# DispenCell versus Limiting Dilution for efficient and affordable single-cell cloning

### Introduction

• Generating monoclonal cell lines is a critical early step for a variety of high added value applications, such as monoclonal antibody production or genome editing<sup>1</sup>. **Concurrent isolation of several clones** is often desired. Downstream steps strongly depend on the quality of the isolated clones.

• "Limiting Dilution", which is based on Poisson's law, is the reference method to obtain monoclonal cell lines in multi-well plates. It is easy to implement yet it has the drawback of multiple empty wells. This limits the number of growing colonies isolated in one experimental round. A supplemental time-consuming step is needed to verify the monoclonality of growing colonies.



DispenCell is an impedance-based automated pipetting robot, enabling gentle and traceable single-cell dispensing<sup>2,3,4</sup>.

We present here a side-by-side comparison between DispenCell and Limiting Dilution for single cell cloning.

## Results



in blue the wells with growing colonies after isolation by Limiting Dilution at 0.3 cells/well (Fig1 left) or with DispenCell (Fig1 right). 0.3 cells/well was chosen as working dilution as it yields less than 10% of wells with polyclonal cell population. With DispenCell, 5x more wells contain growing colonies.

• The well-plates represent

*Figure 1:* Performance of DispenCell as compared to Limiting Dilution: experimental data.

• The pie charts show the distribution of the type of well content (empty, polyclonal, monoclonal) as modelled by Poisson's law (Fig 2 left), and the average type of well content obtained with DispenCell (Fig 2 right, 4% empty, 95% monoclonal). 74% wells are empty after Limiting Dilution and with DispenCell 95% of the wells contain a single cell.



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Monoclonal

*Figure 2:* Performance of DispenCell as compared to Limiting Dilution: statistical model

# SEED's DispenCell versus Limiting Dilution: Generalities



### **Material & Methods**

Single cells were isolated from a pool of eGFP-modified CHO-S cells. The same suspension was used with DispenCell and Limiting Dilution for side-by-side comparison. After seeding, the plates were placed in a  $CO_2$  incubator at 37°C and imaged at days 0, 4, 7, and 15 with the CloneSelect Imager (Molecular Devices). The wells reaching a cell confluence of >25% day 15 are considered.

DispenCell is the efficient, convenient, easy-to-use & cost-effective single cell dispensing alternative to limiting dilution.

### **Contact us**

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#### References

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