Quantitative analysis of the crystalline bond lattice of bio-improved soils using micro-computed tomography

Analyse quantitative du réseau de liaisons cristallines de sols bio-améliorés au travers de la micro-tomographie aux rayons X

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ABSTRACT: The study presents a comprehensive characterization of the fabric of bio-improved soils by introducing an image-based quantitative analysis of the crystalline bond lattice, as a means of better understanding and interpreting the observed mechanical response. Calcium carbonate ($CaCO_3$) bonds, produced under the presence and metabolism of the soil bacterium *Sporosarcina Pasteurii*, are analyzed through micro-Computed Tomography (μ-CT). Using a digital-based image processing methodology, and subsequent three-dimensional reconstruction of the solid matrix, we determine the bond particle orientation, sizes and volumetric fraction. Such parameters referring to the morphology and spatial distribution of crystals have been treated so-far in the literature, mainly, in qualitative methods. The study presented herein provides with new insight into the irregular morphology of the crystalline bond lattice and the critical role of the contact area between soil particles and calcite nuclei. Results from fine- and medium-grained bio-improved sands are validated against experimentally measured parameters. $CaCO_3$ bonds are found to exhibit distinctive morphological trends which affect the contact area and ultimately, the mechanical response of the bio-cemented geo-material.

RÉSUMÉ : La présente étude a pour objectif de caractériser la structure des sols bio-améliorés en implémentant une analyse quantitative du treillis de liaisons cristallines, afin de mieux comprendre et interpréter la réponse mécanique obtenue. Des liaisons de la calcite ($CaCO_3$) produites sous la présence et activité métabolique d'une bactérie présente dans le sol *Sporosarcina Pasteurii*, sont soumises à la micro-tomographie aux rayons X (μ-CT). En utilisant une méthode de traitement d'images, suivie par la reconstruction tridimensionnelle de la matrice solide, nous déterminons l'orientation des particules de la calcite, ainsi que leur taille et fraction volumique. Ces paramètres, liés à la morphologie et distribution spatiale des cristaux, sont traités dans la littérature de manière plutôt qualitative. La présente étude fournit un nouvel aperçu de la morphologie particulière du réseau de liaisons cristallines et le rôle-clé de surface de contact entre les grains du sol et les noyaux de calcite. Les résultats obtenus, concernant deux types de sable bio-améliorés, fin et moyen, ont validé les paramètres déterminés grâce à des essais de laboratoire.

KEYWORDS: image processing, soil fabric, shear strength, microstructure

1 INTRODUCTION.

Soil bio-improvement is emerging as a potential solution to geo-engineering problems related to weak and unstable soils. Engineers have been seeking answers in the natural process of biologically driven calcite mineralization for improving the bearing capacity of foundation substrates, for stabilizing slopes, restoring weak foundations, mitigating liquefaction risk in seismic zones, or even using earth as a building material (**Dejong et al.**). Indeed, the formation and growth of calcite mineral crystals inside the soil matrix imparts granular soils with real cohesion, enhanced rigidity and strength. Though, the main challenge related to soil bio-improvement refers to the ability of controlling and adapting the treatment technique in such a way so as to provide solutions that target the specific needs of various geo-engineering problems for various soil types found in-situ (**Terzis & Laloui**).

The paper puts the focus on the characterization of the crystalline structure of two different bio-cemented geo-materials, in order to provide an improved understanding of their fabric and interpret the obtained mechanical response. The critical role of soil fabric, as well as that of fabric anisotropy in granular materials, has led to the development of constitutive models with fabric-dependent parameters (**Li & Dafalias**), as well as to the incorporation of micro-scale inspired variables in modeling formulations to account for evolving soil fabric (**Gajo et al.**). Moreover, available experimental evidence which provide insight into the fabric of granular materials, allows better designing and addressing numerical problems using DEM codes (**Fonseca et al.**, **Nguyen et al.**).

