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Soil Stabilisation by Microbial-Induced Calcite Precipitation (MICP): Investigation into Some Physical and Environmental Aspects

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ABSTRACT

This work investigates an emerging and promising soil stabilisation method known as bio-cementation using microbial-induced calcite precipitation (MICP). MICP utilises bacteria to hydrolyse urea to give carbonate ions which react with a calcium-rich solution (i.e. calcium chloride) to produce calcium carbonate (calcite) that binds the soil particles together leading to increased soil strength and stiffness. In this paper, the effectiveness of bio-cementation of silica sand under different environmental and physical conditions was investigated including the initial soil density, temperature and pH of soil. A set of laboratory tests were conducted including soil permeability, unconfined compression strength and determination of calcium carbonate content. The results indicate that bio-cementation is more effective for sand of high initial density. The results also demonstrate that although the calcium carbonate production was facilitated at an elevated temperature of 50°C, the build-up of strength was less efficient than at room temperature. Also alkaline (pH 9.5) or acidic (pH 3.5) conditions were adverse to strength development. Sufficient permeability was retained by all bio-cemented samples, which indicates good drainage ability that allows rapid dissipation of the excess pore water pressure upon loading.

A new promising and innovative modification of MICP treatment was also evaluated using the seawater as a replacement for one of the reactants in production of calcium carbonate. This new process provides a high potential for using bio-cementation in maritime environment for applications such as coastal erosion prevention. Treatment using the seawater to replace the calcium chloride has resulted in stabilised soils that exhibit reasonable strength and efficient crystal formation, which confirms the viability of the proposed seawater process for bio-cementation.

Keywords: Microbial-induced calcite precipitation (MICP), soil stabilisation, bio-cementation

1 INTRODUCTION

Soil stabilisation is often a necessary part of civil construction because stability of structures is related to the foundation on which they rest. The goal of soil stabilisation is the transformation of soil, which has insufficient strength for its desired use, into a stable foundation. With roughly 40,000 projects that require soil improvement world-wide each year, adding up to AUD$6 billion (DeJong et al. 2010), geotechnical engineers are challenged in providing workable ground for the structures. The most commonly used ground improvement methods currently available are: soil replacement, mechanical stabilisation by compaction, drainage by soil consolidation and chemical treatment. Among these methods, chemical treatment is widely employed, where the mineralogical structure of soil is altered by chemical additives to improve the physical and mechanical properties of soil. The key principle of chemical treatment involves injecting synthetic grouts (e.g. lime, cement, fly-ash or slag) into the soil pore space to bind the soil particles together, thereby increasing soil strength and stiffness (Hausmann 1990). However, chemical grouts are increasingly under the scrutiny of public policy as they are environmentally detrimental (Charles 2002; DeJong et al. 2010). For instance, in 1974, an acrylamide grout was leached into the surrounding water sources in Japan and has led to five cases of water poisoning, resulting in the ban of nearly all chemical grouts (Karol, 2003). Moreover, initiatives in certain countries such as the US proposed to ban most synthetic grouting materials (DeJong et al. 2010). Another popular chemical additive is Ordinary Portland Cement (OPC), which is commonly used for its simplicity of application and compatibility with any soil type. However, Ariyanti et al. (2011)

mentioned that up to 7% of the world CO_2 emission is currently caused by the manufacturing of OPC. This is due to the calcination of raw materials (e.g. calcium carbonate, $CaCO_3$) and the associated high energy consumption (Ali et al. 2011). Therefore, the need for developing a new soil stabilisation method that is economically viable, environmentally sustainable and can achieve optimum performance continues to grow.

In this paper, a new emerging and promising soil stabilisation technique that has recently gained interest by many researchers and geo-engineers is introduced and presented. The technique is called bio-cementation using microbial-induced calcite precipitation (MICP), which has proved its sustainability and capability to alter and improve most soil engineering properties (Cheng and Cord-Ruwisch, 2014). This technology utilises the metabolic pathway of ureolytic bacteria to form $CaCO_3$ precipitation throughout the soil matrix resulting in an increase in strength and stiffness while maintaining adequate permeability. Compared to OPC cemented soil, MICP treatment shows similar strength while retaining a significant higher permeability of 10-100 times (Cheng et al. 2013). This paper further investigates on MICP and its practicality towards its usage in ground improvement, which will be achieved by examining some physical and environmental parameters that may affect the effectiveness of bio-cementation for ground improvement. The influence of the initial soil density, degree of temperature at which bio-cementation occurs and initial pH of soil will be investigated. The paper also investigates the possibility of using the seawater as a calcium source for bio-cementation, which can significantly reduce the cost of bio-cementation treatment.

