Fluorescence

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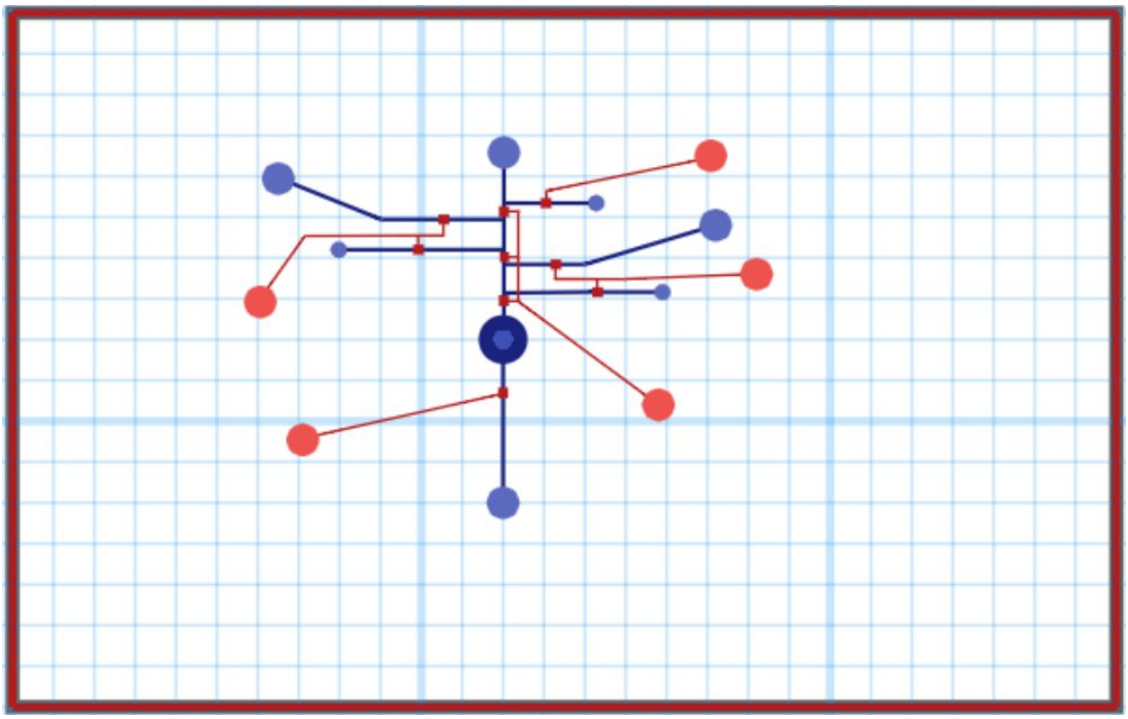
Overview

Fluorescence

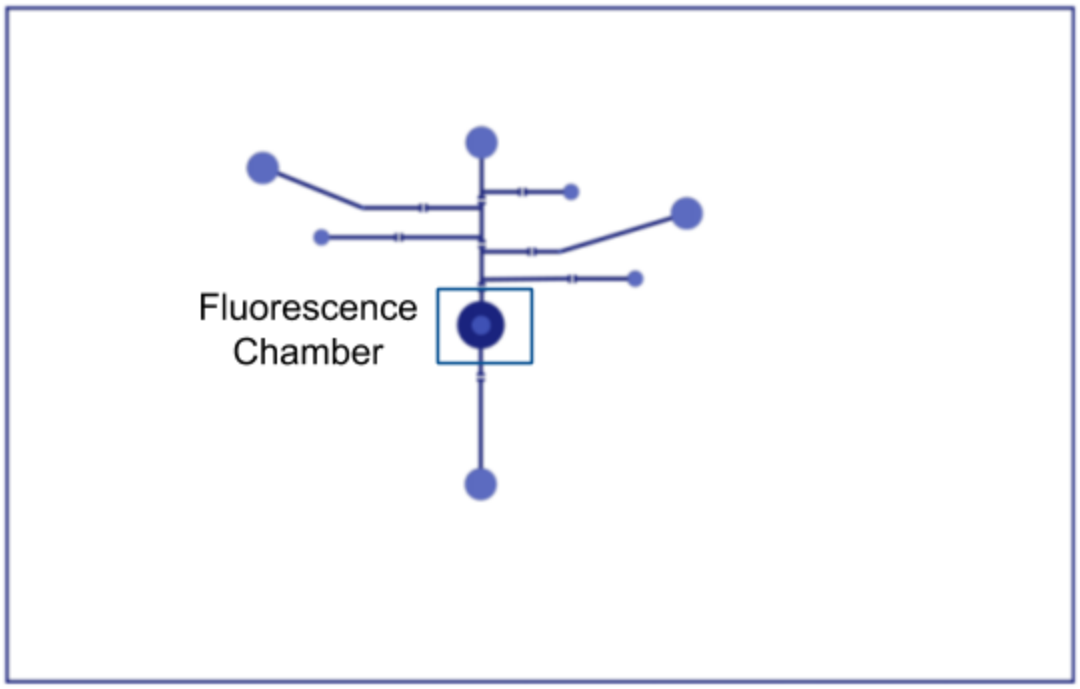
Designed by Dinithi Samarasekera
Date Completed: 10/25/2017

