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Studies on injection method for sand solidification by microbial metabolism

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ABSTRACT

Studies were conducted about the influences on the solidification of sand by Microbial Carbonate Precipitation of various factors like the density of the nutrient salt, the methods of injection, the discharge in the continuous injection, and so on.As the result, it was verified desirable that the period is instituted within the period in the microbial functions is active. It seems advantageous of solidification that the density of the nutrient salt and the discharge of the injection are settled considering the effective period of the injection. It is considered the injection of the thin nutrient salt with much volume and the intermittent injection can be effective of not solidification but also the mitigation of the environmental influence.

1. INTRODUCTION

In this research, microbial carbonate precipitation (MCP), a ground improvement technique that uses microbial metabolism (Whiffin 2007) was focused. In our past investigation (Inagaki 2012), it was understood that the discharge, the staying time, and so on of the nutrient salt can have influence on solidification.

Studies were conducted about the influences on the solidification of sand by Microbial Carbonate Precipitation of various factors like the density of the nutrient salt, the methods of injection, the discharge in the continuous injection, and so on. The unconfined compression tests were carried out to specimens prepared by the injection of the culture and the nutrient salt.

2. EXPERIMENTAL METHOD

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Specimens were prepared with the reactions as Eq.(1), Eq.(2) and Eq.(3) which was ureolysis by " *Sporosarcina pasteurii* "(ATCC 11859) .

(Ureolysis) $CO(NH_2)_2 + 3H_2O \rightarrow 2NH_4^+ + 2OH^- + CO_2$ (1)

(Precipitation of calcium carbonate)

$$CO_2 + H_2O \rightarrow HCO_3^- + H^+ \quad (2)$$

$$HCO_3^- + Ca^{2+} + OH^- \rightarrow CaCO_3 + H_2O \quad (3)$$

Unconfined compression tests were carried out prepared the specimens. After tests, the amounts of $CaCO_3$ precipitated in the specimens were measured. The consecutive experimental procedure were shown Fig.1.

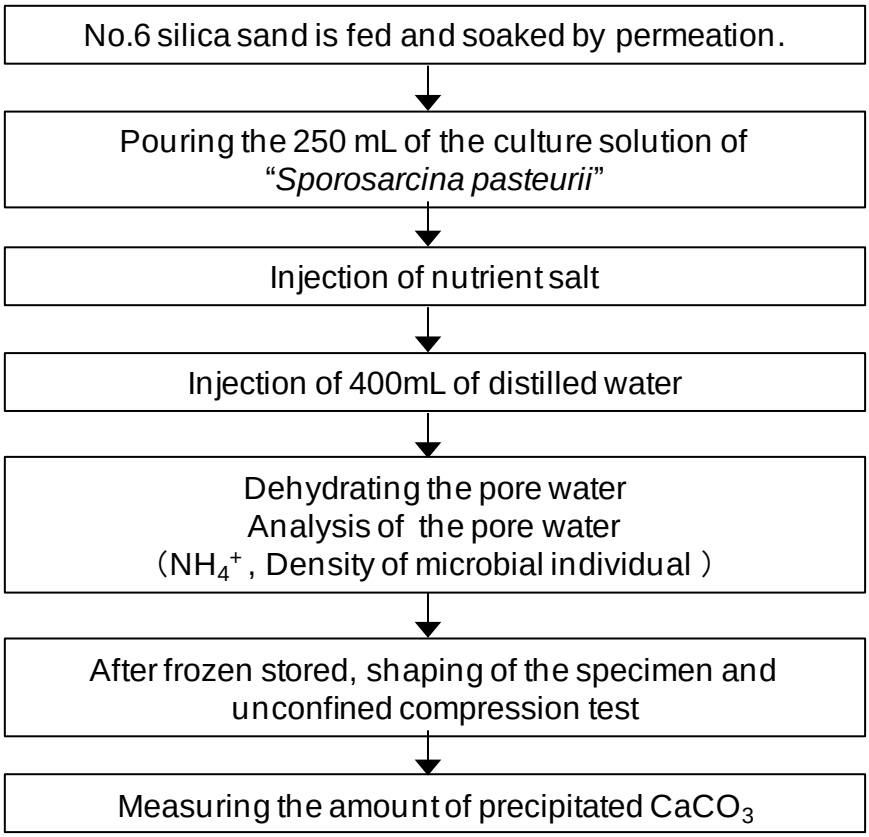


Fig.1 Experimantal procedure

Specimens were prepared in a PVC mold with a diameter of 5 cm and a height of 15 cm (Fig.2).No.6 silica sand (1.662 g/cm^3 for maximum dry density ρ_{dmax} and

1.376g/cm^3 for minimum dry density ρ_{dmin}) was fed into the mold through the aerial fall and soaked by permeation with distilled water directly under the filter layer. To prepare the solidified specimens, 250 mL of a culture solution of *Sporosarcina pasteurii* was fed from above the mold after the No.6 silica sand was soaked.