2 EXPERIMENTAL PROGRAM

2.1 Material Tested

Industrial pure silica sand provided by Cook Industrial, Minerals Pty. Ltd. Western Australia was used in this study, which has the grain size distribution shown in Figure 1. The sand is classified as poorly graded according to the Unified Soil Classification System (ASTM 2011) with a particle size of 0.425 mm. This sand was used as it exhibits undesirable engineering properties for most geotechnical engineering applications.

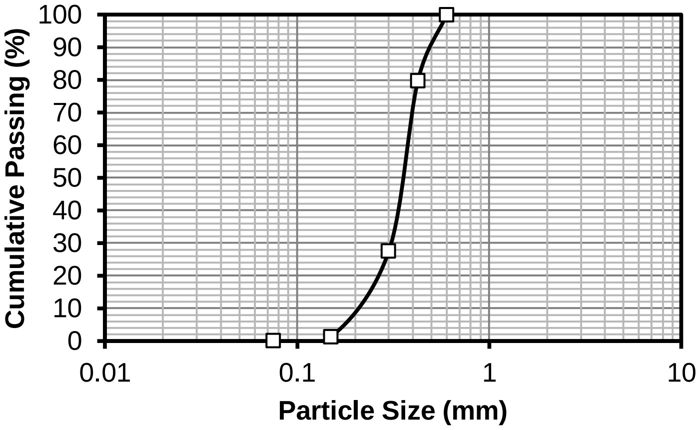

Figure 1. Particle size distribution curve of the sand used

2.2 Bacteria Culture and Cementation Solution

The urease active strain used in this study was *Bacillus sphaericus* (MCP-11) (DSM 23526, available from DSMZ, Germany). The MCP-11 strain was cultivated under sterile aerobic batch condition in yeast extract based medium (20 g/L yeast extract, 0.17 M ammonium sulphate, 0.1 mM Ni^{2+}, pH 9.25). After 24 hours of cultivation at 28^oC, the bacteria culture was collected and stored at 4^oC prior to use. The optical density (OD_{600}) of the culture varied between 0.6–1.0, and the urease activity was approximately 5 U/mL (1 U = 1 μmol urea hydrolysed per min). The cementation solution contained a mixture of 1 M calcium chloride (111 g/L) and 1 M urea (60 g/L). For the seawater experiments, the cementation solution was prepared by adding 10 mM urea (0.6 g/L) into artificial seawater, which consisted of different concentrations of salts (g/L): NaCl (23.9), Na_2SO_4 (4.0), $CaCl_2{\cdot}2H_2O$ (1.5), $MgCl_2{\cdot}6H_2O$ (10.8), KCl (0.7), $NaHCO_3$ (0.2), KBr (0.1), and H_3BO_3 (0.03).

2.3 Preparation of Sand Columns

Sample preparation of compacted and uncompacted sand columns was made by the packing of sand into polyvinyl chloride (PVC) columns of 180 mm in height and 45 mm inner diameter. The final dry densities of the compacted samples ranged between 15.9-16.1 kN/m^3, whereas they were between 14-14.2 kN/m^3 for the uncompacted samples.

2.4 Bio-cementation Treatment

Bio-cementation using MICP employs an introduction of aerobically cultivated ureolytic bacteria, i.e. *Bacillus pasteurii* (also known as *Sporosarcina pasteurii*), into soil with a highly active urease enzyme that is used to catalyse the hydrolysis of urea. From this reaction, ammonium and carbonate are produced, as follows:

$$CO(NH_2)_2 + 2H_2O \rightarrow 2NH_4^{+} + CO_3^{2-} \qquad (1)$$

In the presence of calcium source (in most cases calcium chloride) the produced carbonate ions react with the calcium ions to produce $CaCO_3$ precipitated crystals, as follows:

