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Potential Improvement of Hydraulic Resistance in Coastal Soils Using Optimized Microbially Induced Calcium Carbonate Precipitation (MICP): A Case Study of Kish Island

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ABSTRACT

In this study, the efficiency of the microbially induced calcium carbonate precipitation (MICP) process using the bacterium Sporosarcina pasteurii was evaluated for improving the hydro-geochemical properties of the soil of Kish Island with the objective of reducing saline water intrusion from the Persian Gulf. Accordingly, the effects of three independent variables, namely urea concentration, calcium chloride concentration, and reaction time, were optimized with respect to response variables including effluent volume, electrical conductivity, and pH. The optimization results indicated that the minimum effluent volume of 5 mL was obtained at a urea concentration of 0.5 M, a calcium chloride concentration of 0.5 M, and a reaction time of 9 days. In addition, the minimum electrical conductivity value of 69.5 dS/m occurred at a urea concentration of 0.5 M, a calcium chloride concentration of 0.5 M, and a reaction time of 15 days. Finally, the maximum pH value of 7.65 was recorded at a urea concentration of 0.5 M, a calcium chloride concentration of 0.5 M, and a reaction time of 15 days. Based on the findings of this research, the application of Sporosarcina pasteurii for the formation of a biological barrier can be introduced as a promising approach for preventing the advancement of saline water from the Persian Gulf into the soil of Kish Island.

Keywords: *MICP; Sporosarcina bacteria; calcium carbonate; optimization; central composite design*

1. Introduction

Coastal regions across the globe are increasingly exposed to the adverse consequences of saltwater intrusion, a phenomenon driven by sea-level rise, excessive groundwater extraction, climate variability, and rapid urbanization. The encroachment of saline water into freshwater aquifers not only threatens drinking water supplies but also undermines agricultural productivity, soil fertility, and ecosystem sustainability, thereby generating complex socio-economic challenges for coastal communities (Dong et al., 2025; Nasiri et al., 2019; Su et al., 2025). Recent assessments of coastal hydrogeological systems demonstrate that saltwater intrusion has accelerated significantly over the past two decades, particularly in arid and semi-arid coastal zones where freshwater recharge is limited and anthropogenic pressures continue to intensify (Dong et al., 2025; Su et al., 2025). These trends highlight the urgent need for innovative and environmentally sustainable mitigation strategies capable of limiting the inland advancement of saline water while preserving groundwater quality and soil integrity.

Traditional engineering approaches to saltwater intrusion control, including physical barriers, pumping management, artificial recharge, and chemical grouting, often involve high construction costs, substantial energy consumption, and long-term environmental side effects. Moreover, many of these methods fail to address the inherent complexity of subsurface geochemical and biological processes that govern soil permeability and contaminant transport (Konstantinou & Wang, 2023; Konstantinou & Wang, 2024). Consequently, growing attention has been directed toward nature-based and bio-mediated solutions that leverage microbial processes to enhance soil stability, modify hydraulic properties, and promote long-term environmental resilience (DeJong et al., 2010; Seifan & Berenjian, 2019; Tianzheng et al., 2023).

Among these emerging technologies, microbially induced calcium carbonate precipitation (MICP) has attracted significant interest as a sustainable and controllable biogeotechnical technique for soil improvement and hydraulic regulation. MICP is a biologically driven process in which ureolytic bacteria hydrolyze urea to produce carbonate ions that subsequently react with calcium ions to form calcium carbonate ($CaCO_3$) precipitates. These precipitates bind soil particles, reduce pore spaces, enhance mechanical strength, and significantly decrease hydraulic conductivity, thereby limiting fluid migration through the subsurface (Al-Thawadi, 2008; DeJong et al., 2010; Hammes, 2003; Whiffin et al., 2007). Extensive laboratory and field investigations have confirmed the potential of MICP to serve as an effective bio-cementation method for erosion control, ground reinforcement, liquefaction mitigation, dust suppression, and permeability reduction in porous media (Maleki Kaklar & Yavari, 2019; Meyer et al., 2011; Qabany et al., 2012; Wang et al., 2023).

The performance of MICP is strongly influenced by a complex interaction of biochemical, physical, and environmental parameters. These include bacterial strain characteristics, urease activity, nutrient availability, calcium source concentration, pH conditions, temperature, salinity, and incubation time (Achal et al., 2011; Mori & Uday, 2021; Ng et al., 2012; Okwadha & Li, 2010). The selection and optimization of these parameters are critical for achieving uniform and durable calcite precipitation within soil matrices. Among the most widely studied ureolytic microorganisms, *Sporosarcina pasteurii* has emerged as a model organism due to its exceptionally high urease activity and strong capacity to induce rapid $CaCO_3$ precipitation (Achal et al., 2011; Fujita et al., 2017; Nafeesa et al., 2021). Strain enhancement and process optimization studies have demonstrated that *Sporosarcina pasteurii* can generate substantial quantities of calcite under controlled conditions, making it highly suitable for geotechnical and environmental applications (Achal et al., 2011; Clarà Saracho et al., 2020; Haihe et al., 2021).

A major challenge in the practical implementation of MICP is maintaining process stability and precipitation efficiency under diverse environmental stresses, including salinity fluctuations, variable pH regimes, and prolonged operational periods. Coastal environments, in particular, impose additional constraints due to high ionic strength, fluctuating salinity, and chemical interactions between seawater constituents and precipitated minerals (Chen & Achal, 2020; Gat et al., 2017). Several studies have shown that exposure to acidic conditions, dissolved salts, and competing ions can destabilize calcium carbonate precipitates, potentially reducing long-term performance of bio-cemented barriers (Chen & Achal, 2020; Gat et al., 2017). Therefore, comprehensive understanding of precipitation mechanisms and their response to environmental conditions is essential for designing robust MICP-based solutions for coastal protection.