The role of particle size, shape and orientation (**Altuhafi et al.**) on the behavioral characteristics of natural sands has been explored in the literature. However, to the authors' best knowledge, such a quantitative approach to the fabric of soils improved through Microbially Induced Calcite Precipitation (MICP) remains to be established. Part of the reason why such an approach seems a rather complicated and challenging task is that, contrary to pure sands where fabric anisotropy is associated with the deposition of particles and their shape (**Li & Zeng**), an additional source of anisotropy enters the problem for MICP-treated soils. That is the anisotropy of the fabric, obtained post-treatment, induced by the bio-chemical conditions provided to the system during application of MICP. With the postulate that treatment takes place via infiltration, the initial particle arrangement remains intact during circulation of reactants and, thus, it is the treatment conditions that govern the morphology of the calcite lattice. Once the new, enhanced fabric is formed, its locked nature is considered to govern stress transmission and the overall mechanical behavior.

The peculiar structure of bio-improved soils, and mainly the presence of distinctive, individual particles of calcite acting as "bridges" (**Venuleo et al.**), differentiates them from other, chemically treated soils, and sets new challenges in the study of the mechanical behavior of bonded, granular materials. A dependency of the morphology of the $CaCO_3$ particles on the chemical concentration of reactant elements (i.e. urea and calcium) has been found in the works by **Al Qabany & Soga** and **Terzis et al.** In addition, **Cheng et al.** showed that the degree of saturation during treatment yields the nucleation of menisci-like crystalline bonds, which is found to affect significantly the engineering properties of the material. The role of biological factors in the treatment process has been addressed in the work by Harkes et al., as well as, more recently by the authors (Terzis & Laloui). In this latter work, the presence of calcified biofilms is captured as a distinct precipitate behaviour, yielding bonds of larger diameters.

Calcite bonds can, thus, vary significantly in size and shape, depending on the treatment process, with complex physical underlying mechanisms governing their formation and ultimately, their growth. With the present study we aim at providing new, preliminary, insight into the crystalline bond lattice by means of μ-CT. We postulate that since the formation of the calcite lattice is a result of reactive bio-chemical phenomena, the yielded structure is affected by the material's physical properties. To this purpose we choose to apply the same treatment conditions to materials of different initial grain sizes.

2 MATERIALS AND METHODS

We apply MICP to fine-grained (Itterbeck sand, SMALS, Netherlands) and medium-grained sands (Bernasconi, Switzerland), following the same treatment conditions. Their grain size distribution (GSD) is shown in figure 1. Both types of sand yielded the same behaviour in the untreated state under triaxial shear, as discussed in Terzis & Laloui.

For the shake of brevity in this paper, we refer to Terzis et al. and Venuleo et al. for details regarding sample preparation using MICP treatment. Bio-cemented samples are subjected post-treatment to micro-Computed Tomography (μ-CT). μ-CT analyses are carried out using the SkyScan 1173 high energy micro-CT (Bruker) for bio-cemented samples of the two base materials.

A series of parameters are determined quantitatively including: (i) particle size distribution, (ii) particle orientation and (iii) volumetric fraction. This quantitative approach is implemented via an image processing technique, followed by 3D reconstruction of the soil fabric, using the commercially available software AVIZO (FEI, Thermo Fisher Scientific). Except for the MICP-treated samples, untreated samples of pure silica sand are subjected to μ-CT, as a means of validating the crucial, user-defined parameters regarding image segmentation and 3D reconstruction of the soil fabric. Results are validated against the experimentally determined grain size distribution (executed according to Swiss Norm SN670'810) and the measured $CaCO_3$ content, determined using acid digestion (details are available in Terzis et al.).

