Life Cycle Assessment of Biocement: An Emerging Sustainable Solution?

Hannah Porter 1, Abhijit Mukherjee 1, Rabin Tuladhar 2 and Navdeep Kaur Dhami 1,*

1 Biologically Activated Materials Laboratory, School of Civil and Mechanical Engineering, Curtin University, Bentley, WA 6102, Australia; Hannah.porter@postgrad.curtin.edu.au (H.P.); abhijit.mukherjee@curtin.edu.au (A.M.)
2 College of Science and Engineering, James Cook University, Douglas, QLD 4811, Australia; rabin.tuladhar@jcu.edu.au
* Correspondence: navdeep.dhami@curtin.edu.au

Abstract: Microbially Induced Calcium Carbonate Precipitation (MICP) is a natural biocementation that takes place in corals, stromatolites and beach rocks. In recent years, researchers have explored the emulation of this process as a sustainable alternative of engineered cement. Although the natural process is undoubtedly sustainable, its engineered variant deviates substantially from the natural process. In this paper, we investigate the environmental and economic performance of the engineered biocementation process vis-à-vis present manufacturing of calcium carbonate. SimaPro 8.0 software and the Ecoinvent V2.2 database were used for materials inputs and AUSLCI along with Cumulative Energy Demand 2.01 software were used for carbon footprint and eutrophication potential. Our results show that different metabolic pathways of MICP have considerably varying environmental impact. We observe that nature performs MICP sustainably at ambient conditions and geological time scales utilizing naturally occurring sources of carbon and calcium at micromoles concentrations. Due to the mandate on duration of construction projects, highly purified reactants in a high concentration are used in the engineered process. This has a negative environmental impact. We conclude that the sustainability of engineered MICP is directly impacted by the metabolic pathway of bacteria as well as the purity of the input chemicals. A few biotic processes are superior to the present industrial process for manufacturing calcium carbonate if ingredients of laboratory grade purity are replaced by industrial grade products. A bigger dividend can be obtained by introducing industry by-products as nutrients. The results of this study help to direct future research for developing sustainable biocement for the construction industry.

Keywords: biocement; MICP; life cycle analysis; sustainability

1. Introduction

Microbially Induced Calcium Carbonate Precipitation (MICP) is a form of mineralisation that is responsible for major carbonate formations in nature such as corals, stromatolites and beach rocks [1]. Similar to industrial cement, the grains of sand can be bound together through MICP. Thus, MICP is biocementation that occurs at ambient conditions with no additional source of energy and with water as the solvent. The construction industry, on the other hand, is heavily reliant on ordinary Portland cement (OPC) that produces roughly the same amount of greenhouse gases as its own weight [2]. Worldwide, nearly 3.6 billion tonnes of OPC is produced, which accounts for approximately 6% of anthropological greenhouse gases. Researchers are exploring the emulation of the natural cementation process as a means of achieving sustainability in construction. Dejong, et al. [3] conclude that harnessing the biological processes that occur in natural formations is the next transformative practise for geotechnical engineering. Biocementation is envisaged to be sustainable due to several factors such as a low embodied energy, reversibility and recyclability and self-healing [4–7]. However, there has been little attempt
to objectively examine the sustainability of engineered biocementation. Biocementation has been happening in nature for millions of years, and there is no doubt that the process is sustainable. However, it occurs over geological timeframes utilizing naturally available reactants often at micromolar concentrations (Figure 1a). When emulating biocementation for construction applications, it becomes necessary to enrich both the bacteria and the nutrient media to allow the process to fit it into the speed, reliability and performance mandates of the construction industry [8–12] (Figure 1b). Investigators have typically used purified laboratory grade chemicals at a much higher concentration than what occurs in nature [8–15]. The purification process of the chemicals is likely to consume considerable energy. Thus, it is important to evaluate the input media to ascertain the environmental impact of biocementation. Clearly, biocementation can be performed in a number of ways, and a methodology to evaluate the processes to identify the best among several alternatives is essential. Life cycle analysis has emerged as a great tool for evaluating sustainability [16], particularly in the areas of sustainable housing technologies, building assessments [17–20] and commonly used construction materials such as cement [21–23] and plastic wastes [24] aided by the enormous amount of field data over a long time. There are few studies on industrial-scale projects with biocement to conduct a full-scale life cycle analysis, although the importance of such a study cannot be overestimated. An initial embodied energy analysis on biocementation by urea hydrolysis reveals that the manufacturing processes for urea and calcium chloride are the key contributors to embodied energy, while for ordinary Portland cement, the key contributor to energy usage is the burning of limestone during the calcination process [25]. The production of ordinary Portland cement is estimated to consume approximately 6.21 MJ/kg [26] of energy while urea consumes 30.54 MJ/kg [27] and calcium chloride 11.76 MJ/kg [28]. Field scale experiments demonstrate that 0.6 kg urea and 1.1 kg calcium chloride is required to produce 1 kg of calcium carbonate for biocement through the ureolytic pathway [10,29]. Thus, there is an imperative of examining the sustainability of engineered biocementation. Early indications on the likely pathways to sustainable options based on already available extensive laboratory experiments would be valuable to chart the research directions towards sustainable biocement technologies for the field. This paper employs life cycle analysis methodologies on the available data to identify the developments that are likely to have the deepest impact on the sustainability of biocement technology.
1.1. Process of Biocementation