Recent advancements in experimental monitoring techniques and computational modeling have significantly improved the ability to characterize and predict MICP behavior in porous media. Electrical resistivity tomography and micro-scale pH monitoring tools now enable real-time visualization of precipitation dynamics, spatial distribution of calcite phases, and temporal evolution of reaction fronts (Zehner et al., 2020; Zhang et al., 2024). Meanwhile, numerical and machine learning approaches have been introduced to optimize treatment design, predict performance under variable conditions, and minimize environmental risks associated with large-scale deployment (Khoshtinat et al., 2024; Konstantinou & Wang, 2024). These technological developments are accelerating the translation of MICP from laboratory research into practical field applications.

The integration of MICP with hydrological engineering has opened new avenues for controlling subsurface flow systems, including the creation of bio-mediated physical barriers for saltwater intrusion mitigation. Recent investigations demonstrate that properly engineered MICP barriers can significantly reduce hydraulic conductivity of sandy aquifers, thereby slowing or even reversing saline water encroachment (Guo et al., 2024; Konstantinou & Wang, 2024). Moreover, the self-healing nature of biologically precipitated calcite offers distinct advantages over conventional cement-based grouts, including enhanced durability, reduced carbon footprint, and compatibility with natural soil processes (Choi et al., 2017; Mostafa et al., 2016).

Despite these promising developments, the effectiveness of MICP remains highly sensitive to operational parameters. Urea concentration governs the rate of carbonate production, calcium chloride concentration controls mineral saturation levels, and incubation time determines the extent of precipitation and spatial uniformity of the resulting cementation (Mori & Uday, 2021; Ng et al., 2012; Qabany et al., 2012). Excessive reagent concentrations can inhibit bacterial metabolism, induce osmotic stress, and generate heterogeneous precipitation patterns that compromise hydraulic performance (Harkes et al., 2010; Mori & Uday, 2021; Whiffin et al., 2007). Conversely, insufficient reactant supply or inadequate reaction time may yield incomplete pore filling and unstable cementation structures (Okwadha & Li, 2010; Soyson et al., 2021). Consequently, identifying optimal parameter combinations remains one of the most critical challenges for successful field-scale MICP implementation.

From a management and decision-making perspective, the deployment of MICP technologies requires rigorous optimization frameworks that integrate engineering performance, environmental sustainability, economic feasibility, and long-term system reliability. Coastal infrastructure planners and environmental managers increasingly recognize the importance of data-driven optimization methods to guide technology selection and operational design under uncertainty (Konstantinou & Wang, 2024; Tianzheng et al., 2023). Multi-criteria decision models, response surface methodologies, and machine learning techniques are now being employed to support strategic planning of bio-mediated ground improvement projects, particularly in high-risk coastal zones (Konstantinou & Wang, 2024; Zhang et al., 2024).

In regions such as Kish Island, where groundwater salinization poses a persistent threat to freshwater resources, tourism infrastructure, and agricultural development, innovative and adaptive management strategies are urgently required. Previous modeling efforts have highlighted the vulnerability of Kish Island’s aquifers to saltwater intrusion and emphasized the necessity of proactive intervention measures (Shahabinejad Raberi et al., 2015). The adoption of MICP-based biological barriers offers a potentially transformative solution by combining geotechnical stabilization with environmentally compatible processes capable of long-term performance under harsh coastal conditions (Guo et al., 2024; Konstantinou & Wang, 2023).

Furthermore, the evolving landscape of coastal resource management increasingly emphasizes integrated approaches that balance engineering efficiency with ecological protection and socio-economic sustainability. Bio-mediated technologies such as MICP align closely with these objectives by minimizing chemical pollution, reducing carbon emissions associated with cement production, and enhancing the resilience of natural systems (Choi et al., 2017; Mostafa et al., 2016; Seifan & Berenjian, 2019). As coastal populations continue to expand and climate-related pressures intensify, the strategic role of such technologies in sustainable coastal development frameworks will only grow.

Nevertheless, substantial knowledge gaps remain regarding the optimal design of MICP systems for saline environments, particularly concerning the interplay between reagent concentrations, reaction kinetics, bacterial viability, and long-term durability of precipitated barriers under seawater exposure (Chen & Achal, 2020; Gat et al., 2017; Khoshtinat et al., 2024). Addressing these gaps requires systematic experimental investigation supported by robust statistical modeling and optimization analysis.

Therefore, the present study seeks to contribute to this emerging field by conducting a comprehensive optimization of key operational parameters governing MICP performance in coastal soils, with direct implications for the management of saltwater intrusion in vulnerable island environments.

The aim of this study is to optimize the key operational parameters of microbially induced calcium carbonate precipitation for minimizing effluent volume and electrical conductivity while maximizing pH in order to enhance the effectiveness of biological barriers for controlling saltwater intrusion in the soil of Kish Island.

2. Methods and Materials

2.1. Description of the Study Area

Kish Island, which is administratively the center of the Kish District of Bandar Lengeh County in Hormozgan Province, covers an area of 90.457 km² and is located in the southern half of the northern temperate zone between 26°28′34″ to 26°34′40″ N latitude and 53°54′04″ to 54°02′12″ E longitude from the Greenwich Prime Meridian, at a distance of approximately 18 km from the southern coast of Iran in the Persian Gulf. According to the Stocklin classification, this island is considered part of the folded Zagros zone. The geological structure of Kish Island is an anticline with a west–southeast axis, which is structurally consistent with the general structure of the Zagros Mountains.