Due to the rather asymmetrical shape of calcite bonds, the $CaCO_3$ particle size distribution, calculated through image processing, refers to the diameters of spheres of an equivalent volume to that of individual crystal bonds. Moreover, the particle orientation θ is determined for $CaCO_3$ bond particles. It corresponds to the particle's major inertia axis in the X-Y plane (perpendicular to the Z-axis of the sample, as described in Avizo) and exhibits values between 0-360°. Samples subjected to μ-CT have a diameter equal to 5 mm and a height equal to 15 mm ("microcolumns"). The voxel size of the obtained images is respectively 6.41 μm and 7.12 μm for the samples of fine- and medium- sand. The obtained images via μ-CT are thresholded using the watershed module, available in Avizo (Avizo). Particles are smoothened prior to 3D surface reconstruction. This latter is achieved through generating a grid of triangular elements, by setting the maximum edge length equal to 0.5 % of the minimum dimension of the analyzed volume (Avizo). The total contact area is determined by accounting for the surfaces of triangles of calcite and sand in contact. Due to increased computational cost, samples are analyzed in subvolumes for quantifying the volumetric fraction, calcite particle size and particle orientation. Subvolumes have dimensions equal to 10 times the mean particles size of each type of sand, which allows considering that results are obtained in the representative elementary volume (REV) scale. Figure 1 illustrates an example of a fraction of the analyzed subvolume and the 3D reconstructed surfaces of $CaCO_3$ particles.

Bio-cemented samples of fine and medium sand, previously subjected to MICP, following the treatment pattern TP4 described in Terzis et al., are tested mechanically under unconfined compressive conditions, according to ASTM D2166/D2166M-13. Samples are cemented at different target calcite contents in order to investigate the evolution of the Unconfined Compressive Strength (UCS). In Terzis & Laloui we report triaxial compression tests for the same type of sands, with results showing a more significant improvement in terms of strength for the medium-grained sand, despite the same calcite content, yielded after the application of MICP.

3 RESULTS

Figure 2 shows the particle size distributions derived from image processing. Results referring to sand particles ($fine_{3D}^{sand}$ and $medium_{3D}^{sand}$) show good agreement with the experimentally measured curves. The volumetric fractions of the calcite phase are found equal to 9.1% and 7.5%, respectively, for the fine- and medium-grained bio-improved samples. For comparison, acid digestion of the same samples in the laboratory yielded calcite contents equal to 8.9% and 7.8%. The observed discrepancy is attributed to the tomography resolution and subsequent image segmentation, as described in section 1. Scanning Electron Microscopy (SEM) surface analyses are carried out in order to investigate smaller particles that are not detected through μ-CT. Figure 3 shows a sand grain covered by single $CaCO_3$ particles of diameters in the range of 5 μm. Particle sizes in that range cannot be captured through μ-CT, where the maximum resolution allows for the detection of particles greater than 6.41 and 7.12 μm, respectively, for fine and medium sand. However, individual particles of this size range are found mainly to cover the sand grain surfaces (figure 3), rather than bridging neighboring particles. Thus, the effect of the presence of smaller particles on the mechanical behavior of the material remains out of the scope of the present study.

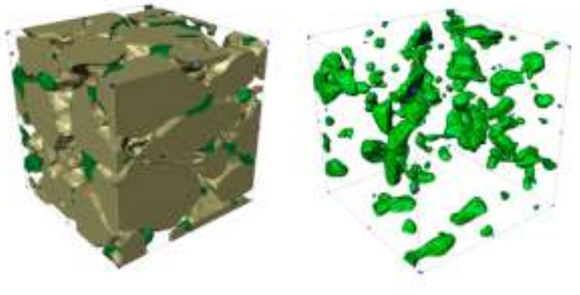

Figure 1. (left) Sand grains (brown) and $CaCO_3$ particles (green) are seen in this example of a fraction of the analyzed subvolume of medium, bio-improved sand; (right) Surfaces of calcite particles of the same fraction of the subvolume

We postulate that since the same conditions are applied during treatment (i.e. initial concentration of biomass, flow rate, chemical concentration of reactants and reaction period allowed) the reaction process adapts differently to the available pore space, yielding crystalline bond structures with distinct properties. Except for the bigger particle sizes yielded for the bio-cemented medium-grained sand, as seen in figure 2, crystals exhibit different trends in their orientations and yield, additionally, different ratio of the number of total bonds over the total number of sand particles. Particle orientation is presented in figure 4, with results referring to the orientation of the major axis . Results show a more homogenous distribution of $CaCO_3$ particle orientations in medium bio-improved sand, compared to fine sand.