In biocementation, the bacteria nucleate the conversion of a water-soluble source of calcium such as calcium chloride into an insoluble one such as calcium carbonate. The bacterial cells secure themselves in the grooves of the substrate and act as the nucleation site for the growth of the calcium carbonate crystals (Figure 2a). Thus, the crystals grip the grains at several places and coat it and gradually grow into mesocrystals (Figure 2b). When the mesocrystals from different sand grains join together, they become cemented (Figure 2c). Through quantitative scans of the substrate, the extent of cementing of the grains can be established (Figure 2d).
sand grains and forms the nucleation sites for the calcium carbonate crystals. (b) The crystals grow to form mesocrystals. (c) The crystals coat the surface and continue to grow. When the crystals bridge the neighbouring sand grains, cementation is formed. (d) Quantitative EDS scans reveal various stages of biocementation [30].

During MICP, naturally occurring microbes act as a biocatalyst for the precipitation of calcium carbonate [3] (Figure 2). MICP can occur via autotrophic or heterotrophic pathways (Table 1), although presently, MICP via urea hydrolysis is the most investigated route for construction applications due to its simplicity [31]. In the case of heterotrophs, mineralization takes place as a by-product of the metabolic activity of the bacteria. In these cases, heterotrophic bacteria utilize organic compounds, such as urea, for energy and cellular material [31,32]. The metabolic activity of the bacteria causes a rise in the pH of the surrounding pore water, resulting in supersaturated conditions and allowing for the precipitation of carbonates when in the presence of an inorganic calcium source. In the cases of autotrophs, bacteria obtain energy from the sun and reduce the atmospheric carbon (in the form of carbon dioxide) for energy and cellular material [1]. Heterotrophic pathways include urea hydrolysis, denitrification, ammonification and methane reduction, while autotrophic pathways include photosynthesis and MICP through carbonic anhydrase-producing bacteria. In both heterotrophic and autotrophic pathways, the rate of MICP is controlled by factors such as the metabolic activity of the bacteria, the availability of calcium, the dissolved inorganic carbon source and the pH of the surrounding environment [3,12,13,33]. Due to differing levels of nutrients and waste products produced during the reaction, different pathways to MICP are likely to have varying impacts on the environment. Their efficacy in terms of sustainability is an important question to answer, particularly when developing MICP as a construction technique for industrial applications.