The geological formations of Kish Island are composed mainly of limestone deposits, and the maximum thickness of its aquifer layers has been estimated at approximately 25 m based on exploratory drilling. According to previous studies, the depth to the groundwater table on the island ranges from 4 to 22 m, with an average depth of 6.87 m.

2.2. Materials

Based on previous studies conducted across Kish Island and analyses of the regional soil map and Geographic Information System (GIS), it was determined that vulnerability to salinity intrusion is greater in the eastern and southeastern regions as well as in the western and southwestern parts of the study area on Kish Island. Therefore, experimental samples were randomly collected from these zones. Soil samples were obtained from the island's coastal areas using standard equipment, including soil samplers and plastic bags. To ensure adequate environmental representation, a sufficient quantity of soil (at least 1000 g) was collected from each location.

High-purity urea and calcium chloride dihydrate were procured from Merck (Germany), along with the required number of Petri dishes and plastic laboratory containers from chemical and laboratory equipment suppliers.

The bacterium used in this study was *Sporosarcina pasteurii* (PTCC No. 1645; DSM 33), which was obtained from the Iranian Research Organization for Science and Technology, Center for Iranian Fungal and Bacterial Culture Collection.

Approximately 30 L of seawater was collected from the coastal area of Kish Island and transported to Tehran.

2.3. Bacterial Preparation

Sporosarcina pasteurii, an ureolytic bacterium, was cultured in nutrient broth medium until reaching the desired concentration and subsequently centrifuged. Specifically, one vial of the bacterium with 2% urea was inoculated into 30 mL of autoclaved liquid nutrient broth at pH 9 and incubated at 30°C for 48 h on a shaker. After 48 h, the broth became completely turbid, indicating bacterial growth.

To confirm bacterial purity, a loopful of the turbid culture was streaked onto solid nutrient agar containing 2% urea. The appearance of single bacterial colonies after 48 h confirmed the purity of the culture.

2.4. Sample Preparation

After drying, sieving, and homogenizing the soil samples, approximately 2 kg of soil was transferred into each sample container with dimensions of 20 cm (length) × 13 cm (width) × 7 cm (height). From this amount, 250 g of soil was placed at the center of the plastic containers between two glass plates spaced 1 cm apart to allow controlled MICP treatment in this section. A specific concentration of the bacterial suspension was then added, and urea and calcium chloride solutions were introduced according to the experimental design (Table 1).

After 7 days, 50 mL of Persian Gulf seawater was added daily to each soil sample, and analyses were conducted from Day 9 onward (no water passage through the samples was observed before Day 9) in accordance with Table 1.

2.5. Measurement of Effluent Volume, Electrical Conductivity, and pH

Beginning on Day 9, and at predetermined intervals as specified in Table 1, the effluent volume was measured using a graduated cylinder, the electrical conductivity (EC) of the effluent was measured using an EC meter (Model 712, Metrohm, Switzerland), and the pH of the effluent was measured using a pH meter (Model 744, Metrohm, Switzerland).

2.6. Statistical Analysis

Response Surface Methodology (RSM) using a Central Composite Design (CCD) with three variables at three levels (Table 1) was employed. The independent variables included calcium chloride concentration (M), urea concentration (M), and time (days), and their effects on three response variables—effluent volume (mL), EC (dS/m), and pH—were examined. Table 1 presents the coded and actual levels of the independent variables.

Table 1

Levels and Values of the Independent Variables

Variable	Symbol	−1	0	+1
Urea (M)	A	0	0.5	1
Calcium chloride (M)	B	0	0.5	1
Time (days)	C	9	12	15

3. Findings and Results

Table 2 presents the results obtained from the Central Composite Design. As observed, the effluent volume ranged from 5 to 45 mL, electrical conductivity varied from 69.5 to 89.9 dS/m, and pH values ranged from 6.59 to 7.65.

Table 2

Levels of the Variables (Urea, Calcium Chloride, and Time) and the Corresponding Responses of Effluent Volume, Electrical Conductivity, and pH

Treatment	Urea (M)	$CaCl_2$ (M)	Time (days)	Effluent Volume (mL)	EC (dS/m)	pH
1	0	0	9	35	85.0	6.87
2	1	0	9	20	89.9	6.60
3	0	1	9	21	89.5	6.80
4	1	1	9	22.5	86.7	6.80
5	0	0	15	45	71.3	7.62
6	1	0	15	33	71.9	7.60
7	0	1	15	35	72.6	7.60
8	1	1	15	36	73.6	7.48
9	0	0.5	12	30	76.1	7.47
10	1	0.5	12	28	80.1	7.37
11	0.5	0	12	25	75.9	7.56
12	0.5	1	12	27	80.9	7.32
13	0.5	0.5	9	5	80.8	6.59
14	0.5	0.5	15	25	69.5	7.65
15	0.5	0.5	12	20	74.1	7.41
16	0.5	0.5	12	19	74.2	7.45
17	0.5	0.5	12	18.5	74.8	7.49

The results of the analysis of variance (ANOVA) in Table 3 indicate that the second-order model for effluent volume, electrical conductivity, and pH is statistically significant at the 95% confidence level, since the *p*-values are less than 0.05. In addition, the quadratic polynomial model shows good agreement with the experimental data, as the lack-of-fit test is not significant for all three parameters—effluent volume, electrical conductivity, and pH—with values of 0.0584, 0.0254, and 0.1086, respectively. Only 3%, 4%, and 3% of the data could not be explained by the proposed models, with coefficients of determination (R^2) of 97%, 96%, and 97%, respectively. Furthermore, the adjusted coefficients of determination for effluent volume, electrical conductivity, and pH were high (92%, 90.45%, and 93%, respectively), confirming the adequacy and robustness of the model fit. These findings indicate that the quadratic equations are sufficient to predict the effects of the three independent variables, and higher-order equations are not required. The second-order equations describing effluent volume, electrical conductivity, and pH—representing mathematical relationships for determining optimal conditions—are presented below:

