Multiphoton Flow Cytometer for High Throughput Analysis of Multicellular Aggregates Like Pancreatic Islets

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Technology description

Regenerative medicine requires a practical, high throughput method of assessing the cellular components of multicellular entities before transplantation. Such a method would have many clinical and research applications. For example, no means currently is available to efficiently assess the functional potential of pancreatic islet cells prior to transplantation without disrupting the islet. Additionally, the characteristics of embryoid bodies used for stem cell research cannot be screened easily prior to dissociation of the bodies.

Flow cytometers are well-established research and clinical instruments that can provide automated, quantifiable and verifiable data on cellular phenotype, differentiation and metabolic state. However, multicellular aggregates such as islets can be disrupted under the forces generated in a typical flow cytometer. Also, conventional flow cytometry monitoring systems are not well-suited for the analysis of three-dimensional cellular structures.

Multiphoton laser-scanning microscopy (MPLSM) is a fluorescence imaging technique that allows imaging of living tissue up to a depth of 600 µm. This technique is useful for live cell imaging studies of large, dense structures such as an islet. However, MPLSM has not been used for high throughput analysis of cells. UW-Madison researchers have developed a system that combines the high throughput characteristics of flow cytometry with the capabilities of MPLSM. This multiphoton flow cytometry system (MPFC) enables deep, high resolution images of large diameter cells and aggregates.

The multiphoton laser can excite intrinsic cellular fluorophores such as NADH, FAD and collagen, allowing both spectral and lifetime data to be acquired. This information then can be used to reveal information on cellular processes like metabolism, viability and the functional potential of cells, pancreatic islets, embryoid bodies and other entities.
The Wisconsin Alumni Research Foundation (WARF) is seeking commercial partners interested in developing a multiphoton flow cytometer that combines microfluidics with multiphoton microscopy to provide high throughput analysis of the cellular components of multicellular entities (i.e., pancreatic islets, embryoid bodies or model organisms). The instrument is capable of detecting fluorescence

signals, including very weak autofluorescence associated with certain metabolites and extracellular matrix proteins.

Additional Information

Buschke D.G., Squirrell J.M., Vivekanandan A., Rueden C.T., Eliceiri K.W. and Ogle B.M. 2014. Noninvasive Sorting of Stem Cell Aggregates Based on Intrinsic Markers. Cytometry A. 85, 353-358.
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Buschke et al. 2013. Large Particle Multiphoton Flow Cytometry to Purify Intact Embryoid Bodies Exhibiting Enhanced Potential for Cardiomyocyte Differentiation. Integr. Biol. (Camb.) 5, 993-1003.
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Application area

High throughput characterization of the inner cells of multicellular entities
Noninvasive labeling, characterization and high throughput sorting of cells prior to transplantation

Advantages

Combines imaging and fluorescence measurements to provide more useful analysis metrics
Enables sophisticated automated cell sorting
Controls flow rates and minimizes shear forces, preventing damage to multicellular aggregates

Institution

Wisconsin Alumni Research Foundation

Inventors

Brenda Ogle
Kevin Eliceiri
Matthew Hanson
Luis Fernandez

联系我们

叶先生

电 话：021-65679356

手 机：13414935137

邮 箱：yeyingsheng@zf-ym.com