



ה מכון לכימיה  
Institute of Chemistry

האוניברסיטה העברית בירושלים  
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Prof. Xiaoyan Cao  
Review Editor  
Jove

Dear Professor Cao,

Thank you for your letter concerning our manuscript entitled “**Calcium carbonate formation in the presence of biopolymeric additives**” (JoVE59638). We are grateful for the reviewers for their comments and we have now fully addressed all the editorial and reviewers' comments and queries and the detailed response is attached below.

On behalf of my co-author, David Azulay, I thank you for considering our manuscript for publication in *Jove*.

Sincerely,  
Liraz Chai

*Liraz chai*



Detailed response to the editorial comments:

1. Please take this opportunity to thoroughly proofread the manuscript to ensure that there are no spelling or grammar issues. The JoVE editor will not copy-edit your manuscript and any errors in the submitted revision may be present in the published version.

Response: Done.

2. Please revise lines 31-32 and 55-56 to avoid previously published text.

Response: We have revised lines 31-32, 55-56.

3. Please obtain explicit copyright permission to reuse any figures from a previous publication. Explicit permission can be expressed in the form of a letter from the editor or a link to the editorial policy that allows re-prints. Please upload this information as a .doc or .docx file to your Editorial Manager account. The Figure must be cited appropriately in the Figure Legend, i.e. "This figure has been modified from [citation]."

Response: The copyright permission is uploaded. Figure legends 3,4 have been changed accordingly.

4. Please rephrase the Introduction to include a clear statement of the overall goal of this method.

Response: We have revised the goal statement (lines 63-67).

5. Please revise the protocol text to avoid the use of any personal pronouns (e.g., "we", "you", "our" etc.).

Response: Done.

6. Please revise the protocol to contain only action items that direct the reader to do something (e.g., "Do this," "Ensure that," etc.). The actions should be described in the imperative tense in complete sentences wherever possible. Avoid usage of phrases such as "could be," "should be," and "would be" throughout the Protocol. Any text that



cannot be written in the imperative tense may be added as a “Note.” Please include all safety procedures and use of hoods, etc. However, notes should be used sparingly and actions should be described in the imperative tense wherever possible. Please move the discussion about the protocol to the Discussion.

Response: Done.

7. [1.1.2.1](#): Please specify the amount of powder ammonium carbonate added

Response: We have changed the text in line 156 (step 1.1.2.1), to account for our accurate actions. We do not weigh the ammonium carbonate, but rather fill in the well, as is now instructed.

8. [1.1.2.2](#): Please list an approximate volume to prepare.

Response: We have added a note to line 162 (step 1.1.2.2), advising the reader to prepare a 10 mL stock solution.

9. Please number the figures in the sequence in which you refer to them in the manuscript text.

Response: Done.

10. Discussion: Please discuss critical steps of the protocol.

Response: Critical steps of the protocol have been added in lines 379, 389-394.

11. Please revise the Table of Materials to include the name, company, and catalog number of all relevant supplies, reagents, equipment and software in separate columns in an xls/xlsx file. Please sort the items in alphabetical order according to the name of material/equipment.

Response: An updated list is now uploaded.



## Detailed response to the reviewers' comments.

### Reviewer: 1

The presented manuscript is well written and contains all the necessary information to perform the described experiment.

Response: We thank the reviewer for the positive feedback.

### Reviewer: 2

In this, paper Azulay [et.al](#) present their results on biomineralization of calcium carbonate in the presence of biopolymers. The motivation of this research is quite interesting and the subject itself is very relevant to biomineralization. Overall, this work is very original and in my opinion, the results presented in this study are significant for understanding  $\text{CaCO}_3$  biomineralization. I can only come up with a few arguments that I think the authors should correct this well-written article:

- 1) The authors should briefly give a note on the thermodynamic stability of each crystalline form of  $\text{CaCO}_3$  this is a key point the crystallization of calcium carbonate.
- 2) Another comment that the authors should give kinetics details on the bio mineralization of calcium carbonate.
- 3) Figure 5- Raman peaks should be assigned to vibrational modes of  $\text{CaCO}_3$ .

In conclusion, I recommend the publication of this paper after the authors' response to the comments raised in this report.

Response: we thank the reviewer for the positive feedback, and our answers are outlined below.

1. We have included a reference to the thermodynamic stability of the different polymorphs of calcium carbonate in the original text (lines 43-45).
2. We thank the reviewer for this comment. Although a kinetic study is out of the scope of this protocol, we have now included this suggestion in the discussion (lines 403-406).
3. We thank the reviewer for this note, and we have now assigned the peaks on the Raman spectrum in Figure 5.