$$Ca^{2}+ + CO_3^{2-} \rightarrow CaCO_3 \qquad (2)$$

The produced precipitated $CaCO_3$ cements the adjacent soil particles together, leading to increased soil strength and forming cemented sand that is very similar to that of calcareous rocks. The MICP process simulates natural digenesis from sand to sandstone, only within a short time instead of million years. It should be noted that the bio-cementation treatment of all samples treated in this study was conducted under fully saturated conditions, to maintain consistency and eliminate possible variations. At the beginning of treatment, the columns were up-flushed with 0.5 void volume (i.e. pore volume of the sand column) of bacterial culture, followed by 0.5 void volume of cementation solution. Then the sand columns were left at the room temperature of 25^{o}C for 24 hours to allow the bacteria to adhere with the sand particles. The columns were then repeatedly treated with one void-volume of cementation solution every 24 hours to achieve different amounts of $CaCO_3$ precipitation. Between each treatment of cementation solution, the sand columns were cured at the room temperature of 25^{o}C. The flow rate of all injections was kept constant at 1 L/hour.

2.5 Samples with Varied Temperature and Initial pH

In order to test the effect of curing of temperature on bio-cementation treatment, a series of sand columns was treated using the standard treatment procedure described in Section 2.4 except that the curing temperature was set at 50±1°C. On the other hand, the effect of the initial pH value of soil on bio-cementation treatment was investigated using two different chemical reactants, i.e. citric acid (pH = 3.5) and sodium hydroxide (pH = 9.5) so as to alter the initial pH of the sand columns by flushing 2 L of each solution into the soil. Then, the sand columns were fully loaded with the chemicals at the room temperature for 24 hours, followed by the standard treatment procedure described earlier.

2.6 Permeability, Unconfined Compressive Strength and Calcium Carbonate Content

The permeability tests were conducted using the constant head method according to the Australian Standards AS1289.6.7.1 (2001) before and after the MICP treatment. These tests measure the reduction in permeability in terms of the percentage of formed $CaCO_3$ crystals (i.e. gram calcite per gram of sand).

The unconfined compression strength (UCS) tests were conducted using the IPC Global Universal Testing Machine (UTM). The purpose of these tests is to compare the strength of the soil samples with respect to its $CaCO_3$ content and crystal formation. Prior to the UCS measurements, all soil samples were flushed with 2 L of tap water and submerged in 2 L of tap water for 24 hours, followed by air-dried process at 25°C for 24 hours. The machine used for the UCS tests was the UTM-25, and the load was applied at a constant rate of 1 mm/min.

The content of calcium carbonate of treated samples was determined by adding 2 mL of 2 M HCl solution into 1-2 g of dry sample, and then the volume of produced CO_2 of gas was measured with a U-tube manometer under standard conditions (25°C, 1 atm). A calibration was made with the analytical grade $CaCO_3$ powder in accordance with Cheng and Cord-Ruwisch (2012).

2.7 Microscopy Investigation

Microstructures of bio-cemented sand were studied under scanning electron microscope (SEM) (PHILIPS XL20 SEM). The microscopy investigation allows a better understanding of the crystal formation and bonding behaviour between the sand particles and $CaCO_3$ crystals.

3 RESULTS AND DISCUSSION

3.1 Initial Density and Grain Size Distribution

Figure 2 shows a comparison of obtained strength between compacted and uncompacted treated soils. It can be seen that the initial density of soil has a significant impact on the effectiveness of bio-cementation. For similar produced $CaCO_3$ content, the compacted samples (i.e. high initial density samples) have greater unconfined compressive strength than those of the uncompacted samples (i.e. low initial density samples). This is because the sand particles in the compacted sand columns are closer together than the uncompacted samples; hence, the $CaCO_3$ crystals are formed over a shorter distance to bridge the sand particles. This result is in agreement with that found by Tsukamot et al. (2013). In terms of permeability of treated soils, it was found that the permeability decreased with the increase of the $CaCO_3$ content, which is a general observation throughout the current study. However, it should be emphasised that there was no significant difference in the permeability reduction between the treated compacted and uncompacted sand samples.

