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Laboratory Study on Soil Desaturation by Microbial Methods

Rima Das[1] Sreelakshmi N[2] and and Dr. K. Muthukkumaran[3]

[1] *Research Scholar, Department of Civil Engineering, National Institute of Technology Tiruchirappalli-620015,*
[2] *M Tech Geotechnical Engineering, Department of Civil Engineering, National Institute of Technology Tiruchirappalli-620015*
[3] *Professor, Department of Civil Engineering, National Institute of Technology Tiruchirappalli-620015,*
kmk@nitt.,edumanika.dasnit@gmail.com, lakshminkamath@gmail.com

Abstract. The soil desaturation method for mitigation of sand liquefaction has gained interest in recent years. The application of microbes for various ground improvement studies has proven to be cost-effective and non-disruptive compared to other conventional methods. The potential of soil desaturation by introduction of non-pathogenic denitrifying bacteria is studied in this paper. This bio-chemical process used to induce partial saturation leads to the nucleation of gas bubbles within the soil pores which generate gas as final product. This evolved gas occupying the pore spaces shifts the soil matrix to quasi-saturated state affecting the rate of pore-pressure. Prior to the evaluation of soil strength properties, batch tests were conducted to understand the effect of initial nitrate concentration on generated gas volume. To understand the effect of microbial desaturation on static behavior of the soil, undrained strain-controlled static triaxial tests were conducted for loose sand condition. Results showed increase in peak deviatoric stress after treatment, change being prominent in loose condition indicating enhanced resistance to static liquefaction. B-value and water content measurements were taken to monitor the change in degree of saturation. The rate of excess pore pressure development in undrained compression loading also showed a decreasing trend.

Keywords: *liquefaction; gas bubbles; desaturation; denitrification.*

1. Introduction

Liquefaction is the loss of shear strength in completely saturated loose sands caused by an excessive buildup of pore water pressure following repeated loading or dynamic excitation, such as an earthquake. Understanding the process of liquefaction and creating strategies for estimating the liquefaction potential at a specific location during a seismic event have been the subject of intense research. Geotechnical engineers have developed a variety of site improvement techniques that can greatly reduce the consequences of liquefaction while liquefaction research is ongoing. Current mitigation methods are expensive and hardly ever applicable to existing structures. There is still a great deal of concern about minimizing the consequences of liquefaction on existing structures.

The primary conventional liquefaction mitigation techniques are soil replacement, sand compaction piles, vibration compaction, dynamic compaction, blast compaction, grouting, the deep mixing pile method, gravel pile method, dissipation through screen pipes, and lowering the groundwater table. These methods have been widely applied in engineering, and the tools and technology they employ for building have been shown to be dependable and efficient. Liquefaction takes place when loose sand-based soil becomes saturated with groundwater. Soil densification, groundwater level decrease, and drainage are the three categories into which liquefaction mitigation measures can be subdivided. Based on liquefaction mitigation principles, researchers divided liquefaction mitigation measures into four groups: soil densification, sand grain bonding, groundwater table lowering, and drainage. It might be challenging to apply these techniques to maintain an existing structure. Because of the potential influence on nearby infrastructure, the high expense of construction, and the risk of damaging nearby environments and water bodies, most approaches are difficult to implement. Lowering the degree of saturation can increase sand strength and resistance to liquefaction, according to certain experimental data (Yoshimi et al., 1989; Okamura 2006; He and Chu, 2014 Wang et al., 2016;Mele et al., 2019; Tsukamoto 2019). A considerable anti-liquefaction impact has been shown to reduce the saturation level of saturated sand by up to 5%. Several techniques have been adopted to achieve the purpose, including air injection (Ishihara et al., 2003; Yasuhara et al., 2008; Okamura et al., 2008, 2011; Raghunandan et al., 2014; Marasini et al., 2015; Zeybek et al.,2017; Amanta et al., 2021; Flora et al.,2021), electrolysis and drainage recharge method (Yegain et al., 2007), chemical method (Bayat et al., 2013,2020; Nababan et al., 2015), biogenic desaturation (Paassen et al., 2010; Rebata -Landa et al., 2012 He et al., 2016; Pham et al., 2016; Donnell et al., 2017a, 2017b; Mousavi et al., 2019;2021; Peng et al., 2021), inducing microbubbles.

