




Abstract

# Use of Bacterial Carbonatogenesis for Construction Materials †

Iuliana Raut<sup>1</sup>, Mariana Constantin<sup>1</sup>, Elvira Alexandrescu<sup>1</sup> , Claudia Ninciuleanu<sup>1</sup>, Monica Raduly<sup>1</sup> , Ana-Maria Gurban<sup>1</sup> , Mihaela Doni<sup>1</sup>, Ionela Petre<sup>2</sup>, Cristian Andi Nicolae<sup>1</sup>, Nicoleta Radu<sup>1,3</sup>, Gelu Vasilescu<sup>1</sup> and Luiza Jecu<sup>1,\*</sup>

<sup>1</sup> Biotechnology Department, National Institute for Research & Development in Chemistry and Petrochemistry-ICECHIM, 202 Independentei Spl., 060021 Bucharest, Romania; iulia\_rt@yahoo.com (I.R.); marriconstantin@yahoo.com (M.C.); elviraalexandrescu@yahoo.com (E.A.); claudia.ninciuleanu@yahoo.com (C.N.); monica.raduly@icechim.ro (M.R.); ana-maria.gurban@icechim.ro (A.-M.G.); mihaela.doni@icechim.ro (M.D.); ca\_nicolae@yahoo.com (C.A.N.); nicolbiotec@yahoo.com (N.R.); gelu\_gvp@yahoo.com (G.V.)

<sup>2</sup> CEPROCIM SA, 6 Preciziei Street, 062203 Bucharest, Romania; ionela.petre@ceprocim.ro

<sup>3</sup> Faculty of Biotechnology, University of Agronomic Sciences and Veterinary Medicine of Bucharest, 59 Mărăști Boulevard, 011464 Bucharest, Romania

\* Correspondence: jecu.luiza@icechim.ro

† Presented at the 17th International Symposium “Priorities of Chemistry for a Sustainable Development” PRIOCHEM, Bucharest, Romania, 27–29 October 2021.

**Keywords:** *Bacillus*; carbonatogenesis; microbially induced carbonate precipitation (MICP)



**Citation:** Raut, I.; Constantin, M.; Alexandrescu, E.; Ninciuleanu, C.; Raduly, M.; Gurban, A.-M.; Doni, M.; Petre, I.; Nicolae, C.A.; Radu, N.; et al. Use of Bacterial Carbonatogenesis for Construction Materials. *Chem. Proc.* **2022**, *7*, 42. <https://doi.org/10.3390/chemproc2022007042>

Academic Editors: Florin Oancea, Zina Vuluga and Radu Claudiu Fierăscu

Published: 16 March 2022

**Publisher's Note:** MDPI stays neutral with regard to jurisdictional claims in published maps and institutional affiliations.



**Copyright:** © 2022 by the authors. Licensee MDPI, Basel, Switzerland. This article is an open access article distributed under the terms and conditions of the Creative Commons Attribution (CC BY) license (<https://creativecommons.org/licenses/by/4.0/>).

Concrete is the most used construction material, but its industrial production from lime consumes 2–3% of the global energy demand, generating 0.73–0.99 t CO<sub>2</sub>/t of cement produced [1]. At the same time, concrete structures are susceptible to physical, chemical and biological factors, which affect their mechanical and durability properties. A viable alternative to reduce cost and environmental impact is considered as the incorporation in the cement matrix of bacteria capable to precipitate calcium carbonate [2,3]. Microbially induced calcium carbonate precipitation (MICP) is possible through the following metabolic pathways: urea hydrolysis, ammonification of amino acids, denitrification, sulfate reduction and photosynthesis. Among them, urea hydrolytic metabolism is the most studied [4]. The aim of this study was to evaluate the capability of several *Bacillus* strains to precipitate calcium carbonate. Each bacterial strain was cultivated on a urea-CaCl<sub>2</sub> medium of different concentrations and CaCO<sub>3</sub> precipitation was evaluated.