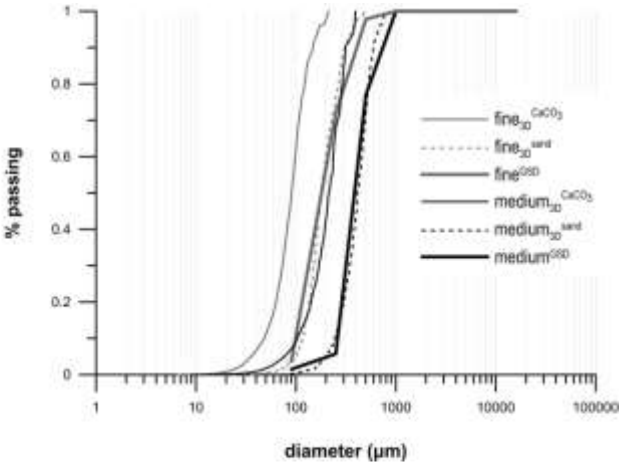

Figure 2. Experimentally measured grain size distribution of fine and medium-grained sand (solid lines); $CaCO_3$ particle size distribution and sand particle size distribution (dashed lines) obtained via 3D image processing.

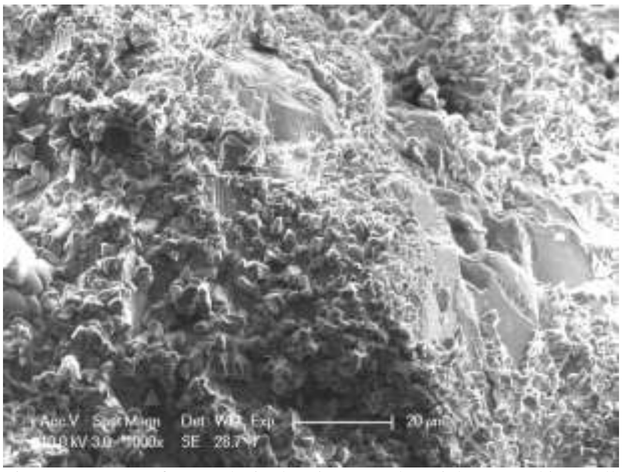

Figure 3. SEM surface analysis showing a sand particle covered in small, single $CaCO_3$ crystals.

Table 1. Material Parameters

	Fine Sand	*Medium Sand*
Mean sand particle size (μm)	190	390
$CaCO_3$ content (%)	9.1	7.5
Subvolume (pixels)	300 x 300 x 300	550 x 550 x 550
Active bonds (% number of total $CaCO_3$)	81	77
N_{CaCO3}/N_{sand} (-) particles ratio	4.3	3.5

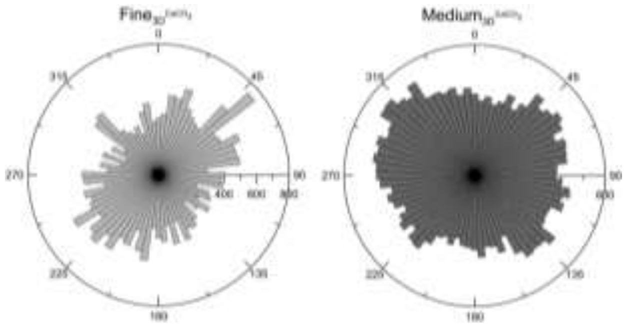

Figure 4. Particle theta-orientation (degrees) of the major inertia axis of individual $CaCO_3$ bonds which precipitate in fine (left) and medium (right) bio-cemented sand.