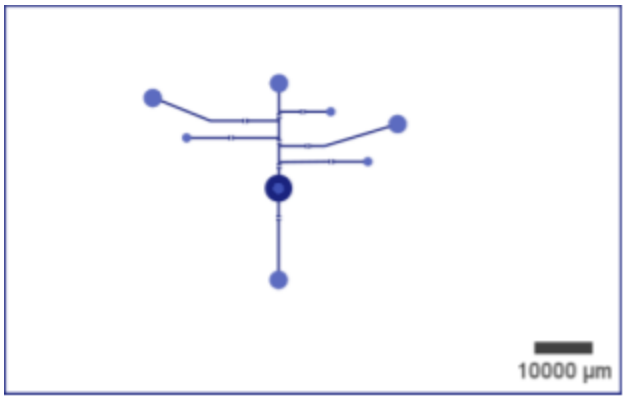


Fluorescence is commonly used as a quantifiable marker in synthetic biology procedures. Following the completion of a protocol, for example an assay test, samples are tested in a plate reader and the results analysed.

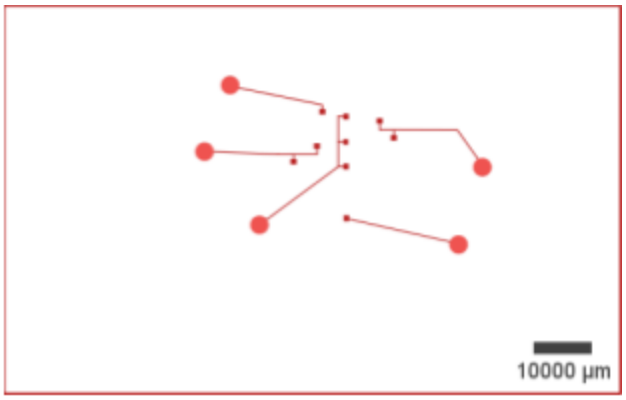
This microfluidic chip is designed to perform a Cell-free Toehold test, however it can be adapted to perform alternative fluorescent tests as desired. The metering primitive dispenses accurate ratio volumes of various substances. These are pushed into the central well which matches the dimensions of a 96-well plate. The chip can then be read in a plate reader with the use of an adapted skirt.

