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Vineeta Bajaj, Ph.D. Review Editor, JoVE

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Resubmission Invited Manuscript

Dear Dr. Bajaj and Editorial Board:

We thank you for the further careful examination of our manuscript entitled "Focused Ion Beam Lithography to Etch Nano-architectures into Microelectrodes" by Shreya Mahajan, Jonah A. Sharkins, Allen H. Hunter, Amir Avishai, and myself, for consideration for publication in the Journal of Visualized Experiments (JoVE).

Please find below our detailed reply to the editors, with point-by-point responses and references to changes in the manuscript. For your convenience, the editor's comments are copied in bolded font, and changes to the revised text were incorporated by track changes throughout for your ease of finding the changes.

Please contact us if there is anything we can do to help make your final decision a favorable one. Thank you for the opportunity to contribute to this exciting body of work!

Best regards,

Evon S. Ereifej, Ph.D.

Editorial comments:

1. The protocol section doesn't show how this is done. The title should be in alignment with the protocol being presented. Please remove these words from the title.

The title has been modified to remove "for Improving Recording Quality and Reducing Neuroinflammation".

2. The length of the protocol section cannot be more than 10 pages including headings and spacing. Presently the protocol is around 11 pages. Please combine shorter protocol steps so that individual steps contain only 2-3 actions per step.

We have combined shorter protocol steps/sub-steps together to ensure that the protocol is no more than 10 pages (currently it is ~7 pages).

3. The hard-cut limit for the highlighted section is than 2.75 pages including headings and spacings. Presently this is around 6 pages. Please adjust the highlights accordingly. Also, if a substep is highlighted, the step/heading associated with it needs to be highlighted as well. So maybe consider removing some of the subheadings and consider combining shorter protocol step so there are 2-3 actions per step and no more than 4 sentences per step.

We have taken all of the advice from the editor to ensure that the highlighted section of the protocol meets the 2.75 page limit.

4. We cannot have more than one note following a step or a sub step. Moved here instead. Please check.

The new placement of the note makes sense, thank you.

5. Some of the details can be moved to the discussion section.

We took the editor's advice and removed this note from the protocol section and ensured that the information is in the discussion section instead.

6. Please include a note stating what is the significance of home staging.

We included a note stating that the importance of home staging is, "Running the home stage operation is a preventative step to ensure the stage axis are read correctly by the software and the microscope is in good condition."

7. Notes cannot be filmed so highlights are removed.

Thank you for this tip, we removed the highlights from all notes in the "to be filmed" sections.

8. We cannot have commercial terms in the manuscript. Please use generic term instead. Please move the commercial term to the Table of Materials and refer to the table wherever needed.

We replaced the term "Nav-Cam" with "navigation camera". The Nav-cam is an optical camera mounted on the microscope that is used to assist with sample navigation, especially when you have more than one sample in the chamber. The term Nav-Cam is probably specific to FEI/ThermoFisher, but there are similar devices on other microscope brands, so replacing with a generic term will be suitable for the protocol.

9. So this is "xT Align Feature" the inbuilt command?

Yes, the xT align feature is built in to the software. It exists on all FEI microscopes made in the last 15 years or so. We added the phrase "built in" before the "xT Align Feature" in the protocol to note that.

10. We cannot have commercial terms in the manuscript. Please move this to the table of materials and use generic term instead. Please refer to the table of materials wherever applicable.

We replaced all reference to "Nanobuilder software" to simply "software", "FEI microscope" to "SEM microscope", "Michigan-style silicon microelectrodes" to "single shank silicon microelectrodes" and removed all model and product numbers from within the protocol.