In the next, a nutrient salt was injected from above the mold. After the nutrient salt of the volume prescribed was injected, 400 mL of distilled water was injected to wash away the components of the nutrient salt in specimens. The injection of these was carried out under the condition which the head of water was settled, as shown Fig.2. When the nutrient or the distilled water was injected, the water from the previous injection and filling the voids was rejected.

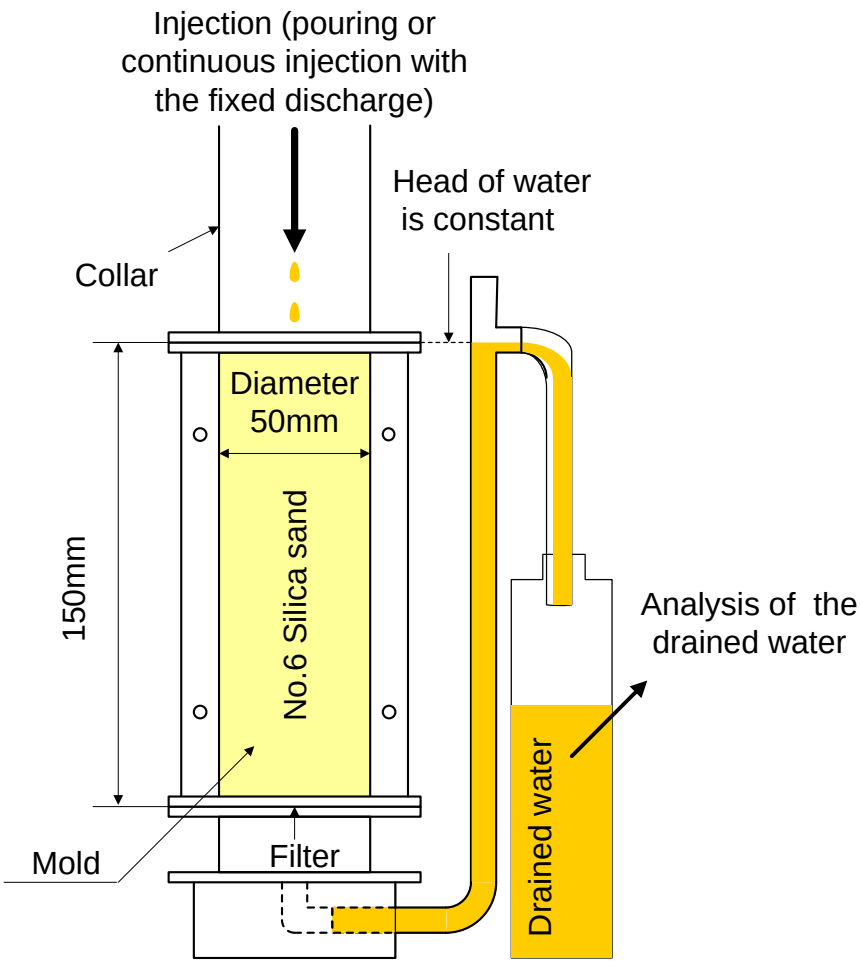


Fig.2 Preparation of specimens

The basic composition of the nutrient salt was as Table.1. The injection conditions were as shown Table.2.

Table.1 Composition of nutrient salt (per liter of distilled water)

Component	Quantity
$CaCl_2$	0.5mol=55.49g
$CO(NH_2)_2$	0.5mol=30.03g
NH_4Cl	10g
Nutrient broth	3g

Table.2 The condition of the injection

Case No.	Method	Discharge	Injection of the nutrient salt			The days to inject the distilled water (days)	The period of the injection (days)
			Density	Days (days)	Volume (mL)		
Case 1	Continuous	0.07 mL/min	1	4	400	4	8
Case 2	Continuous	0.04 mL/min	1	8	400	8	16
Case 3	Continuous	0.07mL/min	0.5	8	800	4	12
Case 4	Continuous	0.14 mL/min	1	2	400	2	4
Case 5	Continuous	0.28 mL/min	1	1	400	1	2
Case 6	Continuous	0.14 mL/min	0.5	4	800	2	6
Case 7	Continuous	0.28 mL/min	0.5	2	800	1	3
Case 8	Intermittent	200 mL/an pouring	0.25	8	1600	2	10
Case 9	Intermittent	200 mL/an pouring	0.5	4	800	2	6
Case 10	Continuous after replacement	0.14mL/min after replacement by 200 mL	0.5	3	800	2	5
Case 11	Continuous	0.14mL/min	0.5	4	800	2	6

About the method at Table.2, “continuous” means the nutrient salt or the distilled water would be injected continuously with fixed discharge. “Intermittent” means that pouring 200 mL of the nutrient salt into the mold and leaving 1 day would be repeated settled times. “Continuous after replacement ” means that after pouring 200 mL of the nutrient salt into the mold to reject the water filling the voids, the surplus 600 mL of the nutrient salt would be injected continuously with fixed discharge. “Discharge” shows that the discharge settled by the quantitative pump in the continuous injection, and that the volume of the nutrient salt to be poured at once in the intermittent injection.