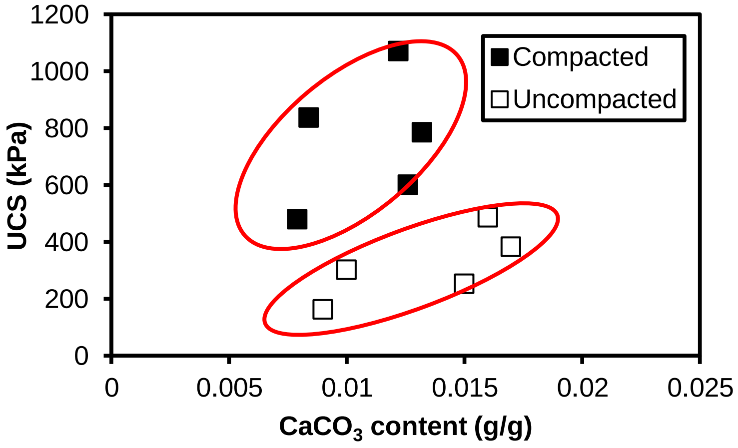

Figure 2. Comparison between compacted and uncompacted strength of treated samples

3.2 Temperature

Figure 3 shows the result of obtained UCS versus $CaCO_3$ content for samples treated at the room temperature (25°C) and at higher temperature (50°C). It can be seen that although about 3 times more $CaCO_3$ crystals were formed at higher temperature of 50°C, the treated samples exhibited about 60% less strength than the samples treated at the room temperature. This decreased strength (with even more amount of $CaCO_3$) at high temperature is interesting in terms of both the scientific and applied aspects. This finding implies that it is not the amount of produced $CaCO_3$ content that governs the strength of treated soils but rather the way the produced $CaCO_3$ crystals are formed. In order to use optimum cementation results for high temperature conditions, such as cementation of oilfield reservoirs, further investigation is needed to explain the formation of less effective crystals at the higher temperature. For example the rate of calcite formation may need to be slowed down by using lower bacterial numbers. It should be noted that there are a number of other factors that may affect the crystal formation, such as the urease activity, nucleation and oversaturation, but they are beyond the scope of this paper and are left for future study.

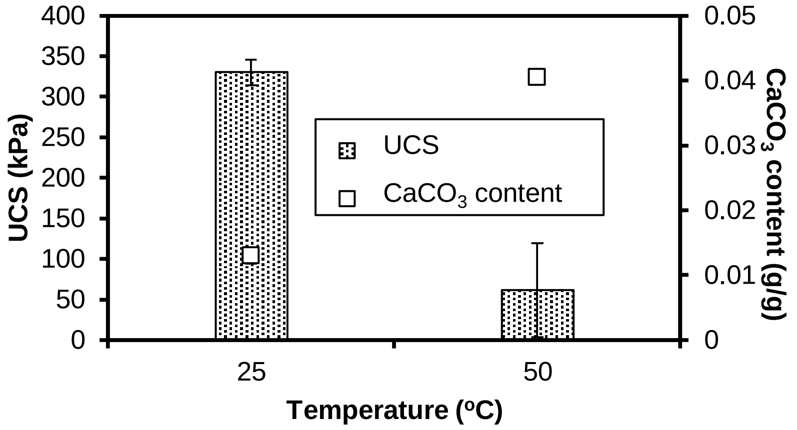

Figure 3. UCS and $CaCO_3$ content of samples treated at different temperatures

In order to further investigate the impact of temperature on the crystal formation of treated samples, SEM analysis was carried out and the results are shown in Figure 4. It was observed from the SEM images and $CaCO_3$ measurements that the amount of $CaCO_3$ precipitated at 50°C is much higher than that formed at low temperature of 25°C, as the high temperature promoted an increase in the urease activity. It was also observed that the crystals produced at higher temperature are relatively small in size (about 2-5 µm in diameter) and fully cover the surface of sand grains (see Figure 4, left column), where the crystals cannot contribute to the strength development. On the other hand, the samples treated at lower temperature produced smaller amount of crystals but of larger size of 15-20 µm (see Figure 5, right column), which can benefit the gap filling between the adjacent sand grains and thus efficiently contributing to the strength development.