Equation 1. Effluent Volume

Water Output = −52.5576 ± 53.3581A ± 41.3581B + 13.9363C + 14.75AB + 0.208333AC + 0.375BC + 38.1831A² + 26.1831B² ± 0.494914C²

Equation 2. Electrical Conductivity

EC = 102.237 ± 6.56599A ± 9.64599B ± 1.57066C ± 3.65AB ± 0.041667AC + 0.141667BC + 10.431A² + 11.631B² ± 0.0380282C²

Equation 3. pH

pH = 1.06843 ± 0.298444A + 0.0735563B + 0.932871C + 0.085AB + 0.108333AC ± 0.0225BC + 0.0239437A² + 0.103944B² ± 0.0326682C²

Table 3

Results of Analysis of Variance (ANOVA) for the Effects of Independent Variables on Effluent Volume, Electrical Conductivity, and pH

(a) Effluent Volume

Source	Sum of Squares	df	Mean Square	F-value	*p*-value
Model	1300.97	9	144.55	20.65	0.0003
Quadratic	47.83	5	9.57	16.40	0.0584
Two-factor	642.24	8	80.28	137.62	0.0072
Interaction effects	594.41	3	198.14	28.31	0.0003
Residual	48.90	7	7.00	—	—
Lack of fit	47.83	5	9.57	16.40	0.0584
Pure error	1.17	2	0.5833	—	—
Total	1349.97	16	—	—	—

$R^2 = 0.97$
Adjusted $R^2 = 0.92$

(b) Electrical Conductivity

Source	Sum of Squares	df	Mean Square	F-value	*p*-value
Model	641.95	9	71.33	17.84	0.0005
Two-factor	115.12	8	14.39	100.40	0.0099
Quadratic	27.71	5	5.54	38.66	0.0254
Interaction effects	87.42	3	29.14	7.29	0.0147
Residual	27.99	7	4.00	—	—
Lack of fit	27.71	5	5.54	38.66	0.0254
Pure error	0.2867	2	0.1433	—	—
Total	669.94	16	—	—	—

$R^2 = 0.96$
Adjusted $R^2 = 0.9045$

(c) pH

Source	Sum of Squares	df	Mean Square	F-value	*p*-value
Model	2.21	9	0.2452	24.11	0.0002
Two-factor	0.3877	8	0.0485	30.29	0.0323
Quadratic	0.0680	5	0.0136	8.50	0.1086
Interaction effects	0.3197	3	0.1066	10.48	0.0056
Residual	0.0712	7	0.0102	—	—
Lack of fit	0.0680	5	0.0136	8.50	0.1086

Pure error	0.0032	2	0.0016	—	—
Total	2.28	16	—	—	—

R^2 = 0.97
Adjusted R^2 = 0.93

3.1. Optimization

As stated, the objective of this study was to optimize the conditions to achieve the minimum effluent volume and minimum electrical conductivity (EC) of the effluent water, while simultaneously obtaining the maximum pH. The optimization results indicated that the minimum effluent volume of 5 mL was obtained at a urea concentration of 0.5 M, a calcium chloride concentration of 0.5 M, and a reaction time of 9 days. Furthermore, the minimum EC value of 69.5 dS/m corresponded to a urea concentration of 0.5 M, a calcium chloride concentration of 0.5 M, and a reaction time of 15 days. Finally, the maximum pH value of 7.65 was achieved at a urea concentration of 0.5 M, a calcium chloride concentration of 0.5 M, and a reaction time of 15 days.

The effluent volume, as shown in Table 2, varied in the range of 5 to 45 mL. The minimum value was associated with experimental condition No. 13, whereas the maximum value occurred under experimental condition No. 5. The three-dimensional response surface plots in Figure 1 demonstrated that calcium chloride concentration exerts a highly significant and nonlinear effect on effluent volume. When the calcium chloride concentration increased from 0 to 0.5 M, the effluent volume decreased markedly, indicating substantial enhancement of the calcium carbonate precipitation process (Al-Thawadi, 2008; Zhang et al., 2022). At 0.5 M, sufficient calcium ions are supplied to react with carbonate produced by *Sporosarcina pasteurii* without generating inhibitory effects, resulting in uniform and effective calcite crystal formation within soil pores and a pronounced reduction in soil hydraulic conductivity (Al-Thawadi, 2008; Zhang et al., 2022).