“Density” means the relative density to the density of the nutrient of the composition as shown Table.1.For example, “density 0.5” means that by half of the 4 kinds of components as shown Table.1 dissolved to 1 L of distilled water. “Injection volume” is the total volume of the injected nutrient salt. In this experiment, the whole

amount of the components was same on the all test cases. The injection volume was proportioned inversely to the density.

"The days to inject the nutrient salt" is the days from the beginning of injection of the nutrient salt to the beginning of injection of distilled water.

"The days to inject the distilled water" is the days from the beginning of injection of 400 mL of the distilled water to the time to dehydrate the specimen. It is equal to the days that takes to inject 400 mL of the distilled water at the same discharge as the injection of the nutrient salt in the case of the continuous injection.
In the intermittent injection, it was 2 days as the days to leave after pouring 400 mL of the distilled water.

"The period of the injection" is equal to the total of the days to inject the nutrient salt and the days to inject the distilled water.

Next to the end of the injection of the distilled water, Each specimen was dewatered by the suctioning and the density of NH_4^+ and the culture individual of the water drawn out. After each specimen was frozen and stored as contained in the mold, these were taken out of the mold and swiftly molded into a shape with a diameter of 50 mm and a height of 100 mm while still frozen. The upper and lower parts of each specimen with a height of 25mm were cut off. Each specimen was quickly placed in the testing machine and left for two hours to thaw. The unconfined compression tests were conducted to the thawed specimen.

