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Automatic Multi-functional Integration Program (AMFIP) towards All-optical Mechano-electrophysiology Interrogation

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Qin Luo[1], Justin Zhang[2], Miao Huang[3], Gaoming Lin[1], Mai Tanaka[4], Sharon Lepler[4], Juan Guan[5], Dietmar Siemann[4], Xin Tang[3]

[1]Department of Electrical and Computer Engineering, Herbert Wertheim College of Engineering, University of Florida, Gainesville, Florida, United States of America;

[2]Department of Electrical and Computer Engineering, University of California, Santa Barbara, California, United States of America;

[3]Department of Mechanical and Aerospace Engineering, Herbert Wertheim College of Engineering, UF, United States of America;

[4]Department of Radiation Oncology, College of Medicine, University of Florida, Gainesville, Florida, United States of America;

[5]Department of Physics, College of Liberal Arts and Sciences, University of Florida, Gainesville, Florida, United States of America

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ABSTRACT

Automatic operations of multi-functional and time-lapse live-cell imaging are necessary for the biomedical science community to study active, multi-faceted, and long-term biological phenomena. To achieve automatic control, most existing solutions often require the purchase of extra software programs and hardware that rely on the manufacturers' own specifications. However, these software programs are usually non-user-programmable and unaffordable for many laboratories. To address this unmet need, we have developed a novel open-source software program, titled Automatic Multi-functional Integration Program (AMFIP), as a new Java-based and hardware-independent system that provides proven advantages over existing alternatives to the scientific community. Without extra hardware, AMFIP enables the functional synchronization of the μManager software platform, the Nikon NIS-Elements platform, and other 3rd party software to achieve automatic operations of most commercially available microscopy systems, including but not limited to those from Nikon. AMFIP provides a user-friendly and programmable graphical user interface (GUI), opening the door to expanding the customizability for myriad hardware and software systems according to user-specific experimental requirements and environments. To validate the intended purposes of developing AMFIP, we applied it to elucidate the question whether single cells, prior to their full spreading, can sense and respond to a soft solid substrate, and if so, how does the interaction depend on the cell spreading time and the stiffness of the substrate. Using a CRISPR/Cas9-engineered human epithelial Beas2B (B2B) cell line that expresses mNeonGreen2-tagged mechanosensitive Yes-associated protein (YAP), we show that single B2B cells develop distinct substrate-stiffness-dependent YAP expressions within 10 hours at most on the substrate, suggesting that cells are able to sense, distinguish, and respond to mechanical cues prior to the establishment of full cell spreading. In summary, AMFIP provides a reliable, open-source, and cost-free solution that has the validated long-term utility to satisfy the need of automatic imaging operations in the scientific community.

ATTACHMENTS

S1 Protocol.pdf

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