The experimental study was carried out with bacterial strains from the Microbial Collection of ICECHIM, as follows: *Bacillus amyloliquefaciens*, *B. licheniformis* and *B. subtilis*. The bacteria were cultured on media with different compositions: (i) tryptic soy broth (TSB); (ii) nutrient broth (NB), with different concentrations of urea and Ca<sup>+2</sup>. The cultures were incubated and then centrifuged, with the obtained pellets being analyzed with different techniques (FTIR-ATR, SEM and TGA).

SEM investigations presented several morphological aspects of the pellets. The bacterial pellets from cultures were characterized by TGA analysis. The results indicated that the pellets from the media with calcium and urea featured two main weight loss steps, 17–24% (138–145 °C) and 19–20% (343–350 °C), respectively. The residue at 700 °C corresponded to a significant weight loss of 45–50%. The behavior of the control samples from the medium, without urea and calcium, was quite different: loss of 65–68% (248–281 °C) and 27–31% residue. This aspect indicated that on the medium with urea and Ca<sup>+2</sup> ions, bacterial strains were able to precipitate calcium carbonate. FTIR spectra of pellets presented the following distinguishable regions: 3000–2800 cm<sup>-1</sup> for cell membrane fatty acids and carbohydrates; 1700–1500 cm<sup>-1</sup> for amide I; band around 1401–1416 cm<sup>-1</sup> assigned to CO<sub>2</sub>; 1200–900 cm<sup>-1</sup> polysaccharides or carbohydrates of microbial cell walls.

The corroboration of the present results with those obtained previously, regarding the secretion of urease [5], indicated *Bacillus subtilis* to be a microorganism with great potential for application in cementitious materials. Further investigation regarding the immersion of bacterial cells in cement will be carried out.

**Author Contributions:** L.J., I.R. and M.D. conceived and designed the experiments; M.C., I.R., A.-M.G., I.P., E.A., C.N., G.V., M.R. and C.A.N. performed the experiments; L.J., M.D., N.R. and A.-M.G. analyzed the data; L.J., I.R., M.C. and M.D. wrote the paper; L.J., A.-M.G. and N.R. reviewed and edited. All authors have read and agreed to the published version of the manuscript.

**Funding:** This work was supported by a grant of the Ministry of Research, Innovation and Digitization, CNCS/CCCDI-UEFISCDI, project number 392/2020, within PNCDI III" and project POC 2016 SECVENT, P\_40\_352, MySMIS: 105684.

**Acknowledgments:** The authors thank to Ministry of Research, Innovation and Digitization of Romania through Program 1-Development of the national research and development system, Subprogram 1.2-Institutional performance-Projects to finance excellence in RDI, Contract no. 15PFE/2021.

**Conflicts of Interest:** The authors declare no conflict of interest.

## References

1. Castro-Alonso, M.J.; Montañez-Hernandez, L.E.; Sanchez-Muñoz, M.A.; Macias Franco, M.R.; Narayanasamy, R.; Balagurusamy, N. Microbially Induced Calcium Carbonate Precipitation (MICP) and Its Potential in Bioconcrete: Microbiological and Molecular Concepts. *Front. Mater.* **2019**, *6*, 126. [[CrossRef](#)]
2. Joshi, S.; Goyal, S.; Mukherjee, A.; Reddy, M.S. Microbial healing of cracks in concrete: A review. *J. Ind. Microbiol. Biotechnol.* **2017**, *44*, 1511–1525. [[CrossRef](#)] [[PubMed](#)]
3. Lee, Y.S.; Park, W. Current challenges and future directions for bacterial self-healing concrete. *Appl. Microbiol. Biotechnol.* **2018**, *102*, 3059–3070. [[CrossRef](#)] [[PubMed](#)]
4. Krajewska, B. Urease-aided calcium carbonate mineralization for engineering applications: A review. *J. Adv. Res.* **2018**, *13*, 59–67. [[CrossRef](#)] [[PubMed](#)]
5. Răut, I.; Călin (Constantin), M.; Gurban, A.M.; Doni, M.; Vasilescu, G.; Alexandrescu, E.; Radu, N.; Jecu, L. Screening of bacterial strains active in microbial induced carbonate precipitation through ureolytic pathway. In Proceedings of the International Conference of the University of Agronomic Science and Veterinary Medicine of Bucharest "Agriculture for Life, Life for Agriculture", Bucharest, Romania, 3–5 June 2021.