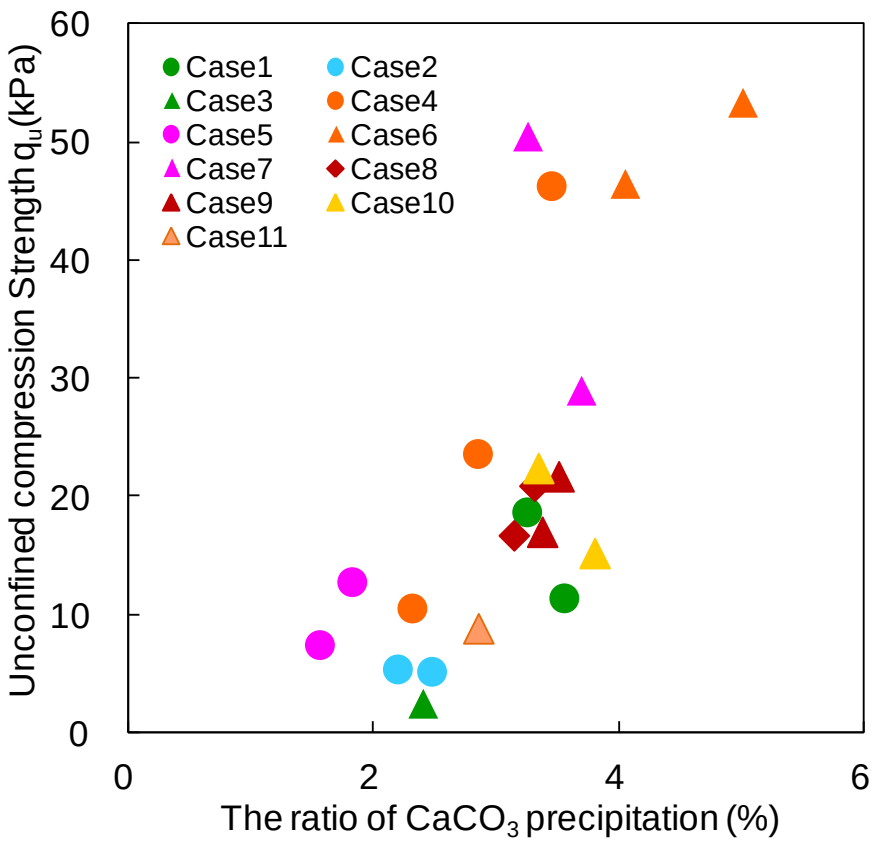


Fig.3 The relationship between $CaCO_3$ precipitation ratio and q_u

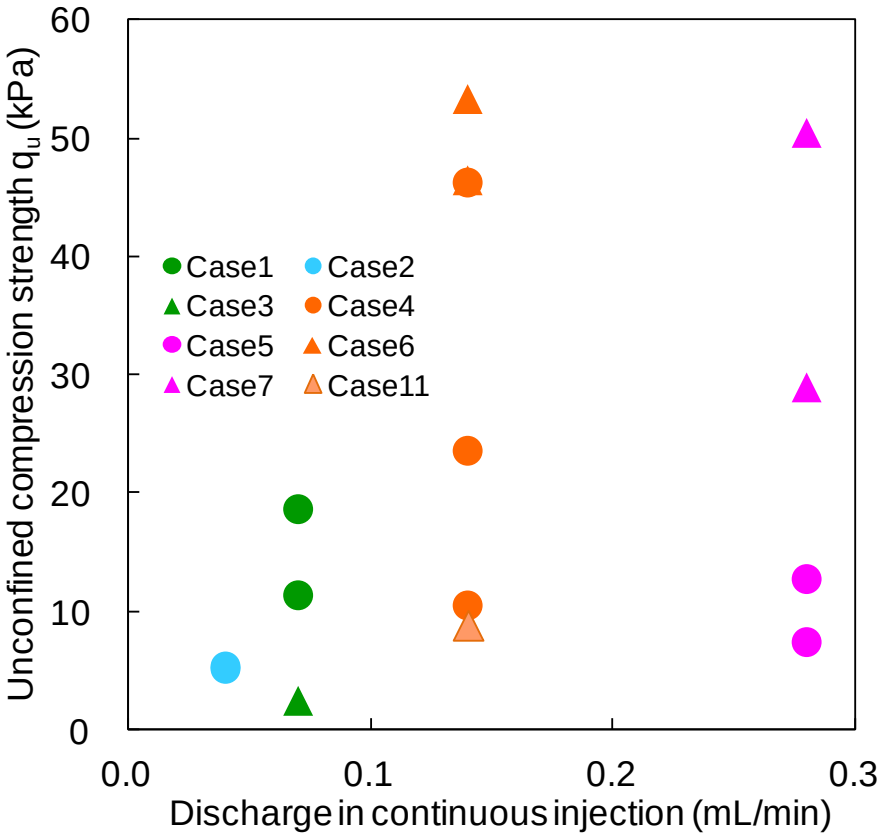


Fig.4 The relationship between the discharge and q_u at the continuous injection

After the completion of the unconfined compression tests, the amount of precipitated $CaCO_3$ in each solidified specimen was calculated based on dry mass changes resulting from the decomposition or elution of $CaCO_3$ by hydrochloric acid.

The above series of tests in 11 cases as shown Table.2. In each case, 2 or 3 specimens were prepared taking un-homogeneity of tests into consideration.The studies were investigated for the results on the specimens, which the problematic matters like a leak did not cause at.

3. EXPERIMENTAL RESULTS

3.1 The Relationship between $CaCO_3$ Precipitation and Unconfined Compression Strength

Fig.3 shows the relationship between $CaCO_3$ precipitation and unconfined compression strength q_u (hereafter q_u). $CaCO_3$ precipitation is expressed as the ratio of $CaCO_3$ precipitate to the mass of sand (hereafter the $CaCO_3$ precipitation ratio).

There were the linear relationship between the $CaCO_3$ precipitation ratio and q_u. With the $CaCO_3$ more, q_u tended to increase.

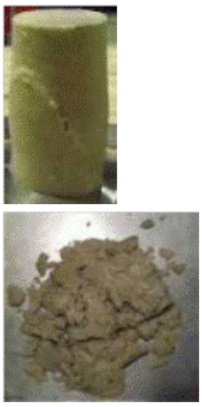
Case 2 (0.04 mL/min)	Case 1 (0.07 mL/min)	Case 4 (0.14 mL/min)	Case 5 (0.28 mL/min)
q_u=5.31kPa	q_u=18.61kPa	q_u=10.47kPa	q_u=7.37kPa
			

Fig.5 The appearance of the specimen after the unconfined compression test
(The example of the density 1)

3.2 The Influence of the Discharge

Fig.4 shows the relationship between discharge and q_u at the continuous injection, about from Case 1 to Case 7, and Case 11. Fig.5 shows the appearance of the specimens after unconfined compression tests in the various the condition of discharge. The cases the nutrient salt of density 1 given are instanced.

Through specimens were prepared at the same case, q_u was different by specimens (Fig.4). Moreover the state was different at every part in a specimen (Fig.5). It considered that q_u was be influenced by this un-homogeneity. But through these matters were considered, the specimens given the nutrient salt with discharge 0.14 mL/min had comparatively more q_u. In Case 2 (discharge 0.04 mL/min) and Case 5 (discharge 0.28 mL/min) , there were not the lumps of sand in the specimen after the unconfined compression tests and the specimen had comparatively less q_u. In Case 2, the surface of sand began to be stopped and the injected nutrient salt settled in the collar without permeation to sand when about 4 days passed from the beginning of the injection of the nutrient salt. In Case2, the amount of permeated nutrient salt in practice was only 200 mL against expected 400 mL. It considered that the supply of the components and the precipitation of $CaCO_3$ had not progressed sufficiently. In Case 6, the whole of injected nutrient salt permeated to sand, but it was considered that the components of the nutrient salt flowed out without enough use.

It is considered that the appropriate condition of discharge to not stop and remain in the void for enough time to precipitate $CaCO_3$ is necessary.