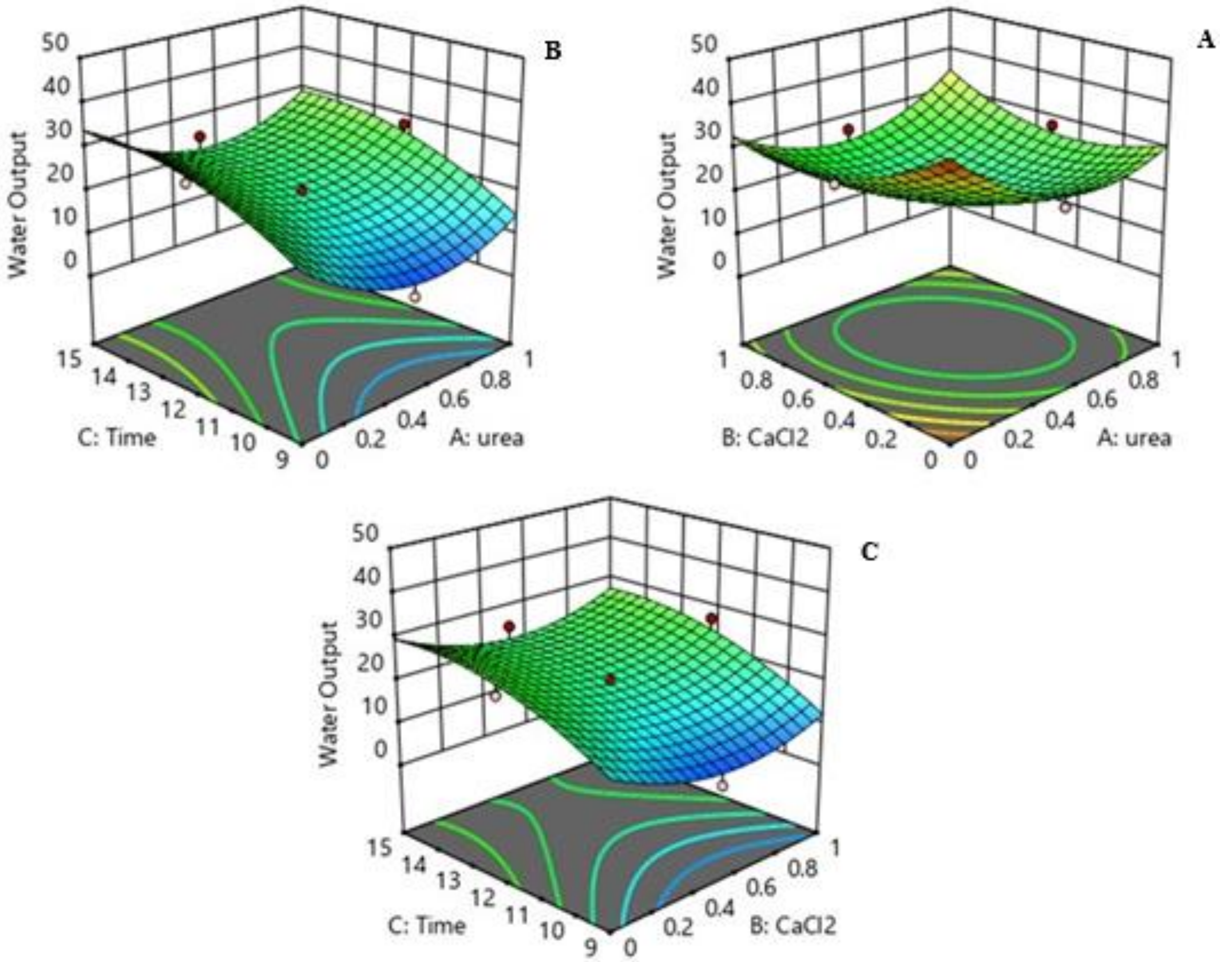
However, further increasing calcium chloride concentration to 1 M led to a renewed increase in effluent volume. This increase can be attributed to two primary mechanisms. First, osmotic and ionic toxicity: high calcium chloride concentrations impose severe osmotic stress and elevated ionic strength, inhibiting urease activity and consequently reducing urea hydrolysis and carbonate ion production, which limits calcite precipitation. Second, formation of heterogeneous and coarse crystals: excessive calcium supply accelerates nucleation and crystal growth, producing larger crystals localized near injection zones, which generate preferential flow paths instead of uniformly sealing soil pores (Harkes et al., 2010; Wang et al., 2023; Whiffin et al., 2007).

Urea concentration exhibited a nonlinear behavior similar to calcium chloride. Increasing urea concentration from 0 to 0.5 M significantly reduced effluent volume, whereas further increase to 1 M reversed the trend. At 0.5 M urea, optimal conditions for urease activity of *Sporosarcina pasteurii* are established, leading to efficient urea hydrolysis, sufficient carbonate and ammonium production, and uniform calcium carbonate precipitation throughout soil pores, which substantially lowers permeability and effluent volume (Al-Thawadi, 2008). Conversely, at 1 M urea, several adverse effects occur: (1) enzymatic inhibition due to competitive inhibition of urease and excessive ammonia accumulation, which raises local pH and suppresses enzymatic activity; (2) osmotic stress and toxicity reducing bacterial viability and metabolic activity; and (3) rapid, heterogeneous precipitation, generating dense layers near injection points while leaving downstream zones under-treated, ultimately increasing overall permeability (Ng et al., 2012; Okwadha & Li, 2010).

The investigation period, defined as incubation time following bacterial and nutrient injection, was selected from Day 9 onward, as water breakthrough was first observed at this time. Response surface analysis revealed that increasing incubation time from 9 to 15 days led to a continuous and approximately linear increase in effluent volume, indicating gradual decline in precipitation efficiency over time. During early stages (e.g., Day 9), bacterial activity is maximal, promoting rapid urea hydrolysis, carbonate generation, and effective pore sealing, yielding minimum effluent volume (Okwadha & Li, 2010). However, extended incubation to Days 12 and 15 increases effluent volume due to: (1) declining bacterial activity and viability as nutrients are depleted and ammonia accumulates, suppressing urease function (Gat et al., 2017); (2) partial dissolution or instability of calcite precipitates under chemical influences of saline pore water; and (3) structural and mechanical changes within the soil matrix over time, generating new flow pathways (Achal et al., 2011; Konstantinou & Wang, 2023; Seifan & Berenjian, 2019).

