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June 6, 2011

Dear Meghan,

We are submitting a manuscript titled, "Live-cell imaging of migrating cells expressing fluorescently tagged proteins in a three-dimensional matrix", by Wenting Shih and Soichiro Yamada for consideration for publication as a article in the Journal of Visualized Experiments. This work has not been submitted to, or published in any form in another journal.

In a three-dimensional (3D) matrix, epithelial cells migrate with a morphology significantly different from cells migrating on a culture dish. Yet, we know very little about how cells migrate in a 3D matrix. This is in part due to the absence of accessible experimental protocols to visualize and analyze 3D cell migration. In this article, we outline our protocol to analyze 3D cell migration using a standard confocal microscopy system. This manuscript contains detailed protocols of the production of fluorescently labeled cell lines, the setup of live-cell imaging, FRAP, and 3D traction force experiments. We believe that wide dissemination of this protocol via JoVE will aid our understanding of 3D cell migration.

Thank you for considering this work for publication in the Journal of Visualized Experiments.

Sincerely,

A handwritten signature in black ink, appearing to be "Soichiro Yamada".

Soichiro Yamada