3.3 The Influence of the Period of the Injection

Fig.6 shows the relationship between the period of injection and q_u. The whole amount of components given was the same in all cases, but the period of the injection

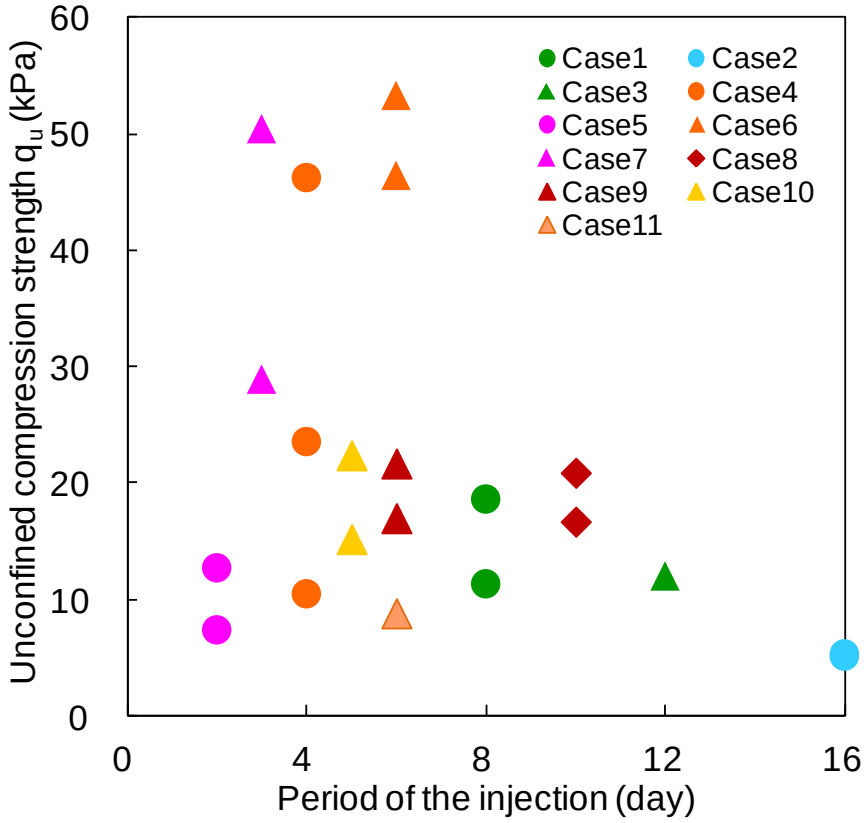


Fig.6 The relationship between the period of the injection and q_u

were different at every case depend on the density, the volume, the days to inject the distilled water.

According to Fig.6, the tendency to be more q_u comparatively in the case which the period of the injection was about from 3 days to 6 day was verified. In Case 5 which has only two days as the period of the injection, $CaCO_3$ precipitation ratio and q_u was less comparatively as shown Fig.3. Presumably, this result is because the time to use the components to precipitate $CaCO_3$ in the void was not obtained sufficiently, as stated above 2).

Conversely in the cases which have the long period of the injection, q_u was less comparatively, too. It was verified that *Sporosarcina pasteurii* would be most active within about 8 days at first on the past study (Inagaki 2014). It seems the reactions as Eq.(1), Eq.(2) and Eq.(3) became dull because of the decline of microbial functions in the cases which have the long period of the injection.

It is considered desirable that the density of the nutrient salt and discharge would be instituted to put the period of the injection within the period in which the microbial functions is active.

3.4 The Influence of the Density of the Nutrient Salt

In the continuous injection, comparisons between the 2 Cases had the same discharge and the different density of the nutrient salt were investigated on Case 1 and Case 3, Case4 and Case 6 and Case 5 and Case 7.

As stated above Fig.4, q_u in the case of the density 1 (Case 1) was more than it in the case of the density 0.5 with the discharge 0.07 mL/min. Conversely with the discharge 0.14 mL/min and 0.21 mL/min, q_u in the case of the density 0.5 (Case 6 and Case 7) was more than it in the case of the density 1. As shown Fig.7, in Case 1, Case 6 and Case7 the bigger clods remained after the unconfined compression tests than in Case 3, Case 4 and Case 5. If the discharge was faster than 0.14 mL/min, in the case of density 0.5 were effective for the solidification than in the case of density 1.

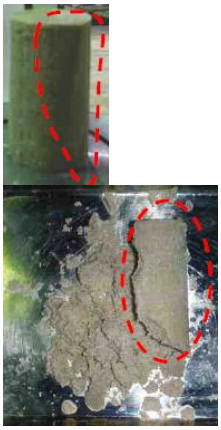
Discharge 0.07mL/min			
Case 3	Case 1		
Density 0.5	Density 1		
q_u=6.29kPa	q_u=18.61kPa		
			
Discharge 0.14mL/min		Discharge 0.28mL/min	
Case 6	Case 4	Case 7	Case 5
Density 0.5	Density 1	Density 0.5	Density 1
q_u=53.29kPa	q_u=10.47kPa	q_u=28.91kPa	q_u=7.37kPa
			

Fig.7 The appearance of the specimen after the unconfined compression test
(By the density and the discharge)

Fig.8 instances the change of the density of NH_4^+ in the pore water drained from the void on from Case 4 to Case 7. NH_4^+ occurs with ureolysis shows microbial functions in the void. In the all cases from Case 4 to Case 7, NH_4^+ was found and it seems that ureolysis progressed.

