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## Overview

#### PCR

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The ability to sort cells by type or physical properties is a valuable tool in many synthetic biology labs. Prior to analysis or in order to perform specialised protocols, creating homogenous cell suspensions from a mixture is necessary. In addition to sorting cells, the removal of cell fragments, activated magnetic particles or unwanted debris through sorting also makes up a key part of purification protocols.

This microfluidic chip design carries out cell sorting as a cell suspension is passed through it. Cells are sorted based on size and pushed to the periphery of the channel. These cells are then carried away from the main solution through the two periphery outputs, and the cell-free solution can be collected from the central output. Chip Design



Flow Layer



# Flow Layer

## **Milling Guidelines**



**Flow Layer** 

### Milling Guidelines

For a comprehensive guide to milling click <u>here</u>. For a list of tool parameters click <u>here</u>.

#### Notes

- 1. This chip should be milled on medium or thick polycarbonate (2.75mm < Z<sub>Polycarbonate</sub>).
- 2. This chip requires thick PDMS (1.0mm <  $Z_{PDMS}$  < 1.5mm)

All the required SVGs for milling this chip are provided in the ZIP file. The layer, depth, and tool required for each SVG is listed in the file name. Below is a key describing how to read an SVG file name:



### Milling Instructions

Mill the layers in the order they are listed with the correct depths and using the correct tools.

Flow Layer		
Order	Layer Name	
1.	F_1700_64	
2.	F_PORTS_8	
3.	F_255_100	
4.	Border	

# **Testing Protocol**

## Flow Layer Setup



Inputs				
Name	Liquid	Flow Rate		
А	Cell Suspension	0.3 mL/hour		

Outputs		
Name	Liquid	
а	Sorted Cells	
b	Cell-free Solution	
с	Sorted Cells	

### Testing the Chip

#### Setup

- 1. Prepare 1 syringe with coloured water
- 2. Attach the syringe to Input A
- 3. Attach your output tubing to Outputs a,b and c; they should connect to eppendorf tubes for collection of the sorted cells and cell-free fluid

#### Running the chip

- 1. Begin flowing the cell suspension into the chip at a rate of 0.3 mL/hour
- 2. Allow the fluid to completely fill the channels and flow out of the three outputs
- 3. Continue to flow liquid through the channels for thirty seconds, pause the syringe pump and dispose of the original eppendorf tubes
- 4. Attach new eppendorf tubes to the outputs and restart the fluid flow

#### Cleaning the Chip

- 1. Remove the eppendorf tubes from your output tubing
- 2. Disconnect your output and input tubing carefully avoiding spills
- 4. Clean the chip following the oil & water protocol listed <u>here</u>
- 5. Store your chip as detailed in the cleaning protocol