**Table 1. Microbial pathways to precipitation of calcium carbonate.**

| Metabolic Type   | Pathway               | Bacteria Type                          | Chemical Reactions                                                                 |
|------------------|-----------------------|----------------------------------------|------------------------------------------------------------------------------------|
| Heterotrophic    | Urea hydrolysis [3,10,29,31,34] | Ureolytic bacteria (Bacillus pasteurii) | (a) Urea + H2O → 2NH3 + CO2  
(b) NH3 + H2O → NH4+ + OH−  
(c) OH− + CO2 → HCO3−  
(d) Ca2+ + HCO3− → CaCO3 + H2O |
|                  | Denitrification [8,14] | Denitrifying bacteria (Pseudomonas denitrificans) | (a) NO3− + 1.25 CH2O → 0.5N2 + 1.25CO2 + 0.75H2O + OH−  
(b) Ca2+ + CO32− + 2OH− → CaCO3 + H2O |
| Ammonification [35–37] | Myxobacteria (Myxococcus xanthus) |                        | (a) Amino acid + O2 → NH3 + CO2 + H2O  
(b) NH3 + H2O → NH4+ + OH−  
(c) OH− + CO2 → HCO3−  
(d) Ca2+ + HCO3− → CaCO3 + H2O |
| Methane Oxidation [38,39] | Methanogens (Methylocystis parvus) |                        | (a) Methane + SO42− → HS− + HCO3− + H2O  
(b) Ca2+ + HCO3− → CaCO3 + H2O  
(c) H+ + HS− → H2S |
| Autotrophic      | Carbonic Anhydrase [40,41] | (Bacillus Megaterium) | Ca2+ + 2HCO3− → CaCO3 + HCO3− + H+ → CaCO3 + CO2 + H2O                                      |
|                  | Photosynthesis [33,42]  | Cyanobacteria (Synechococcus) | CO2 + H2O + 2HCO3 + Ca2+ → CH2O + CaCO3 + O2 + H2O                                                   |

1.2. Cost of Biocementation

The economic cost of biocement has been estimated by a few researchers. De Muznck, et al. [43] conducted an economic assessment of biocement as a surface treatment for building materials. The urea/calcium chloride cementation media was found to be the highest contributor to costs, responsible for approximately 47% of the overall cost of the treatment. The cost of application and added value of the product accounted for 41% of
the overall cost while the bacterial growth media was approximately 12% or the total cost. The overall cost of biocement as a surface treatment was 23–28 EUR/sqm, (AUD 34–42).

One way of controlling the cost and embodied energy of biocement is to use unprocessed natural materials or industry by-products. Our prior studies have demonstrated the potential of corn steep liquor or lactose mother liquor as an alternative, growth medium for the ureolytic bacterial strain *Sporosarcina pasteurii* [44,45]. Research has also demonstrated the potential of using acetic acid and limestone as an alternative calcium source for ureolytic bacteria [46]. However, acetic acid typically has a higher environmental carbon footprint (1.556 kg CO$_2$/kg) and embodied energy (53.35 MJ/kg) when compared to the laboratory grade calcium chloride (0.854 kg CO$_2$/kg and 10.96 MJ/kg), making this option unlikely to have a lower overall environmental footprint than the traditional alternatives [28,47]. Eggshells as an alternative calcium source has also been explored [46,48]. Whilst the study demonstrated that laboratory grade calcium chloride can be replaced with eggshells, the overall sustainability of this method cannot be assessed as the resources required to collect and reuse eggshell waste is unknown.

With this background, we will investigate the environmental and economic performance of engineered biocementation (limestone) via a range of bacterial metabolic processes and compare its performance with the present manufacturing process of calcium carbonate, which has not been done before. We will also analyse the impact of different nutrient sources and industrial grade and lab grade chemicals on the LCA of biocementation for the first time. SimaPro 8.0 software and the Ecoinvent V2.2 database will be used for materials inputs and AUSLCI along with Cumulative Energy Demand 2.01 software will be used for carbon footprint and eutrophication potential.

2. Materials and Methods

From the above discussion, it is clear that a comprehensive assessment of alternative processes is essential to evaluate the sustainability of biocementation and direct future research in the area. However, lack of industrial scale data inhibits a full life cycle analysis. This paper applies the life cycle analysis methodologies on the available experimental results to compare the environmental and economic costs of different routes to biocementation to facilitate an informed future research with a view to sustainability. An assessment has been conducted on calcium carbonate precipitated through different MICP metabolic pathways as compared to an equal mass of manufactured calcium carbonate.