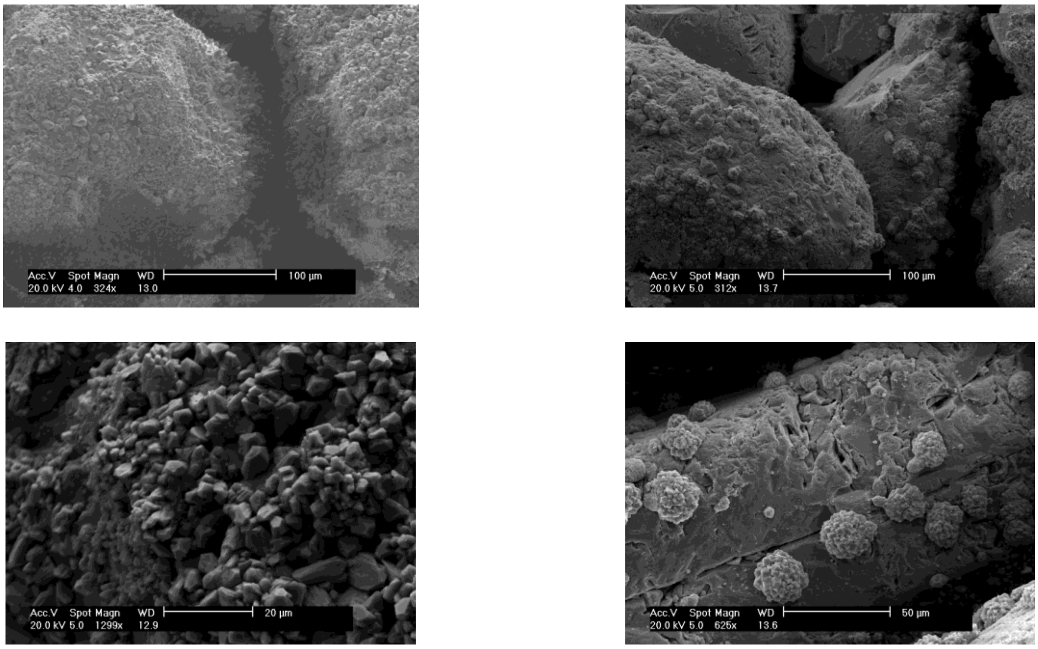

Figure 4. SEM of $CaCO_3$ crystals formed at different temperatures of 50°C (left) and 25°C (right)

3.3 Initial pH of Soil

As shown in Figure 5, the initial pH of soil has a significant impact on the final compressive strength of treated samples. It can be seen that both the acidity and alkalinity conditions have negative effects on the treated samples, resulting in a decrease in strength performance even in the presence of high content of $CaCO_3$ crystals. As mentioned by many researchers (e.g. Sanderson et al. 1996; McWhirter et al. 2002; Harkes et al. 2010), the pH value can influence the bacteria transport and adhesion, which is an essential factor for achieving homogeneously improved strength of treated soils. The initial pH can also affect the formation of crystals, as the solubility of $CaCO_3$ varies according to the pH value. This may be due to the urea hydrolysis reaction, which continuously produces a mixture of $NH_3/NH_4^+/CO_3^{2-}/HCO_3^-$ solution with a strong pH buffer capability. The effect of the initial pH of soil on bio-cementation requires further investigation to better understand the feasibility of bio-cementation for the Australian soils, where the pH may vary from 4 to 10.5 (De Caritat et al., 2010).