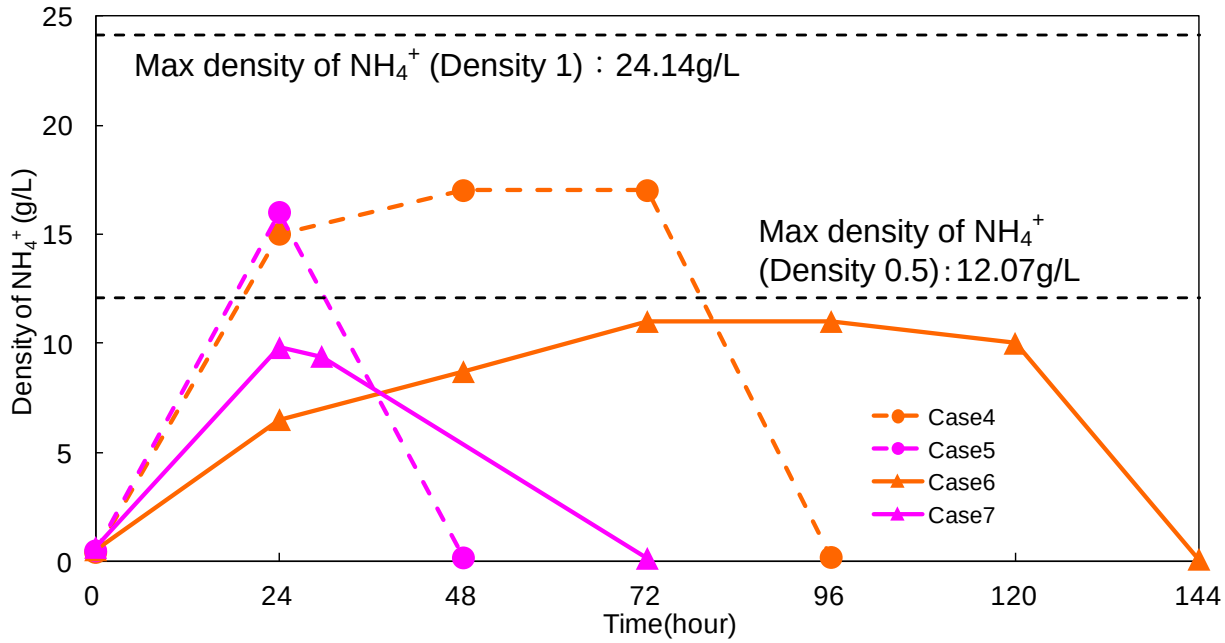


Fig.8 The change of the density of NH_4^+ in the period of the injection

As the difference depends on the density of the nutrient salt, the comparisons were made between Case 4 and Case 6 and between Case 5 and Case 7 respectively. In the Case 6 or Case 7 which is the density of nutrient salt 0.5, the density of NH_4^+ was thinned because the density of urea given was half. But if attention is paid to the ratio density of NH_4^+ to the density of NH_4^+ in the case all urea dissolved, it seems that ureolysis progresses well in Case 6 or Case 7 than in Case 4 or Case 5. In these cases, it is considered that q_u increased, because ureolysis and $CaCO_3$ precipitation continued longer and more regularly than in the cases of density 1.

It is required that the environmental influence caused by injections of the nutrient salt is decreased when the practical ground is solidified. It is considered that the injection of the thin nutrient salt with much volume can be effective to decrease the density of the ions from the nutrient salt.

3.5 The Influence of the Method of the Injection

As the influence of the method of the injection of the nutrient salt, the results of from Case 8 to Case 11 were compared each other.Pouring of the 200 mL of nutrient salt and leaving were repeated in the intermittent injection of Case 8 and Case 9. In the Case 11, the nutrient salt was injected with fixed discharge.In Case 10, after 200 mL of the nutrient salt was poured and the culture solution was replaced, the 600 mL of the

nutrient was injected with fixed discharge. These 4 cases were carried out at the same time, using the culture solution prepared in the same container.

Fig.9 shows the relationship between the ratio of $CaCO_3$ precipitation and q_u about these 4 cases. In Case11 which the nutrient salt was injected continuously, the ratio of $CaCO_3$ precipitation and q_u was small than in the other 3 cases.

Fig.10 shows the states of each specimen after the unconfined compression tests. In Case 8 and Case 9 which the nutrient salt was injected intermittently, each clods were small, but many clods were found comparatively. It seems that the extent solidified was large. In Case 10 or Case11 which the part or all of the nutrient salt was injected continuously, a few large clods were found comparatively. It seems that the extent solidified was small than Case 8 or Case 9.

From these results, it seems that the uniformity of solidification depended on the method of the injection. So that the ratio of $CaCO_3$ precipitation of the upper and lower parts cut off at the shaping were examined.

Fig.11 shows the comparison about the ratio of $CaCO_3$ precipitation between the upper or the lower part and specimens. The ratio of the lower part to the specimen and the ratio of the upper part to the specimen about the ratio of $CaCO_3$ precipitation were expressed on the horizontal axis and vertical axis, each other. If the ratio of $CaCO_3$ precipitation is equal on the upper part and the lower part, the point is recorded on the dotted line of Fig.11.