Figure 1

Three-dimensional response surface plots showing the effects of independent variables on effluent volume

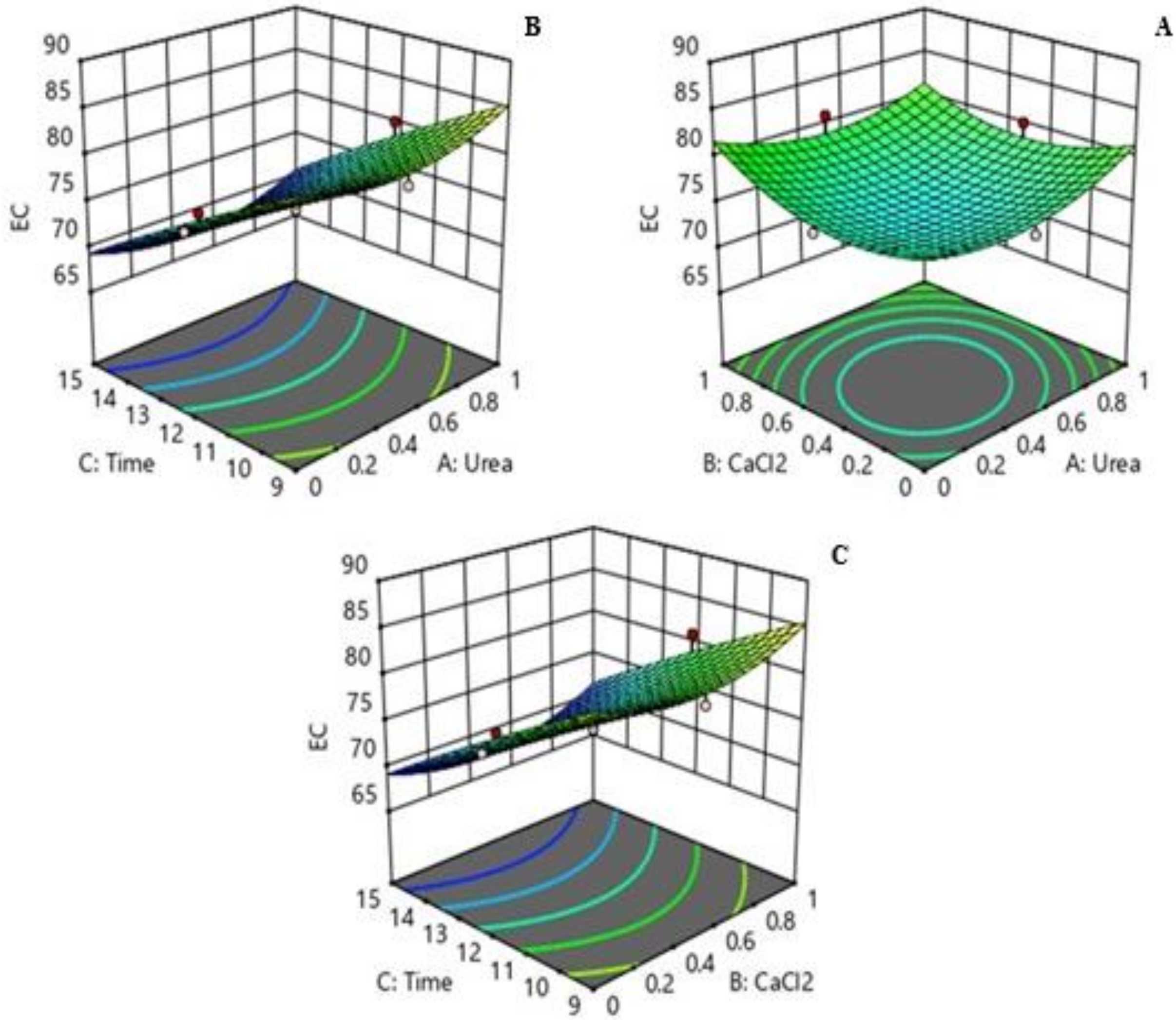


3.2. Electrical Conductivity (EC)

Table 2 indicates that EC varied from 69.5 to 89.9 dS/m, with the minimum corresponding to experimental condition No. 14 and the maximum to No. 2. Response surface plots demonstrated that EC generally increased with higher calcium chloride and urea concentrations and with shorter incubation times. Elevated concentrations introduce larger quantities of soluble ions into the system, while incomplete MICP under short incubation allows residual ions to exit with effluent water. In contrast, longer incubation enables more complete urea hydrolysis and ion immobilization within calcite, thereby reducing EC. Consequently, optimal reduction of EC was achieved under moderate reagent concentrations and extended incubation, consistent with earlier findings (Mori & Uday, 2021; Qabany et al., 2012; Wang et al., 2023; Zhang et al., 2024).

Figure 2

Three-dimensional response surface plots showing the effects of independent variables on EC