Induced partial saturation (IPS) refers to the process of transitioning soil from a saturated to an unsaturated state using various methods. A cost-effective liquefaction mitigation strategy is to move the soil from a saturated to a partially saturated state. By inducing a very little amount of gas or air in the pores of fully saturated sands, Induced Partial Saturation aims to prevent liquefaction. IPS will have a cost-effective implementation for both new and existing structures, which will give it an advantage over alternative mitigating strategies. Due to the higher compressibility of the gas/air-water mixture in the voids, the generation of gas/air reduces the excess pore water pressure.

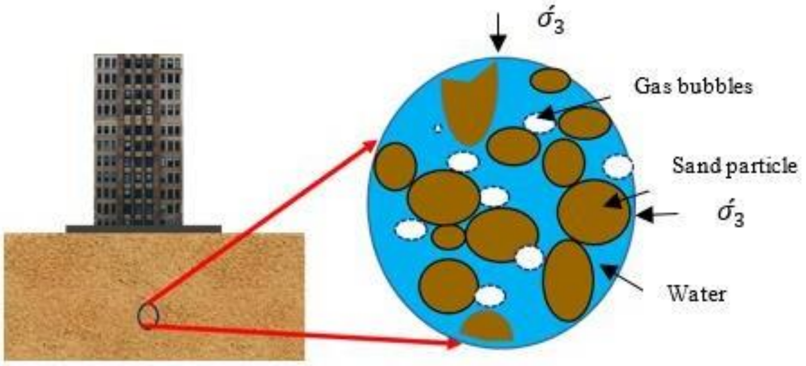


Fig. 1. Concept of IPS technique

Theoretically, it is possible to demonstrate that cyclic loading reduces the extra pore water pressure in sands with trapped gas and air (Fig.1.).By assessing soils in an undrained environment, the laboratory can investigate how liquefaction temporarily suppresses water drainage in the field. Theoretically, both static loading ($\Delta\sigma'$) and dynamic loading can show an excessive accumulation of pore water pressure. According to Terzaghi's effective stress concept, the soil will experience a change in effective stress when a static load ($\Delta\sigma$) is applied.

$$\Delta u = \frac{1}{1+\frac{n*[SC_W+\frac{1-S}{u_a}]}{Cs}}\Delta\sigma \qquad (1)$$

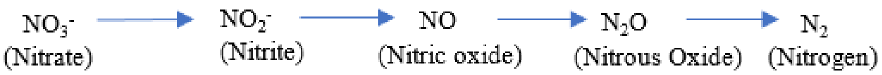
The preceding equation 1 implies that since C_w is almost zero at S=1.0, Δu will be equal to the applied loading. Due to the (1-S)/u_a element in the equation, however, Δu will be lower than the applied loading when S< 1. On the other hand, the air's ability to be compressed is dependent on the pore air pressure. Finn et al., (1976) estimated the excess pore water pressure in fully saturated sand during one loading cycle of basic shear tests as follows.

$$\Delta u = \frac{\Delta\in_{vd}}{\frac{1}{Er}+\frac{n}{K_W}} \qquad (2)$$

Where Δu denotes, Excess pore pressure per load cycle,$\Delta\in_{vd}$ denotes net volumetric strain increment corresponding to the decrease in volume occurring during the load cycle in drained case, E_r denotes one dimensional rebound modulus of sand at σ_v', n denotes porosity of the soil, K_w denotes bulk Modulus of Water. Due to the rise in the compressibility expression of the pore fluid, the excess pore water pressure generated during each loading cycle in air-entrapped sands will be lower than in fully saturated sands. The objective of the study is to evaluate the impact of microbial induced partial saturation on the undrained static liquefaction of loose sand.