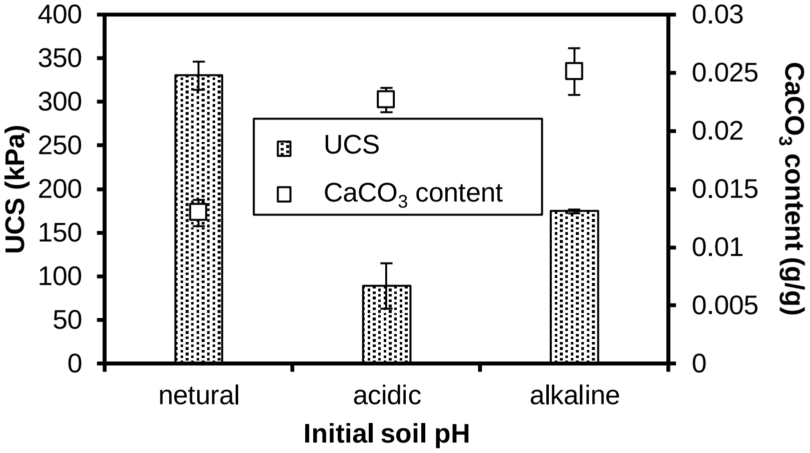

Figure 5. UCS and $CaCO_3$ content of samples treated with different initial pH

3.4 Seawater as Calcium Resource

The cost of the MICP process including bacterial cultivation, chemical usage, equipment and labour may prevent the progress of further commercial development of this emerging ground improvement technique. Consequently, an attempt is made in the current study to exploit the potential benefit of using the seawater in the bio-cementation process to reduce the cost of the MICP process, bringing soil bio-cementation closer to be a commercially and environmentally acceptable ground improvement alternative. The use of seawater as a relatively dilute calcium solution requires many subsequent treatment (flushes); however, each treatment can be completed within a shorter period of time (e.g. 6 hours for the current study) compared to the 24 hours needed for a single treatment using the method discussed earlier. The results of using the seawater in the MICP treatment are shown in Figure 6, which clearly demonstrate the feasibility of using the seawater as a chemical reagent for bio-cementation. It can be seen that the strength of the seawater treated samples significantly increases with the number of treatment (flushes) between 60 and 80, but slightly enhanced after 80 flushes. This can be attributed to the loss of bacterial activity after the long-term treatment process, which reduces the efficiency of the bio-cementation reaction that may be caused due to the encapsulated cells in the $CaCO_3$ crystals or extracellular urease enzyme decomposition. A similar decrease in bacterial urease activity during the bio-cementation process has been observed by other researchers (e.g. Whiffin et al. 2007; van Paassen et al. 2010), therefore, additional supplementation of urease active bacteria during the subsequent treatment is necessary. It should be noted that more work is still needed to investigate the feasibility of using this proposed method in situations of real world applications. This is because the injection method used in the current study may not be suitable for treatment of coastal cliffs or beach sand in large scale. In subsequent phase of this work, this issue will be one of the main foci of this ongoing research.

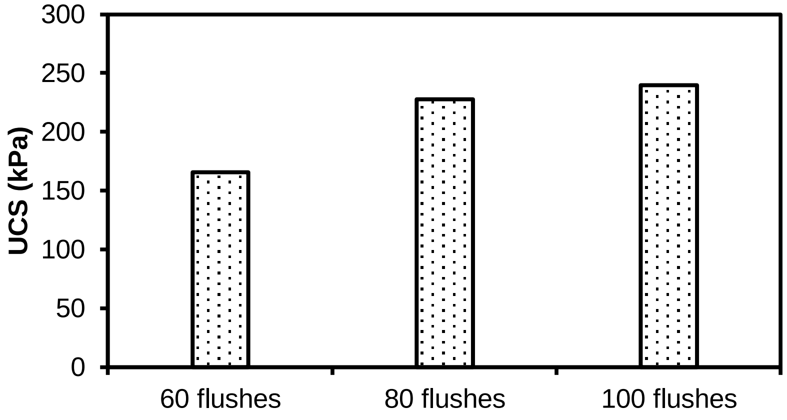

Figure 6. UCS of treated samples after different number of seawater treatment

4 CONCLUSIONS

The results presented in the current study demonstrated that bio-cementation using microbial-induced calcite precipitation (MICP) can significantly improve the engineering properties of silica sand with enhanced compressive strength and retained permeability. However, the efficiency of bio-cementation in improving the soil strength varied significantly according to the physical and environmental conditions. The compacted soil of high initial density (i.e. dense sand) was found to achieve greater compressive strength compared to the uncompacted sand of lower density (i.e. loose sand). The study also indicated that bio-cementation is able to process in different environmental conditions, such as extreme pH and high temperature. However, the compressive strength of treated soils varies significantly depending on the environmental conditions, with lower strength gained at hot temperature of 50^{o}C and at pH values different from the neutral pH of 7.

The results from this study also showed that it is possible to use the seawater as a chemical reagent for bio-cementation to replace the calcium chloride, and a significant strength improvement can be achieved after repeated treatments. This finding is interesting as it extends the application of bio-cement to broader areas, such as ground improvement in marine environments. Marine bio-cementation can potentially contribute to more sustainable human activities and significantly benefit society in areas of offshore and onshore infrastructure protection and maintenance, as well as coastline erosion prevention.

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