate that structural strengthening has developed effectively. Therefore, the 14-day incubation period represents the most effective MICP condition for improving soil stiffness and the stability of the elastic response.

Comparison of E_0 at 7 and 14 Days Incubation

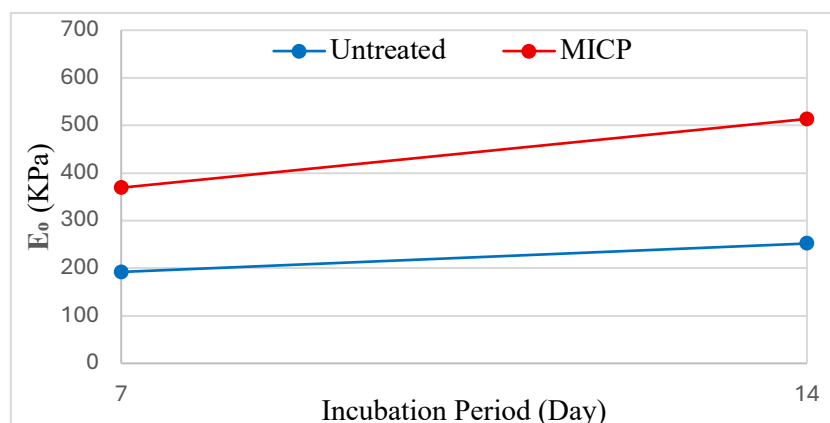


Figure 6. Elastic Modulus (E_0)

Figure 6 presents a comparison of the initial elastic modulus (E_0) between untreated soil and soil treated with biostimulated MICP at incubation periods of 7 and 14 days. The difference in E_0 values between the two conditions indicates the direct effect of MICP treatment on the initial stiffness of the soil. At the 7-day incubation period, the E_0 value of the untreated soil is 192,07 kPa, whereas the MICP-treated soil exhibits an E_0 value of 369,08 kPa, corresponding to an increase of approximately 92% relative to the untreated condition at the same incubation time. At the 14-day incubation period, the E_0 value of the untreated soil is 251,81 kPa, while the E_0 of the MICP-treated soil reaches 513,59 kPa, indicating an increase of approximately 104% due to MICP treatment. The greater increase observed at the 14-day incubation period indicates that the effectiveness of biostimulation improves as the biological processes develop. This comparison confirms that the enhancement of initial soil stiffness is primarily governed by biostimulated MICP rather than by the natural soil condition or incubation time alone. The calcium carbonate (CaCO_3) precipitates formed through microbial activity strengthen interparticle contacts, thereby significantly improving the initial elastic response of the soil compared to the untreated condition.

Comparison of E_t at 7 and 14 Days Incubation

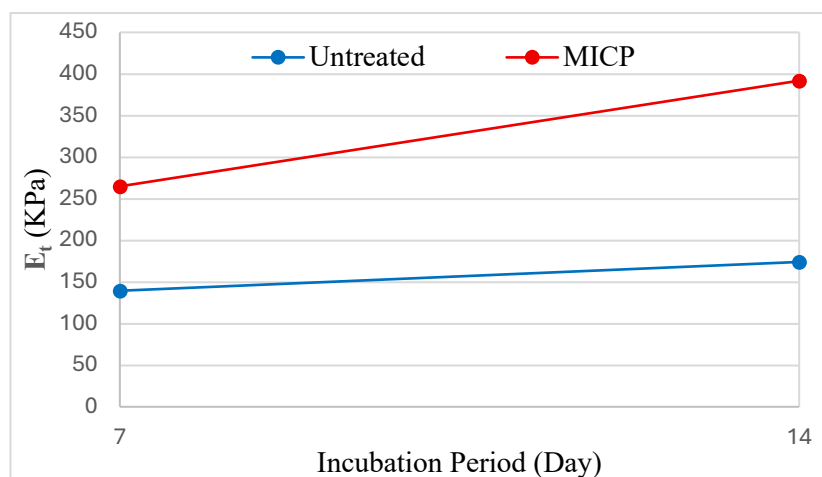


Figure 7. Elastic Modulus (E_t)

Figure 7 presents a comparison of the tangent elastic modulus (E_t) between untreated soil and soil treated with biostimulated MICP at incubation periods of 7 and 14 days. The clear difference in E_t values between the two conditions indicates the direct effect of MICP treatment on soil deformation behavior near the plastic state. At the 7-day incubation period, the E_t value of the untreated soil is 139,47 kPa, whereas the MICP-treated soil exhibits an E_t value of 264,98 kPa, corresponding to an increase of approximately 90% relative to the untreated condition at the same incubation time. This result indicates that MICP-treated soil is able to maintain an elastic response for a longer duration before entering plastic behavior. At the 14-day incubation period, the E_t value of the untreated soil is 174,09 kPa, while the E_t of the MICP-treated soil reaches 392 kPa, indicating an increase of approximately 125% due to MICP treatment. The greater increase observed at the longer incubation period demonstrates that the effectiveness of biostimulation continues to develop with increasing incubation duration. This comparison confirms that the enhancement of E_t is primarily governed by biostimulated MICP rather than by the natural soil behavior. The calcium carbonate (CaCO_3) precipitates formed through microbial activity strengthen interparticle bonding and improve resistance to plastic deformation, resulting in a more stable mechanical response of MICP-treated soil compared to untreated soil.

ANOVA Test

Table 1. One Way ANOVA Test

Parameter	P-Value	
	7 Days	14 Days
E_0	7.71×10^{-4}	3.61×10^{-7}
E_t	2.97×10^{-4}	3.25×10^{-10}

The outcomes of the ANOVA are presented in Table 1. For the E_0 parameter, the obtained p-values were 7.71×10^{-4} at 7 days and 3.61×10^{-7} at 14 days of incubation. In contrast, the E_t parameter yielded p-values of 2.97×10^{-4} at 7 days and 3.25×10^{-10} at 14 days. Since all p-values were below the 0.05 significance level, the differences observed in all evaluated parameters were statistically significant.

Conclusion

This study demonstrates that the application of MICP through the biostimulation approach significantly enhances the stiffness of low-plasticity clay (CL) at both 7 and 14-day incubation periods. At 7 days of incubation, the initial elastic modulus (E_0) and tangent elastic modulus (E_t) of MICP-treated soil increased by approximately 92% and 90%, respectively, compared to the untreated condition, indicating the onset of soil structure modification through early-stage biocementation. With extended incubation to 14 days, the improvements became more pronounced, with E_0 and E_t increasing by approximately 104% and 125%, respectively, reflecting the development of a more consolidated and stable soil structure. These results confirm that biostimulation-based MICP effectively enhances soil stiffness and resistance to plastic deformation through the formation of calcium carbonate (CaCO_3) precipitates that strengthen interparticle bonding. The findings highlight the potential of biostimulated MICP as an efficient and environmentally friendly soil stabilization technique for infrastructure applications in tropical regions.

Future research is recommended to establish quantitative relationships between calcium carbonate (CaCO_3) precipitation and the enhancement of elastic stiffness parameters, particularly E_0 and E_t , to support the development of predictive models for performance-based geotechnical design. The long-term mechanical performance of biostimulated MICP-treated clays under sustained loading and varying environmental conditions also warrants further investigation to assess durability. However, this study was limited to laboratory-scale testing under controlled conditions and did not include detailed CaCO_3 quantification or microstructural analysis. Therefore, field-scale investigations with varied biostimulation conditions are required to fully evaluate the efficiency and long-term stability of MICP application in tropical soils.

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