2. Biologically induced partial saturation

Several strategies for reducing the liquefaction potential through induced partial saturation have been used in the past. The most frequent biogenic gases identified near the surface were CO_2, H_2, CH_4, and N_2. Nitrogen gas (N_2) is produced during denitrification, an anaerobic dissimilatory reduction of nitrate mediated by microbes. N_2 is a particularly suitable biogenic gas because it is neither explosive nor a greenhouse gas. It has a limited solubility in water and is chemically inert.



2.1 Estimation of gas production

Based on two extreme scenarios—no growth and maximal growth—the thermodynamic method suggested by Heijnen et al., (2010) is used to calculate the stoichiometry of the denitrification process using acetate $C_2H_3O_2$ as the energy and carbon source and nitrate as the oxidizing agent and nitrogen source. Denitrification stoichiometry for maximum growth circumstances is (Pham et al., 2016):

$1.21\ C_2H_3O_2^- + 0.97\ NO_3^- + 0.76\ H^+ \rightarrow 1CH_{1.8}O_{0.5}N_{0.2} + 1.41\ HCO_3^- + 0.39\ N_2 + 0.59\ H_2O$ (3)

And for conditions of no biomass growth, but only maintenance:

$C_2H_3O_2^- + 1.6\ NO_3^- + 0.6\ H^+ \rightarrow 0.8\ N_2 + 2\ HCO_3^- + 0.8\ H_2O$ (4)

The reaction stoichiometry of denitrifying organisms is between these two boundaries at any metabolic state. As a result, the projected range of the N_2/NO_3^- ratio is 0.40 to 0.50, and the ratio of HCO_3^-/NO_3^- is 1.45 to 1.25, corresponding to the maximum and no growth boundaries, respectively. These ratios can be used to determine the ranges of generated N_2 and HCO_3 when the consumed concentration of NO_3 is known.

3. Materials and microbes culture

3.1Soil properties

Bulk quantities of soil have been collected from an excavation site near Nagapattanam. Tests were conducted to determine physical and engineering properties of soil samples as per respective IS codes. Hence, the soil is classified as SP (poorly graded sand) (as per IS 1498:1970). The soil sample particle size distribution shows that it is enclosed inside the limits of extremely prone to liquefiable soil (Tsuchida, 1970) (Fig. 2.). The results index on collected soil were presented in table 1.

Table 1. Index properties of sand used in this study

Index properties	Values
Specific gravity	2.622
Maximum void ratio	0.725
Minimum void ratio	0.643
Sand content	92%
Silt content	8%
c_u	2.14
c_c	3.78
plasticity	non-plastic
Soil classification	SP

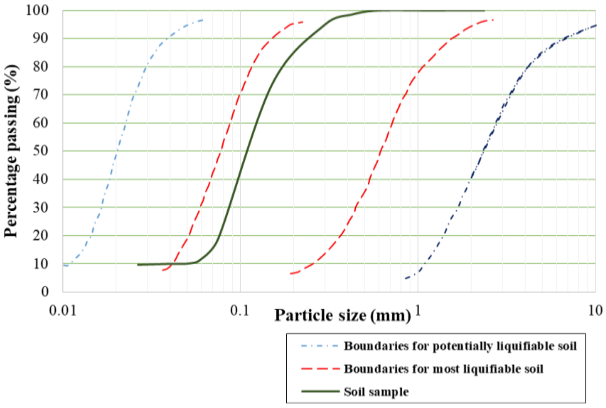


Fig. 2 Cumulative % finer vs sieve size for soil sample (Tsuchida, 1972)

3.2 Microbial culture

Pseudomonas stutzeri is a denitrifying, Gram-negative, rod-shaped bacterium. It is a mesophilic organism that grows best at a temperature of around 35 °C, while it can grow at temperatures as low as 4 °C and as high as 44 °C. This bacterium has a doubling time of about 53 minutes when cultured on a nitrate broth medium at 32 °C. This bacterium generally grows at a pH of 7. *Pseudomonas stutzeri* was chosen to perform the soil desaturation experiment. Dry powdered *Pseudomonas stutzeri* (MTCC 863) was purchased and cultured on a petri dish stabilized with nutrient agar (Fig.3.)