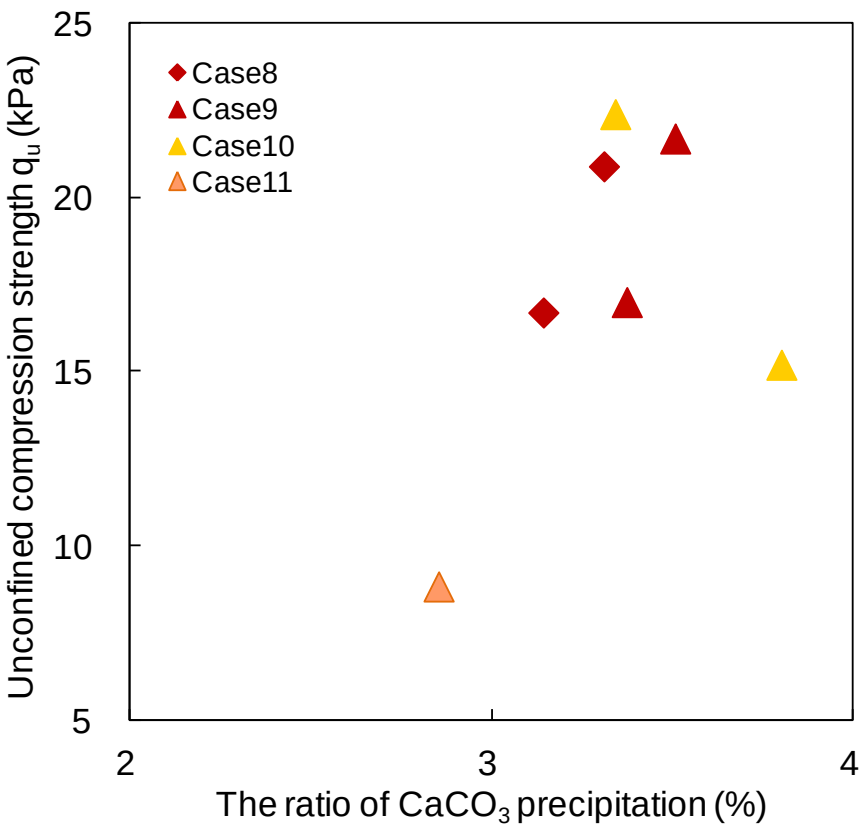


Fig.9 The influence of the method of the injection on the ratio $CaCO_3$ precipitation and q_u

Case 8	Case 9	Case 10	Case 11
Intermittent (Density0.25)	Intermittent (Density0.5)	Continuous after replacement (Density 0.5)	Continuous (Density 0.5)
q_u=16.7kPa	q_u=21.7kPa	q_u=15.2kPa	q_u=8.83kPa
			

Fig.10 The appearance of the specimen after the unconfined compression test
(By the method of the injection)

In Case 9, the ratio of $CaCO_3$ precipitation was nearly equal on the specimen, the upper part and the lower part. But in the other 3 cases, the ratio of $CaCO_3$ precipitation was large in the order of the upper part, the specimen and the lower part. This tendency was remarkable in Case 10 and Case11 which included the continuous injection. It seems that solidification of sand began in the upper part at first and the remaining components were used to precipitate $CaCO_3$ in the specimen and the lower part when the nutrient salt were injected continuously. In Case8 and Case9, it seems that the pore water was replaced by the nutrient salt poured into at first and the precipitation of $CaCO_3$ progressed in the all parts when the sand was leaved. Trough the density of the nutrient salt were different, the nearly equal effect on solidification were got in Case 8 and Case9 (density 0.25 and density 0.5).

It is considered that the continuous injection is adapted to the concentrated solidification to limited extent and that the intermittent injection is adapted to the homogeneous solidification to large extent comparatively.

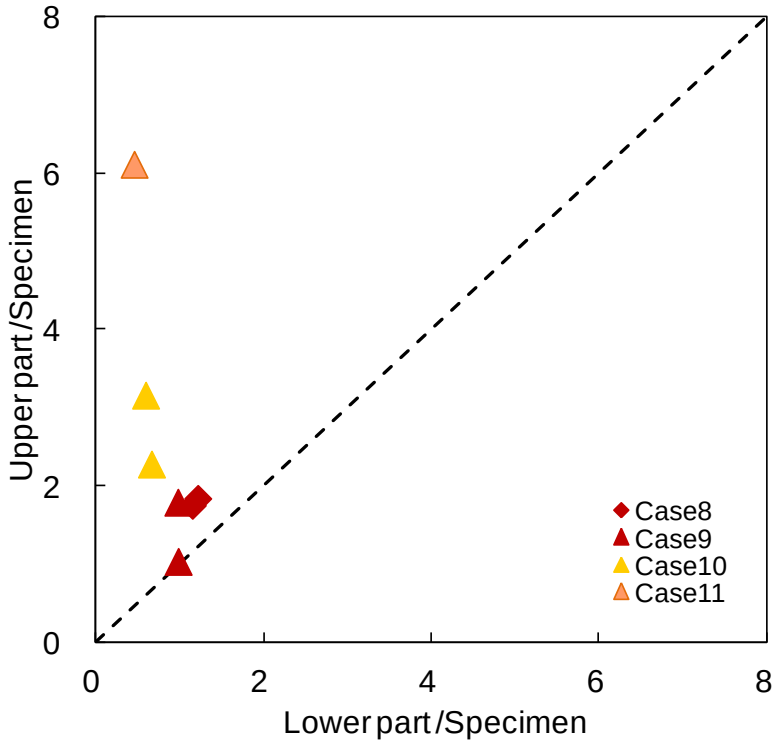


Fig.11 The comparison about the ratio of $CaCO_3$ precipitation between the parts