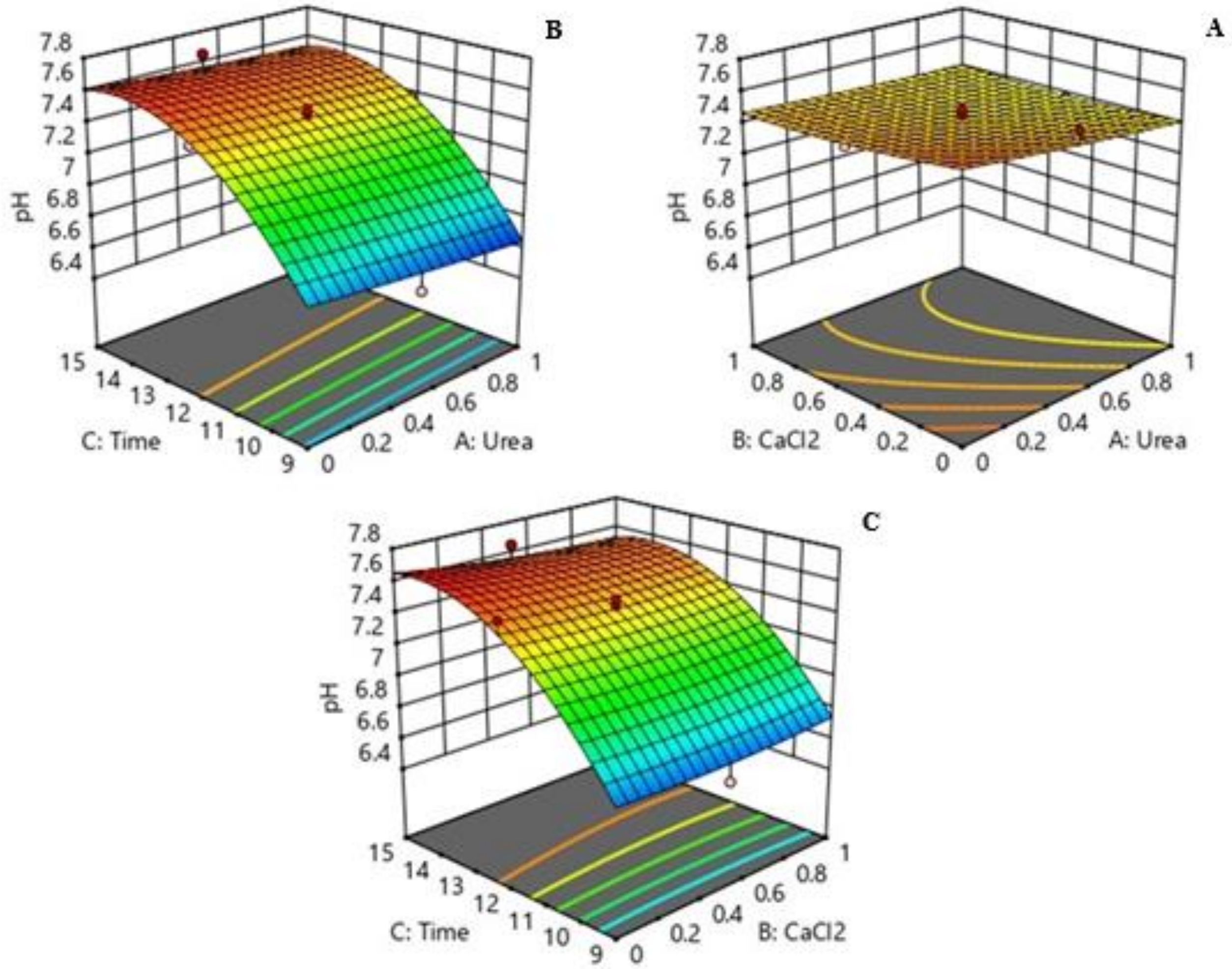


3.3. pH

As shown in Table 2, pH ranged from 6.59 to 7.65, with the lowest and highest values observed in experimental conditions No. 13 and No. 14, respectively. Response surface analysis revealed that increasing calcium chloride and urea concentrations decreased pH, whereas increasing incubation time elevated pH. High reagent concentrations limit bacterial hydrolysis efficiency and promote carbonate precipitation reactions that release dissolved carbon dioxide, shifting buffering equilibria toward lower pH (Khoshtinat et al., 2024). Conversely, prolonged incubation permits sustained urea hydrolysis, continuous ammonia generation, and stable elevation of pH. Therefore, maximum pH, which represents the study's optimization target, occurred under low reagent concentrations and extended incubation, conditions that also enhance long-term calcium carbonate stabilization and effective seawater intrusion control (Haihe et al., 2021; Nafeesa et al., 2021).

Figure 3

Three-dimensional response surface plots showing the effects of independent variables on pH



4. Discussion and Conclusion

The present study demonstrates that microbially induced calcium carbonate precipitation (MICP) can be effectively optimized to control seawater intrusion by simultaneously minimizing effluent volume and electrical conductivity while maximizing pH, thereby improving the hydro-geochemical stability of coastal soils. The observed responses of effluent volume, electrical conductivity, and pH clearly indicate that the interaction among urea concentration, calcium chloride concentration, and incubation time governs the efficiency of the biocementation process. These findings are consistent with the growing body of research indicating that MICP performance is highly sensitive to biochemical reaction conditions and that non-linear responses are inherent to biologically driven mineralization processes (Mori & Uday, 2021; Ng et al., 2012; Tianzheng et al., 2023).

The minimum effluent volume achieved at moderate reagent concentrations (0.5 M urea and 0.5 M $CaCl_2$) and shorter incubation (9 days) confirms that excessive reagent loading does not necessarily enhance performance. Instead, optimal reagent balance enables efficient urease activity, controlled carbonate generation, and uniform calcite precipitation within soil pores. This behavior directly aligns with earlier experimental findings demonstrating that moderate reactant concentrations maximize precipitation efficiency and pore filling while avoiding inhibitory effects on bacterial metabolism (Achal et al., 2011; Okwadha & Li, 2010; Qabany et al., 2012). The sharp reduction in effluent volume under optimal conditions reflects substantial pore clogging and hydraulic conductivity reduction, consistent with the soil reinforcement mechanisms described by

Whiffin and colleagues and DeJong et al. (DeJong et al., 2010; Whiffin et al., 2007).

The non-linear influence of calcium chloride concentration observed in this study further supports the established understanding of calcium's dual role in MICP systems. At moderate concentrations, calcium ions act as the limiting reactant enabling effective calcite nucleation and crystal growth. However, excessive calcium supply induces osmotic stress, enzymatic inhibition, and formation of coarse heterogeneous crystals that fail to uniformly block pore spaces, ultimately increasing permeability. Similar inhibitory effects of high calcium concentration on bacterial activity and precipitation uniformity have been documented in previous investigations (Harkes et al., 2010; Mori & Uday, 2021; Wang et al., 2023). This phenomenon underscores the necessity of precise reagent management for achieving long-term hydraulic performance in MICP-treated soils.

