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Polyploidy in Cardiomyocytes: Machine Learning for Single Cell Analyses

Jeremy Scott Mitchell; Mark Alan Sussman, PhD

Department of Biology, San Diego State University, San Diego, California

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Introduction

Understanding the dynamics of cardiomyocyte ploidy is of paramount importance for deciphering the cellular mechanisms propelling cardiac development,[1] regeneration,[2] and disease.[3] Cardiomyocytes possess diverse ploidy levels that can vary significantly among the cell population. Both polyploidization and ploidy reduction are well documented in liver hepatocytes in response to injury and repair, but these processes are poorly understood in the context of the heart. To address this gap in scientific understanding, a novel approach using single-cell, flow cytometry–based imaging and software-driven machine learning algorithms has been employed.

Current Limitations

The existing technical limitations associated with traditional histologic or microscopic examination of selected cell populations include potential subjectivity and low-throughput data collection. These methods often rely on small sample sizes and can introduce bias, affecting the accuracy and precision of experimental findings. In addition, precise and accurate nuclear ploidy counts for cardiomyocytes in mass have not been easy to obtain.

Recent Developments

In the present study, the Amnis ImageStream X Mark II imaging flow cytometer (Cytek Biosciences), IDEAS software analysis, and machine learning were used to revisit ploidy in cardiomyocytes. Amnis ImageStream flow cytometry enables photomicroscopy of individual cells and fluorescence image acquisition (Fig. 1), leading to high-throughput data collection from thousands of cells in a single experimental session. Photomicrographs were processed using IDEAS Software's machine learning module to obtain target cellular data. Configured to perform population-based analysis of thousands of cells, this software extracted key features from the images, allowing for precise measurements of nuclear ploidy in each cardiomyocyte. This approach greatly improves analysis compared to traditional methods by expanding the population size, avoiding subjective bias, and enhancing accuracy.

Future Directions

With this trinity of tools, the groundwork has been laid for precise quantification of nuclear polyploidy in cardiomyocytes. This technique offers swift and reliable processing of data sets consisting of single-cell images and em-

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Corresponding author: Mark Sussman, PhD, Biology Department, 5500 Campanile Dr, San Diego, CA 92182 (msussman@sdsu.edu)

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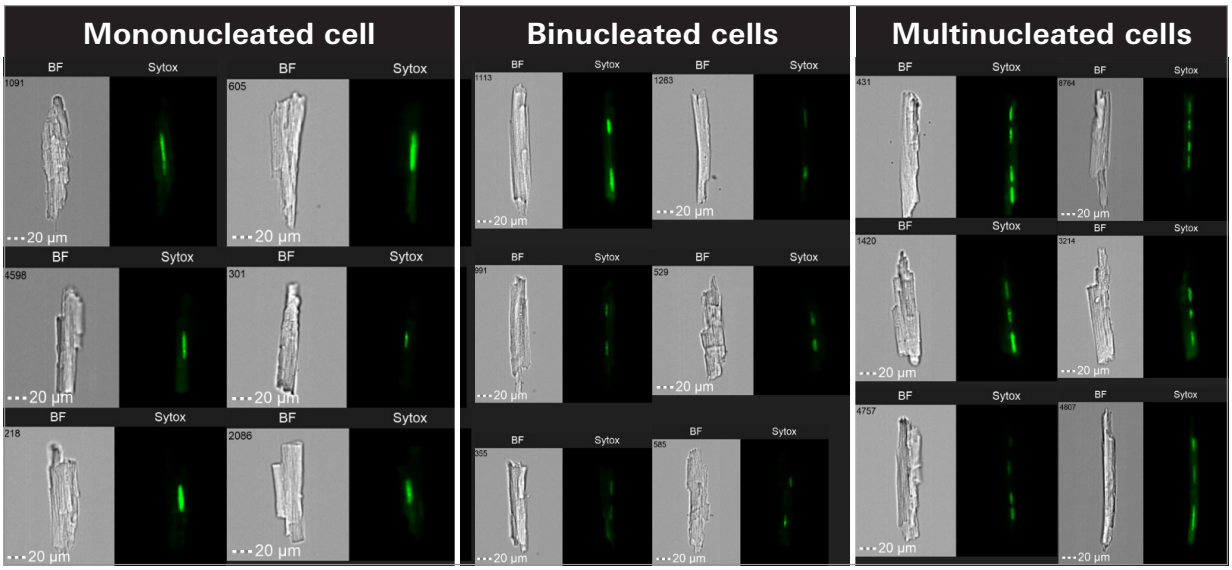

Fig. 1 Images captured using the Cytek Amnis ImageStream X Mark II imaging flow cytometer of enzymatically isolated, single-cell adult mouse cardiomyocytes. Images were analyzed using IDEAS software to determine nuclear count and ploidy level, using a custom-scripted machine learning analysis module. From left to right: mononucleated cells, binucleated cells, and multinucleated cells.

powers population-based studies of polyploidy, paving the way for advanced explorations of ploidy dynamics in myocardial biology. This methodology will be used to reevaluate ploidy response to myocardial injury, repair, and regeneration.

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References

1. Broughton KM, Sussman MA. Adult cardiomyocyte cell cycle detour: off-ramp to quiescent destinations. *Trends Endocrinol Metab.* 2019;30(8):557-567. doi:10.1016/j.tem.2019.05.006
2. Broughton KM, Sussman MA. Myocardial regeneration for humans—modifying biology and manipulating evolution. *Circ J.* 2017:81(2):142-148. doi:10.1253/circj.CJ-16-1228
3. Broughton KM, Ebeid D, Esquer C, Wang B, Moshref M, Sussman M. Polyploidy increases in murine cardiomyocytes following myocardial infarction. *Circ Res.* 2019;125:A401. doi:10.1161/res.125.suppl_1.401

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