3.6 The Influence of Density of the Microbial Individual

The examinations for from Case 8 to Case 11 were conducted using the same culture solution, at the same time. The density of microbial individual in this culture solution was 3.8×10^7/mL. Fig.12 shows the relationship between the density of the microbial individual in the pore water at the finish of the period of the injection and q_u.

In all Cases from Case 8 to Case11, the density of microbial individual decreased to from 1-10^4th to1-tength compared with it of the culture solution. Comparatively q_u increased though the density of microbial individual stayed on was small in Case 8 and Case 9 which the nutrient salt was injected intermittently.

This results shows that the rest of many microbial individual is not necessarily advantageous of the solidification. In the case of the intermittent injection, it seems that microbial individual decreases in void because the microbial individual is pushed out of the pore with pore water when the pore water replaces by new nutrient salt poured

It is considered that the intermittent injection can be advantageous of the control of the microbial individual remaining in the pore and the mitigation of environmental influences if the drained pore water is collected and treated appropriately.

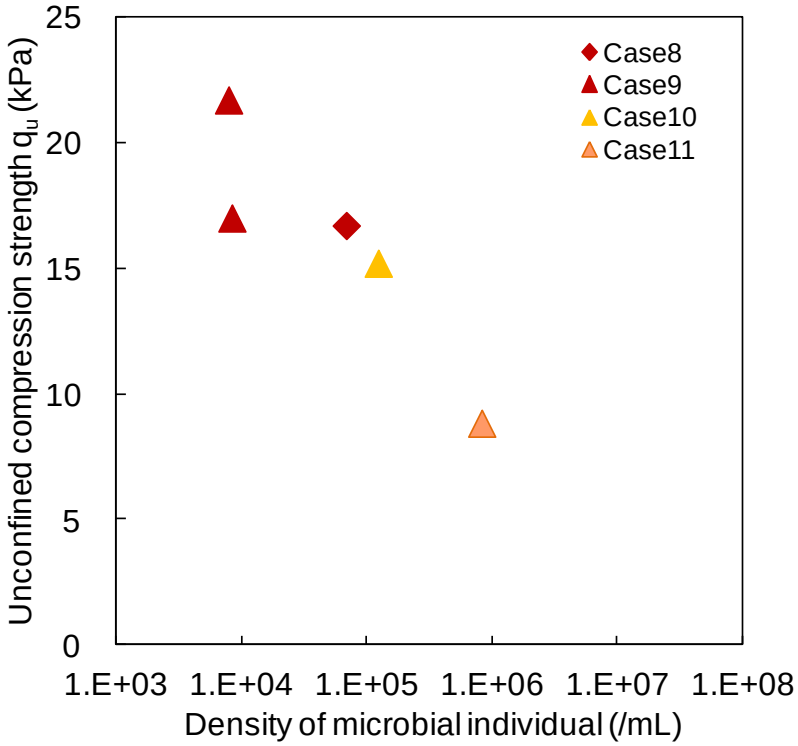


Fig.12 The relationship between the density of the microbial individual in the pore water and q_u.

4. CONCRUTIONS

Studies were conducted about the influences on the solidification of sand by Microbial Carbonate Precipitation of various factors like the density of the nutrient salt, the methods of injection, the discharge in the continuous injection, and so on. The unconfined compression tests were carried out to specimens prepared by the injection of the culture and the nutrient salt.

Followings were investigated under the condition the whole amount of the components was same on the all test cases

・There were the linear relationship between the $CaCO_3$ precipitation ratio and q_u. With the $CaCO_3$ more, q_u tended to increase.

・It is considered that the appropriate condition of discharge to not stop and remain in the void for enough time to precipitate $CaCO_3$ is necessary.

・The period of the injection is not necessarily advantageous of solidification. It is desirable that the period is instituted within the period in the microbial functions is active.

・It is considered that the injection of the thin nutrient salt with much volume can be effective not to decrease the density of the ions from the nutrient salt but also solidification also the mitigation of the environmental influence.

・It is considered that the continuous injection is adapted to the concentrated

solidification to limited extent and that the intermittent injection is adapted to the homogeneous solidification to large extent comparatively.

・The density of microbial individual stays in the pore after the finish of the period of the injection depends on the methods of injection. In the case the nutrient salt supplied by the intermittent injection the density of microbial individual was lower than by the continuous injection. In the case of the intermittent injection, it seems that the microbial individual is pushed out of the pore with pore water when the pore water replaces by new nutrient salt poured.

・The rest of many microbial individual is not necessarily advantageous of the solidification.

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