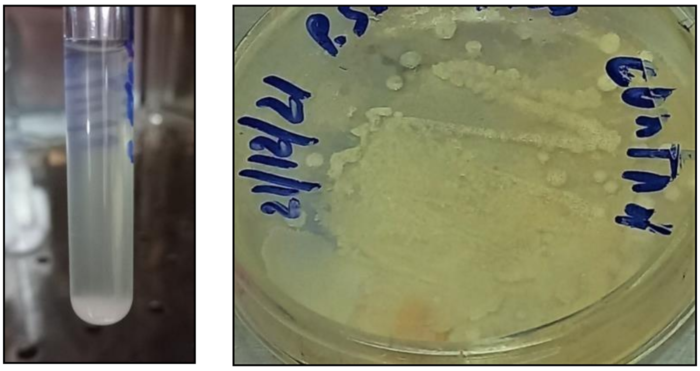

(a) (b)

Fig.3. positive microbial growth in (a) test tube and (b) petridish.

4. Experimental test procedure

To address the requirements of the investigation, a cyclic triaxial setup with a load pressure control system was employed and modified .The apparatus consists of pneumatic Actuator with a maximum frequency upto 10Hz and capable of dynamic loading from ±10N to ±4kN, control panel to apply confinement, back pressure and vacuum and automatic data logger connected to computer system. The mould used for the sand sample is shown in Fig. 4(a) and after removing the mould, the sample placed in pedestal before inserting confining chamber is shown in Fig 4(b). A volume measurement system made up of a closed burette connected to the drainage value was added to the typical triaxial configuration.

4.1 Specimen Preparation and testing procedure: Each test specimen, 50mm in diameter and 100mm high had prepared with wet pluviation technique in a split mould not attached to triaxial cell. Liquid nutrient medium containing denitrifying was placed into the mould in the case of the treated sample.

Back pressure and cell pressure were increased to 70kPa and 75kPa in 10kPa increments to saturate the sample. The B-value was more than 0.95 at a back pressure of 70 kPa, indicating that the sample was saturated. The material was then isotopically consolidated at confining pressures of 50 kPa, 75 kPa, and 100 kPa. After that, the samples were sheared monotonically/dynamically in undrained conditions at 0.5 mm/min for static testing and at loads equal to 0.1, 0.2, 0.3, and 0.4 for cyclic loading. Both treated and untreated samples undergo testing for evaluation and comparison. The volume measurement instrument can determine how much water is expelled. For five days leading up to the reaction, the volume of the expelled liquid was measured to determine the degree of saturation by phase relationship.

(a) (b)

Fig. 4. (a) Mould used for the test (b) Sample installation on triaxial apparatus

5. Batch test on soil and gas production estimation

The initial substrate concentrations, reaction stoichiometry, pore pressure, and the type and solubility of the gas produced all affect how much gas can be produced at the maximum rate. If all of the nitrate is completely converted to nitrogen gas, as required by the reaction's stoichiometry, each mole of nitrate produces a half mole of nitrogen gas. In coarse-grained soils, gas pressure is believed to be almost equivalent to pore liquid pressure (Paassen et al., 2018). Henry's law, which states that under equilibrium circumstances, the ratio between a gas's dissolved concentration in water and its partial pressure in the gas phase remains constant, governs a gas's solubility.

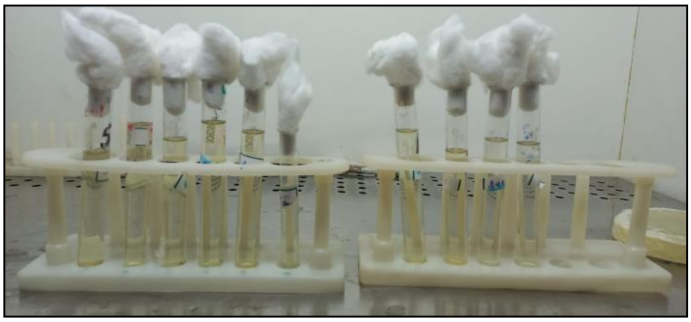


Fig. 5 Cultures inoculated with various initial nitrate conc

The inoculated cultures were then stored at $35^{o}C$ for 24hrs and then applied into the soil sample taken in beaker and saturated, mixed thoroughly and kept closed to avoid any escape of water (Fig.5) and maintain anaerobic conditions. Samples were taken after 24hr intervals for 8 days and extracts were tested for nitrate levels. The variation in concentration, % gas saturation and pH values for first eight days is given in Table 2.In the table C^{tot}_{N2} is the total produced mol-N2 per L of liquid; C_{N2}^{aq} is the dissolved concentration of N_2 in mol-N_2 per L liquid; n_{N2}^{g} is the amount of N_2 gas in moles; V_g is the total volume of gas and S_g denotes the % gas saturation.