Table 1 summarizes key parameters related to the materials subjected to MICP. The higher ratio of number of calcite particles over the number of sand particles obtained for fine sand, implies that the total calcite mass is fragmented in a larger number of smaller particles, compared to medium sand, where similar mass of calcite is found to yield a smaller absolute number of particles. Thus, increased contact area does not necessarily imply a larger number of particles, but rather particles which grow in size.

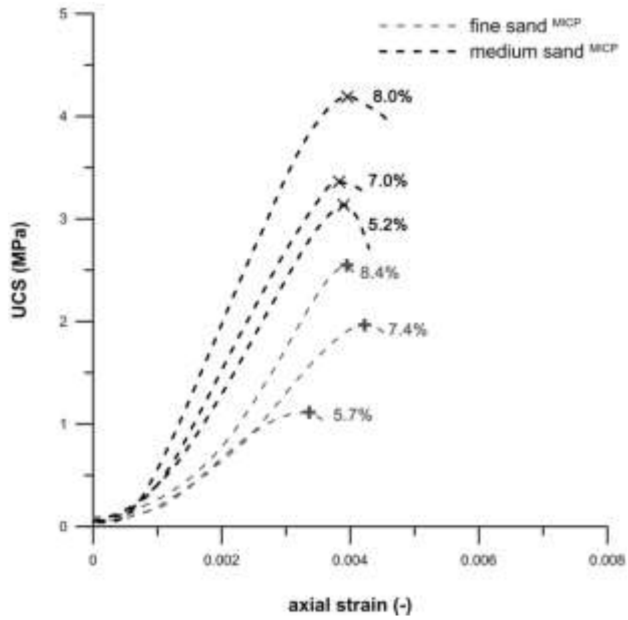

Figure 5. Unconfined Compressive Strength for samples of fine and medium MICP-treated sand for various degrees of calcite content.

Figure 5 presents the evolution of the Unconfined Compressive Strength for both materials, to various calcite gravimetric contents. Results reveal that MICP-treated samples of medium sand yield higher values of strength compared to fine sand. Brittle failure is observed in all tested samples occuring in the small-strain region, at around 0.4% of axial strain. This provides a further validation of the hypothesis adopted previously in Terzis and Laloui (2017), based on triaxial shear tests. In this latter work, the same type of medium sand is found to yield higher values of peak deviatoric strength, compared to fine sand, for similar content in $CaCO_3$. It should be noted that both base materials reach the same peak strength in the untreated state and yield similar dilatancy rates.

4 CONCLUSIONS

The peculiar structure of bio-improved geo-materials, which differentiates them from other artificially cemented soils, implies that its formation needs to be studied in relation with its deformation. Such an approach is adopted in this paper where we seek answers in the material's microstructure in order to interpret the obtained mechanical response. An experimental approach is presented that allows quantifying key characteristics, previously treated in qualitative methods, related to the geometry and spatial distribution of bonding particles.

The study provides quantitative insight into the crystalline bond lattice of bio-improved soils, formed under Microbially Induced Calcite Precipitation. We determine key parameters related to the fabric of bio-cemented sands, such as the size of calcite bonds, their relative number with respect to the number of sand grains, the contact area and particle orientation. MICP is found to respond differently when applied to materials of different initial grain sizes, after following the same treatment conditions. This offers preliminary evidence towards interpreting the mechanical response of materials which, despite the similar calcite content they exhibit, yield different trends in their mechanical response. This finding can contribute towards a more efficient design of the treatment process, by accounting for the material properties of in-situ soils. Finally, the evolution of the material's strength with respect to varying calcite content is captured through UCS tests.

The evidence condensed in the present study can provide with a new understanding around the material's engineering properties, their evolution with respect to calcite content and fabric, as well as their dependency on the materials' initial properties. Such new information can lead to the conception of microscale-inspired parameters in constitutive modeling or to statistically reproducing similar trends of the solid skeleton in numerical simulations using DEM analyses.

