FORMULATRIX

“The Mantis reduces variability allowing for more efficient screen development”

- Christopher Hubbs, Scripps Research Institute

Cell dispensing in a high-content, primary neuron based, screening platform using The Mantis Liquid Handler

Introduction

Alterations in synaptic connections are implicated in nearly all brain disorders. In particular, synapse loss is a particularly profound problem in brain disorders that attack cognitive function, such as schizophrenia and Alzheimer’s disease. As part of a research program aimed at discovering novel mechanisms that trigger increases in neural connectivity as a strategy to combat these illnesses, a high content, primary neuron based, screening platform has been developed to facilitate the discovery of novel chemical probes that trigger increases in synaptic connectivity. These probes will serve as developmental platforms for future generations of drugs that treat a wide range of brain disorders.

In order to ensure reproducible batch to batch results as part of a high content screening platform, it is imperative that cells are plated consistently and that the cells are viable for the duration of the experiment. This application note describes an experimental procedure that highlights the suitability of The Mantis liquid handler as a precise, reliable, and robust solution for dispensing even the most sensitive primary neuronal cells.

Materials

1x PDL coated 1536 well Aurora Microplate

50 mL culture medium containing:

- 1X Neurobasal-A medium
- 2.5% Glutamax-I
- .02% Gentamicin Reagent
- 2% B-27 Supplement
- 10 uM FUDR

$5x10^6$ primary cortical cells extracted from P0 mouse pups

Formulatrix Mantis Liquid Handler

Eppendorf 5810 R Centrifuge

GE IN Cell Analyzer 6000 laser-based confocal imaging platform

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Methods

1. Neuronal cells were plated across 1536 well Aurora microplate
2. 15 µL of culture medium containing 3 x 10^3 cells was dispensed to each of the 1536 wells
3. Plates were centrifuged at 200 g for 1 min
4. Cells were incubated for 7 days without feeding
5. Cells were counted via DIV7 Phase Contrast using IN Cell Analyzer 6000

Results

Following a 7 day incubation, the average phase cell count was comparable to a 96 or 384 well plate and the CV of cell count uniformity across the entire 1536 well plate was less than 10%.

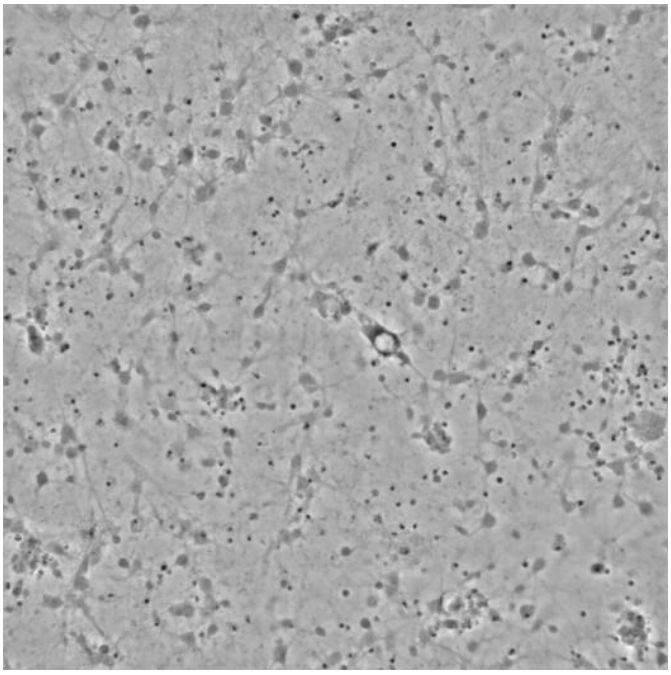

Figure 1. Primary neuronal cells in a single well of a 1536 well Aurora microplate

Conclusion

The Formulatrix Mantis consistently dispenses cells, even in low volume 1536 well format. Moreover, through the use of gentle, low shear force, dispensing technology, the instrument promotes prolonged in vitro cell viability.

Acknowledgements

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