Table 2. Calculated gas volumes as percentage of pore volume for each initial nitrate concentration applied

Initial Conc.	Nitrate Consumed	C_{N2}^{tot}	C_{N2}^{aq}	n^{g}_{N2}	Vg	S_g (%) (Gas Saturation)	pH
125.75	0.94	0.42	6.48E-04	50.90	1.24	4.45	8.67
146.58	1.03	0.47	6.48E-04	55.78	1.36	4.87	8.46
173.66	1.15	0.52	6.48E-04	62.29	1.52	5.44	8.77
198.66	1.25	0.56	6.48E-04	67.71	1.66	5.91	8.73
229.91	1.36	0.61	6.48E-04	73.68	1.80	6.43	8.65
246.58	1.42	0.64	6.48E-04	76.93	1.88	6.72	8.63
275.75	1.56	0.71	6.48E-04	84.52	2.07	7.38	8.76
300.75	1.7	0.77	6.48E-04	92.12	2.25	8.04	8.81

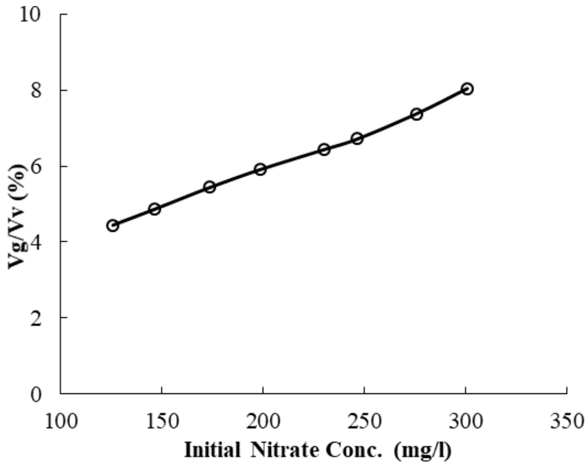


Fig. 6 Change in gas volume with initial nutrient content

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Fig.7. shows the gradual decrease in nitrate concentration as fraction of applied concentration with days for all initial concentrations used in the study. Maximum change was observed in 2 to 3 days while the rate of reaction allowed down by 5 days and stabilized by 7 days

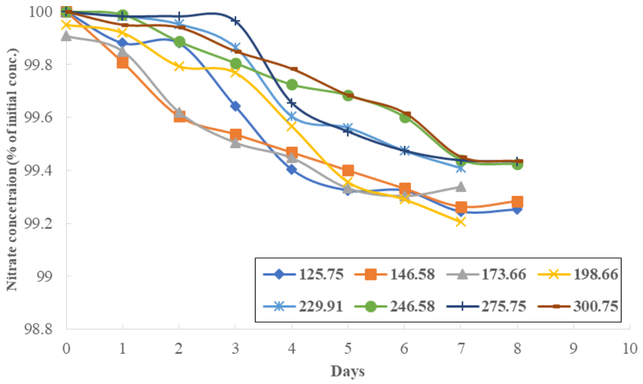

Fig. 7. Change in the nitrate concentration with time for each initial nutrient content

6. Results and Discussion

6.1Undrained response under static loading

Fig. 8**.** shows stress strain response of sample at 30% relative density before and after microbial treatment. A comparison of excess pore pressure development is shown in Fig .9**.** For treated and untreated sand sample. Using the first two letters, samples are designated as treated (TR) and untreated (UN). The third letter designates the sample's state "L" and the last component designates the sample's confining pressure in kPa.