The functional unit (FU) for the assessment was defined as 1 kg of precipitated calcium carbonate. The scope of the assessment for microbially precipitated calcium carbonate included the extraction and processing of raw materials, as well as the environmental impact of any by-products produced during the MICP (Table 1). It may be remembered that the scale of allocation of MICP is relatively small. Therefore, certain factors could not be estimated. Transportation of raw materials to the plant and the energy required to operate the fermenter were excluded from the analysis. The analysis was conducted using SimaPro 8.0 software, which is the most widely used LCA software to analyse and model complex life cycles in the building sector [49,50]. The software provides the user interface, the environmental information database and the options for the impact assessment method for the LCA practitioner [50]. This software comes with numerous advantages including flexibility, ease of use, transparent results and accuracy with large amounts of data. This software includes a large amount of background data and carbon dioxide equivalence factors for greenhouse gas emissions. There are several databases available in SimaPro, but the most widely used one in the construction sector is Ecoinvent V2.2 database [26,50,51], which was utilised in the current study. This database provided material inputs with adaptations to an Australasian context where available. In the study of (Martiney-Rocamora, et al. [52], wherein they compared different LCA databases, Ecoinvent was reported to stand out for its integrity, usability and dedicated resources. The carbon footprint and eutrophication potential were calculated using Australian Life Cycle Inventory Database (AUSLCI) Version 3.0 (http://www.auslci.com.au, accessed on 10 December 2020) containing datasets...
gathered from Australian-specific sources [53]. The embodied energy for each scenario was calculated using the Cumulative Energy Demand 2.01 methodology (Available from http://www.ecoinvent.org accessed on 10 December 2020). This is a high-level assessment for the purpose of comparing major impacts generated by different metabolic pathways, based on laboratory scale data for the purpose of directing future research. A full life cycle analysis, including other impact factors (such as water and land use, mineral and fossil fuel depletion, etc.) is outside the scope of this study and is a subject of future investigation.

The following assumptions have been made in this analysis:

- All the reaction efficiencies are 100% and all of the provided calcium source is converted into calcium carbonate.
- The effect of the metabolic rate of different pathways of MICP has not been considered.
- Waste products generated by the MICP process have been included in the analysis. However, treatment and recycling of these products is not considered.
- Production of laboratory grade calcium carbonate through the carbonation process were based on the cradle-to-gate assessment conducted by Mattila, et al. [54].
- Costs (AUD) for laboratory grade materials were based on published rates from suppliers [55–57], whilst costs for commercial grade chemicals (in bulk) were based on data published by ICIS [58].

Methods, including statements of data availability, assumptions and scope of the assessment are available in the online version of the paper and in Supplementary Material.

3. Results and Discussions

The overall environmental impact of calcium carbonate produced through MICP using laboratory grade chemicals is shown in Figure 3. The environmental impacts of 1 kg of calcium carbonate produced using carbonic anhydrase producing bacteria, methanogens, cyanobacteria, denitrifying bacteria, ureolytic bacteria and myxobacteria are compared to the environmental impact of 1 kg of calcium carbonate produced using the traditional carbonation process.

With the exception of MICP through denitrification, calcium carbonate produced through MICP has a lower carbon footprint than an equal quantity of calcium carbonate produced through the traditional carbonation process. This is due to the fact that MICP occurs under ambient conditions and does not have the energy requirements of the calcination process. Autotrophic pathways to MICP (carbonic anhydrase bacteria and photosynthesis) utilized carbon dioxide from the atmosphere, reducing the overall carbon footprint by 0.44 kg/kg CaCO$_3$. Calcium carbonate produced using the carbonic anhydrase enzyme and had the lowest carbon footprint (0.681 kg CO$_2$/kg CaCO$_3$), 70% lower than the carbon footprint of the traditional carbonation process.

The eutrophication potential for the production of 1 kg of CaCO$_3$ using laboratory grade chemicals is also shown in Figure 3. MICP via ureolytic bacteria has the highest eutrophication potential (0.24 kg SO$_4$/kg CaCO$_3$), followed by myxobacteria (0.065 kg SO$_4$/kg CaCO$_3$) and denitrifying bacteria (0.02 kg SO$_4$/kg CaCO$_3$). All three scenarios have a higher eutrophication potential than the production of 1 kg of CaCO$_3$ through the carbonation process. In the case of urea hydrolysis and denitrification, the eutrophication potential can be attributed to the ammonium gas produced during the reaction. In the case of denitrifying bacteria, the eutrophication potential is largely due to the nitrogen gas produced during MICP. The eutrophication potential of all other MICP pathways is minimal. These results demonstrate that the gaseous by-products of MICP undertaken by the more commonly investigated routes (ureolytic bacteria, denitrification and ammonification) have the potential to cause significant environmental damage. If MICP is to become a truly sustainable construction technology, consideration must be given to treating or reusing the by-products of MICP in a way that does not adversely impact other environmental factors such as carbon footprint, embodied energy, etc. One example of the reuse of ammonia produced by the ureolytic pathway would be as an ammonia fertilizer product for plants [3].
Figure 3. Environmental impact ranking and key material contributors for the production of 1 kg of CaCO$_3$ through different metabolic pathways using laboratory grade inputs.