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6 REFERENCES

Al Qabany, A., and Soga, K. 2013. Effect of chemical treatment used in MICP on engineering properties of cemented soils. *Géotechnique*, 63(4), 331.

Altuhafi, F. N., Coop, M. R., and Georgiannou, V. N. 2016. Effect of Particle Shape on the Mechanical Behavior of Natural Sands. *Journal of Geotechnical and Geoenvironmental Engineering*, 142(12), 04016071.

ASTM Designation: D2166/D2166M – 13, Standard Test Method for Unconfined Compressive Strength of Cohesive Soil

Avizo. 2009. Avizo User's Guide. Bordeaux, France:Visualization Sciences Group.

Li, B., and Zeng, X. 2014. Effects of fabric anisotropy on elastic shear modulus of granular soils. *Earthquake Engineering and Engineering Vibration*, 13(2), 269-278.

Li, X. and Dafalias, Y. 2012. Anisotropic critical state theory: role of fabric. *J. Engng Mech.* 138, No. 3, 263–275.

Cheng, L., Cord-Ruwisch, R., and Shahin, M. A. 2013. Cementation of sand soil by microbially induced calcite precipitation at various degrees of saturation. *Canadian Geotechnical Journal*, 50(1), 81-90.

DeJong, J.T., Soga, K.S., Kavazanjian, E., Burns, S., van Paassen, L., Al Qabany, A., Aydilek, A., Bang, S.S., Burbank, M., Caslake, L., Chen, C.Y., Cheng, X., Chu, J., Ciurli, S., Fauriel, S., Filet, A.E., Hamdan, N., Hata, T., Inagaki, Y., Jefferis, S., Kuo, M., Laloui, L., Larrahondo, J., Manning, D.A.C., Martinez, B., Montoya, B.M., Nelson, D.C., Palomino, A., Renforth, P., Santamarina, J.C., Seagren, E.A., Tanyu, B., Tsesarsky, M., Weaver, T. "Biogeochemical Processes and Geotechnical Applications: Progress, Opportunities, and Challenges" *Geotechnique*, 2013, Vol. 63, No. 4, pp. 287-301.

Fonseca, J., Nadimi, S., Reyes-Aldasoro, C. C., & Coop, M. R. 2016. Image-based investigation into the primary fabric of stress-transmitting particles in sand. *Soils and Foundations*, 56(5), 818-834.

Gajo, A., Cecinato, F., & Hueckel, T. 2015. A micro-scale inspired chemo-mechanical model of bonded geomaterials. *International Journal of Rock Mechanics and Mining Sciences*, 80, 425-438.

Harkes, M. P., Van Paassen, L. A., Booster, J. L., Whiffin, V. S., & van Loosdrecht, M. C. 2010. Fixation and distribution of bacterial activity in sand to induce carbonate precipitation for ground reinforcement. *Ecological Engineering*, 36(2), 112-117.

Nguyen, H. B. K., Rahman, M. M., & Fourie, A. B. 2016. Undrained behaviour of granular material and the role of fabric in isotropic and K 0 consolidations: DEM approach. *Géotechnique*, 67(2), 153-167.

Terzis, D., Bernier-Latmani, R., Laloui, L. 2016. Fabric characteristics and mechanical response to bio- improved sand to various treatment conditions. *Géotechnique Letters* 6(1):50-57

Terzis, D. and Laloui, L. 2017. On the Application of Microbially Induced Calcite Precipitation for Soils: A Multiscale Study in Advances in Laboratory Testing and Modelling of Soils and Shales (ATMSS) (Lyesse, L. and Ferrari, A., eds.): 388-394

Venuleo, S., Laloui, L., Terzis, D., Hueckel, T., Hassan, M. 2016. Microbially induced calcite precipitation effect on soil thermal conductivity. *Géotechnique Letters* 6(1): 39-44