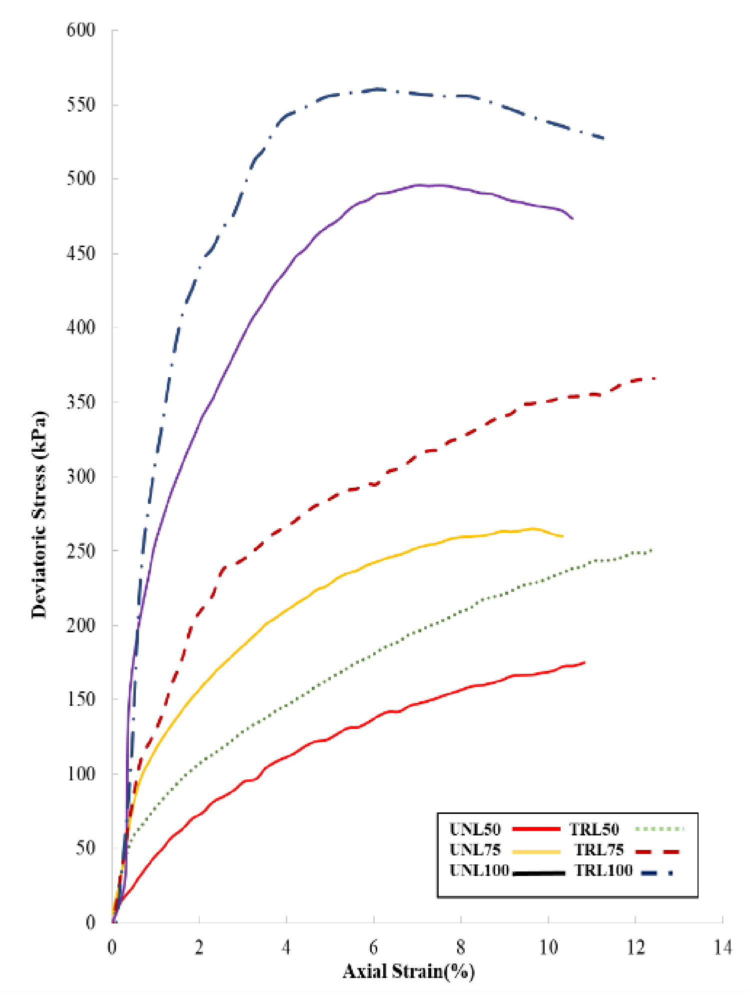

Fig. 8. Deviatoric stress v/s strain response of loose sands under static loading

As the specimen has enough contractive behavior it liquefied because of zero effective stress before reaching high axial strains for all effective confining pressure applied for untreated samples. Specimen exhibited constant deviatoric stress beyond axial strain of about 7%. The deviatoric stress attained was 1.4 times higher after treatment for 100kPa confining pressure .Generated gas bubbles has replaced few pore volumes of water and reduced the effective stresses within the system, thereby pore pressure generation was also inhibited with Ru value stabilized at 0.4 to 0.3 at same axial strain.

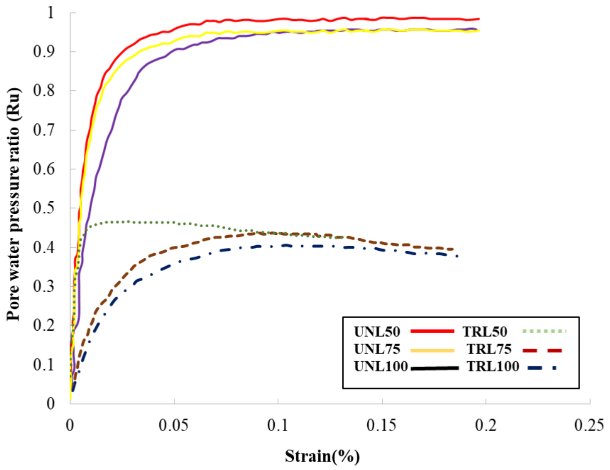

Fig. 9. Excess pore pressure v/s axial strain response of loose sands under static loading