Urea concentration exhibited a comparable non-linear effect on effluent volume and system performance. The optimal urea concentration (0.5 M) provided sufficient substrate for urease-driven carbonate production while avoiding the detrimental impacts of excessive ammonia accumulation and enzyme inhibition that occurred at 1 M urea. The inhibitory influence of high urea concentrations has been repeatedly observed in laboratory and field-scale studies, where elevated ammonia production leads to extreme pH shifts and suppressed urease activity (Fujita et al., 2017; Mori & Uday, 2021; Ng et al., 2012). The present findings thus reinforce the importance of regulating nitrogen input not only for maximizing calcite yield but also for maintaining microbial viability and metabolic stability.

The temporal dynamics of incubation revealed that the most effective hydraulic sealing occurred at early stages (9 days), whereas extended incubation led to gradual performance degradation. This behavior reflects the natural life cycle of bacterial populations and the progressive depletion of nutrient resources, resulting in reduced carbonate production over time. Additionally, prolonged exposure to saline conditions may destabilize previously formed calcite structures through dissolution or mechanical restructuring. These mechanisms are consistent with observations by Gat et al. and Chen and Achal, who reported long-term stability challenges of microbially precipitated carbonates under chemically aggressive environments (Chen & Achal, 2020; Gat et al., 2017). Consequently, the results highlight the necessity of designing MICP systems that achieve maximum sealing efficiency early in the treatment period.

Electrical conductivity trends further validate the effectiveness of the optimized treatment. The lowest EC values occurred under low reagent concentrations and extended incubation, indicating that most dissolved ions were immobilized within the calcite matrix rather than discharged with effluent water. This behavior confirms that optimized MICP not only reduces permeability but also significantly improves effluent water quality by trapping dissolved salts and reaction byproducts within the solid phase. Similar reductions in effluent salinity have been reported in hydrological applications of MICP, particularly in coastal aquifer protection systems (Guo et al., 2024; Konstantinou & Wang, 2024; Zhang et al., 2024). The present findings therefore extend existing knowledge by quantitatively demonstrating the dual hydraulic and chemical benefits of optimized MICP treatment.

The pH response exhibited a complementary pattern: while high reagent concentrations suppressed pH elevation due to enzymatic inhibition and carbonate buffering effects, extended incubation produced sustained alkalinity through continued ammonia generation. The highest pH values obtained under low reagent concentrations and long incubation reflect optimal ureolysis and minimal buffering interference from excess calcium ions. These results are consistent with the mechanistic models of pH-dependent precipitation dynamics described by Khoshtinat et al., Haihe et al., and Lai et al. (Haihe et al., 2021; Khoshtinat et al., 2024; Lai et al., 2022). Elevated and stable pH is critical for promoting calcite stability and long-term durability of bio-cemented structures, especially in saline coastal environments.

From a broader management perspective, the study confirms that MICP can function as an effective biological barrier for mitigating seawater intrusion in vulnerable coastal zones such as Kish Island. When properly optimized, the technology offers a low-carbon, environmentally compatible alternative to conventional physical and chemical remediation approaches. These advantages align with contemporary sustainability goals and integrated coastal zone management frameworks that prioritize ecological resilience alongside infrastructure protection (Dong et al., 2025; Konstantinou & Wang, 2023; Su et al., 2025). The ability of MICP to enhance soil integrity, regulate hydraulic conductivity, and improve water quality simultaneously positions it as a strategically valuable tool for long-term coastal resource management.

The findings further contribute to the growing consensus that successful implementation of MICP requires site-specific optimization and continuous monitoring. Advances in real-time monitoring techniques such as electrical resistivity tomography and micro-scale pH tracking provide powerful tools for controlling treatment quality and predicting long-term performance (Zehner et al., 2020; Zhang et al., 2024). When combined with computational optimization and machine learning frameworks, these tools enable decision-makers to design highly efficient and adaptive bio-mediated remediation systems (Khoshtinat et al., 2024; Konstantinou & Wang, 2024). The present study provides empirical evidence supporting the feasibility and reliability of such integrated approaches.

Overall, the results strongly support the hypothesis that carefully optimized MICP treatment can significantly enhance soil performance and provide a robust defense against seawater intrusion. The convergence of biological efficiency, hydraulic control, and environmental sustainability observed in this study reflects the transformative potential of biogeotechnical solutions for future coastal infrastructure protection and groundwater management.

This study was conducted under controlled laboratory conditions that cannot fully capture the complexity of natural coastal environments. Factors such as heterogeneous soil composition, fluctuating groundwater chemistry, temperature variability, biological competition, and large-scale hydrodynamic forces were not explicitly modeled, which may influence MICP performance in real field conditions.

Future studies should investigate long-term field-scale performance of optimized MICP systems under natural coastal conditions, including multi-seasonal monitoring, coupled geochemical-hydrological modeling, evaluation of microbial community dynamics, and integration of smart sensor networks for real-time process control and adaptive optimization.

For practical implementation, coastal managers and engineers should adopt phased deployment strategies, prioritize site-specific optimization, integrate monitoring and feedback mechanisms into system design, and develop operational guidelines that ensure environmental safety, regulatory compliance, and economic feasibility of large-scale MICP-based coastal protection projects.

Authors' Contributions

Authors contributed equally to this article.

Declaration

In order to correct and improve the academic writing of our paper, we have used the language model ChatGPT.

Transparency Statement

Data are available for research purposes upon reasonable request to the corresponding author.

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Declaration of Interest

The authors report no conflict of interest.

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Ethics Considerations

In this research, ethical standards including obtaining informed consent, ensuring privacy and confidentiality were considered.

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