| Pathway                          | Carbon Footprint (CO$_2$ / FU) | Enutrification Potential (SO$_2$ / FU) | Embodied Energy (MJ) | Cost (AUD / FU) | Overall Ranking |
|----------------------------------|--------------------------------|----------------------------------------|----------------------|-----------------|-----------------|
| (i) Carboxylic anhydrase producing bacteria | 0.671                          | 1.77e-3                                | 12.9                 | 62              | 1               |
| (ii) Carbonate                   | 2.37                           | 7.5e-4                                 | 7.2                  | 61              | 2               |
| (iii) Methanogens                | 1.15                           | 1.79e-3                                | 13.3                 | 71              | 3               |
| (iv) Cyanobacteria               | 1.38                           | 2.97e-3                                | 20.1                 | 99              | 4               |
| (v) Denitrifying bacteria        | 3.91                           | 2.67e-2                                | 13.6                 | 102             | 5               |
| (vi) Ureolytic bacteria          | 2.06                           | 2.4e-1                                 | 28.4                 | 107             | 6               |
| (vii) Myxobacteria               | 2.34                           | 6.5e-2                                 | 23.4                 | 138             | 7               |
MICP undertaken using laboratory grade chemicals has a significantly higher embodied energy than the traditional carbonation process. MICP via ureolytic bacteria had the highest embodied energy (28.4 MJ), whilst MICP using carbonic anhydrase-producing bacteria had the lowest embodied energy (12.9 MJ). The embodied energy requirements for MICP pathways are predominantly due to the laboratory grade calcium chloride, which contributes between 44 and 98% of the total embodied energy.

The unit costs for MICP using laboratory grade chemicals generally exceed that of calcium carbonate produced through the carbonation process. The key contributors to the overall costs are shown in Figure 3. Calcium chloride is a key cost across all MICP routes. Other significant costs include the cost of urea (for urea hydrolysis) and sodium nitrate (for denitrification). The most expensive route was MICP via ammonification or methane oxidation (138 USD/kg CaCO$_3$), while the most economic route was MICP via cyanobacteria (62 USD/kg CaCO$_3$).

The overall environmental impact of calcium carbonate produced through MICP using commercial grade chemicals is shown in Figure 4. The environmental impacts of 1 kg of calcium carbonate produced using carbonic anhydrase producing bacteria, methanogens, cyanobacteria, denitrifying bacteria, ureolytic bacteria and myxobacteria are compared to those of 1 kg of calcium carbonate produced using the traditional carbonation process.

For the laboratory grade scenarios it was assumed that the calcium source (CaCl$_2$) was produced using the traditional Solvay process [28]. However, for the commercial grade scenarios, it was assumed that the calcium source was less pure, and calcium chloride produced through the hypochlorination of allyl chloride [59] was adopted for the analysis. In the case of denitrification, sodium nitrate was also replaced with a nitrate fertilizer [27].

The substitution of a commercial grade calcium chloride dramatically decreased the carbon footprint across all MICP routes. A decrease between 18% and 49.62% was recorded across all MICP pathways. Key contributors to the carbon footprint are also shown for each metabolic pathway. A key component for all processes is the provided calcium chloride. Clearly, future research should investigate alternatives to laboratory grade calcium sources to further enhance the sustainability of carbonates produced through MICP. In the case of MICP via urea hydrolysis, or denitrification, the provided organic carbon source (urea or nitrate) results in a greater contribution to the carbon footprint than the provided calcium chloride. In the case of MICP via cyanobacteria or myxobacteria, the provided bicarbonate or glucose also results in a significant contribution to the overall carbon footprint.

A significant decrease in embodied energy (between 43 and 95%) was achieved in all pathways, with the exception of denitrification. This decrease is largely attributed to the replacement of the laboratory grade calcium chloride with a commercial grade product. MICP using carbonic anhydrase bacteria required the least embodied energy (0.619 MJ), 91% less than calcium carbonate produced through the traditional carbonation process.

While the cost for MICP-produced carbonates is still higher than calcium carbonate produced through the traditional carbonation process, (0.4 USD/kg CaCO$_3$), it may be recalled that the MICP processes do not enjoy the economies of scale that the present industrial process has. However, use of commercial grade inputs does improve the viability of carbonates produced using the MICP process, bringing the costs into the same range as calcium carbonate produced through carbonation.