6.2 Cyclic Triaxial test results

For liquefaction study, cyclic load was applied to the samples at 30% relative density for confining pressure of 75kPa. For the required stress ratio, the size of the cyclic load to be applied is estimated. The ratio of the intended deviator stress to the effective consolidation is known as the desired stress ratio (SR). The load to be applied calculated form desired Cyclic stress ratio according to the equation:

$$Load(kg) = 2 * CSR * \sigma 3c * A \qquad (5)$$

CSR denotes Cyclic Stress Ratio, $\sigma 3c$ denotes effective confining pressure, Ac denotes area of Cross-section of sample

For sand specimens with a 30% relative density and a 75 kPa effective confining pressure, Fig. 10**.** plots the excess pore pressure ratio with cycle number for various stress ratios. When the sand samples were subjected to cyclic load in a loose condition, they produced an excess pore pressure that, for CSR values of 0.2, 0.3, and 0.4, respectively, reached the effective confining pressure at 11, 18, and 24 cycles. The excess pore pressure ratio was maintained at 0.7, 0.4, and 0.3 for the same number of cycles after 5 days of microbe treatment**.** Therefore these findings suggested that microbial desaturation could lower the liquefaction potential of loose sands.

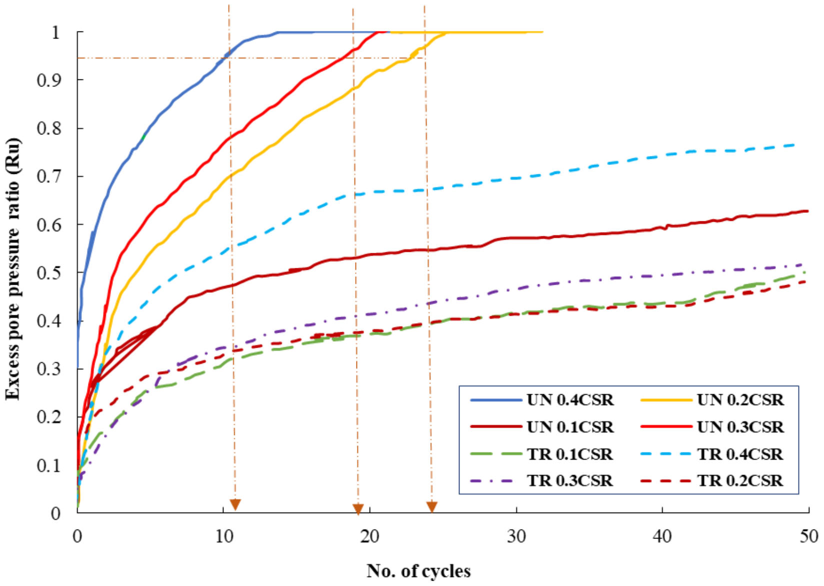

Fig. 10. Excess pore pressure ratio with number of cycles for loose sands under cyclic loading at different loads

7. Conclusions

A laboratory study was performed to study the effects of microbial treatment on strength and liquefaction behavior of soils. For this study, sandy soil was collected and characterized to be in liquefiable range. Microbes were cultured and maintained for 30 days before each sub-culturing at 4^oC. From the lab tests the few conclusions are drawn. The selected microbe was cultured and preserved in incubator with minimum contamination and colonies were able to produce the reductase enzyme necessary for nitrification process. Optical density of culture was maximum in day 2 suggesting the time of injection into soil after subculturing for maximum efficiency of the process. Batch tests indicates the saturation level can be controlled by initial nitrate concentration applied in the soil. For loose sands, increase in peak effective stress by 43 % indicates undrained strength under compression loading was enhanced. Maximum excess pore pressure was much lower after treatment but stabilized at same axial strain rate of 0.1%. In dynamic tests, the sand liquefied for applied CSR above 0.1 for all confining pressures. After treatment, excess pore pressure ratio was limited to 0.45 for low CSR and 0.65 for CSR=0.4. The pore water pressure was reduced substantially due to desaturation particularly in loose sands under low confining pressures with reduced saturation up to 82%. Therefore, it can be concluded that this method is effective in developing resistance against liquefaction.

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