The metabolic pathways to MICP were ranked according to the overall environmental impact. When using laboratory grade input materials, MICP through carbonic anhydrase producing bacteria, cyanobacteria or methanogens all had a lower overall environmental impact when compared to the production of an equal amount of calcium carbonate using the traditional carbonation process. When commercial grade materials were used for MICP, the overall environmental impact and cost for carbonates produced during the MICP process decreased remarkably. To date, the majority of MICP investigations have utilised laboratory grade products, and as such, MICP may not be techno-economically competitive when compared to other construction materials such as Portland cement. Going forward,
research in the area of MICP must prioritize low-cost commercial alternatives or waste products as the calcium and organic carbon source for the MICP reaction.

Figure 4. Environmental impact ranking and key material contributors for the production of 1 kg of CaCO$_3$ through different metabolic pathways using commercial grade inputs.
In the case of MICP via urea hydrolysis, denitrification or ammonification, the provided organic carbon source (urea, nitrate or glucose) results in an equal contribution to the carbon footprint, embodied energy and cost as the provided calcium source. The gaseous waste produced by these pathways (ammonium or nitrogen) also results in high eutrophication potential, which makes carbonates produced through these pathways less sustainable than the traditional carbonation method. For carbonates produced through urea hydrolysis, denitrification or ammonification to be truly sustainable, recycled alternatives such as lactose mother liquor and corn steep liquor should be investigated. By-products of the MICP reaction must also be treated in a way that does not significantly increase the impact in other areas (carbon footprint, embodied energy, etc.).

A key component identified in the carbon footprint, embodied energy and cost for all MICP metabolic routes was the calcium source (calcium chloride). The substitution of laboratory grade calcium chloride with a commercial equivalent resulted in decreases in the carbon footprint between 26.7 and 82%. Clearly, alternatives to laboratory grade calcium sources should be investigated and included in future experiments to further enhance the sustainability of carbonates produced through the MICP process. Directions for future research for the industrialisation of MICP are shown in Figure 5.

A preliminary, high level cost analysis of carbonates produced through MICP revealed that as long as laboratory grade calcium and carbon sources are used, the cost of raw materials is significantly higher than that of traditional methods, such as carbonation, or when compared to commonly used construction binders, such as Portland Cement. In order for MICP to be viable economically, laboratory grade chemical inputs, typically adopted in the laboratory must be replaced with commercial grade alternatives.

4. Conclusions

Nature has been performing MICP for millions of years utilising different classes of microbes. As engineered MICP follows the same metabolic pathways, it is claimed
by several researchers that they are sustainable. This paper notes that engineered MICP deviates substantially from the natural one to suit the timeframe of construction projects. Thus, sustainability of the cementation process via different microbial metabolic routes has been examined. The major conclusions of our study are:

- Microbially induced calcium carbonate produced using carbonic anhydrase producing bacteria is the most environmentally sustainable route for engineered MICP applications followed by methanogens (methane oxidation) and then cyanobacteria (photosynthesis).
- The most widely used metabolic route for engineered MICP via ureolytic pathway has poor sustainability due to high carbon footprint and embodied energy of the supplied urea, as well as the eutrophication potential of ammonium waste produced during the MICP reaction.
- The sustainability of engineered MICP via ureolytic and other routes can improve significantly via utilisation of naturally found nutrient sources, recycled wastes for the source of microbial nutrients and cementation reagents as well as by the utilisation of commercial grade chemicals compared to lab grade chemicals.

The outcome of this study is highly valuable for designing the course of further research, as well as for tailor-made applications of biocement in different construction applications. Having a holistic view of the overall performance based upon the economic and environmental dividends biocement can provide, engineers and scientists can work together on bringing this novel technology out of labs and into real field scenarios.

Supplementary Materials: Details of methodologies, scenarios and boundaries, information on chemicals including unit rates have been included in the supplementary file. The following are available online at https://www.mdpi.com/article/10.3390/su132413878/s1, Figure S1: Scenarios included in analysis, Figure S2: Boundaries of Environmental Assessment (a) Biologically precipitated calcium carbonate (b) Laboratory grade calcium carbonate, Table S1. Summary of inputs/outputs for production of 1 kg of CaCO₃ through microbial route, Table S2. Unit Rates for Laboratory Grade Chemicals, Table S3. Unit Rates for Commercial Grade Chemical Replacements.

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