Chip Design



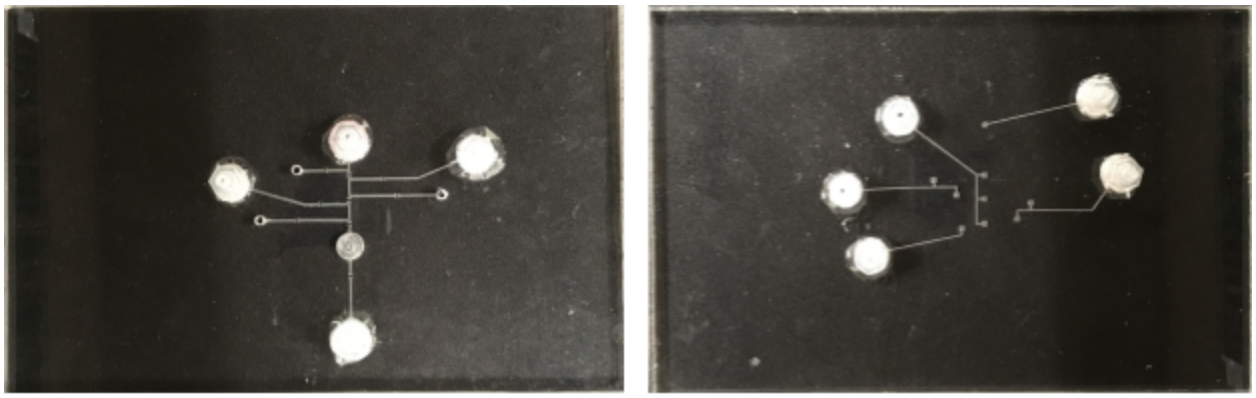


Flow Layer



Control Layer

Milling Guidelines



Flow Layer | Control Layer

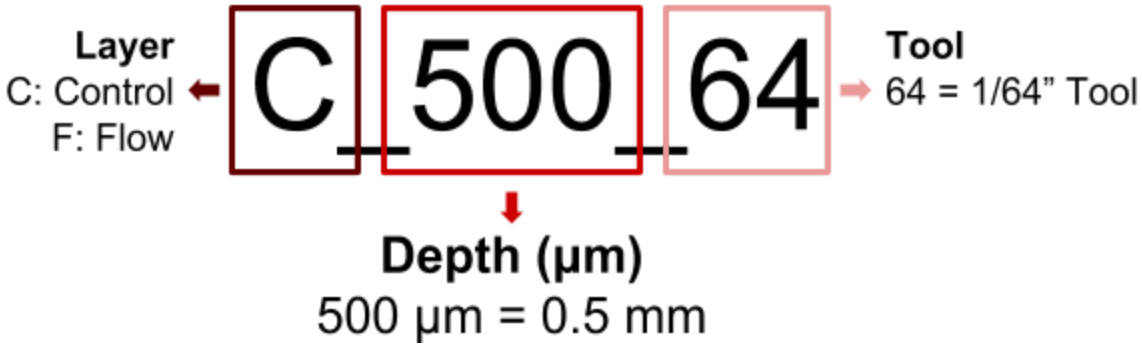
Milling Guidelines

For a comprehensive guide to milling click here. For a list of tool parameters click here.

Notes

1. This chip should be milled on medium or thick polycarbonate (2.75mm < $Z_{Polycarbonate}$).
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

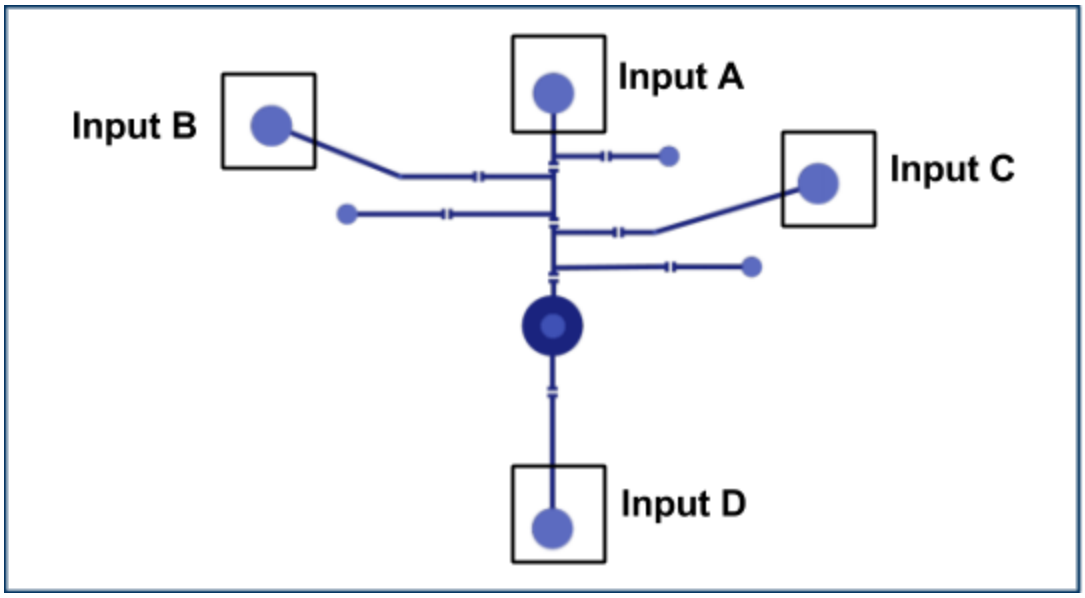
Mill the layers in the order they are listed with the correct depths and using the correct tools.

Flow Layer	
Order	**Layer Name**
1.	F_CHAMBER_16
2.	F_500_64
3.	F_PORTS_8
4.	Border

Control Layer	
Order	**Layer Name**
1.	C_300_100
2.	C_PORTS_8
3.	Border

Testing Protocol

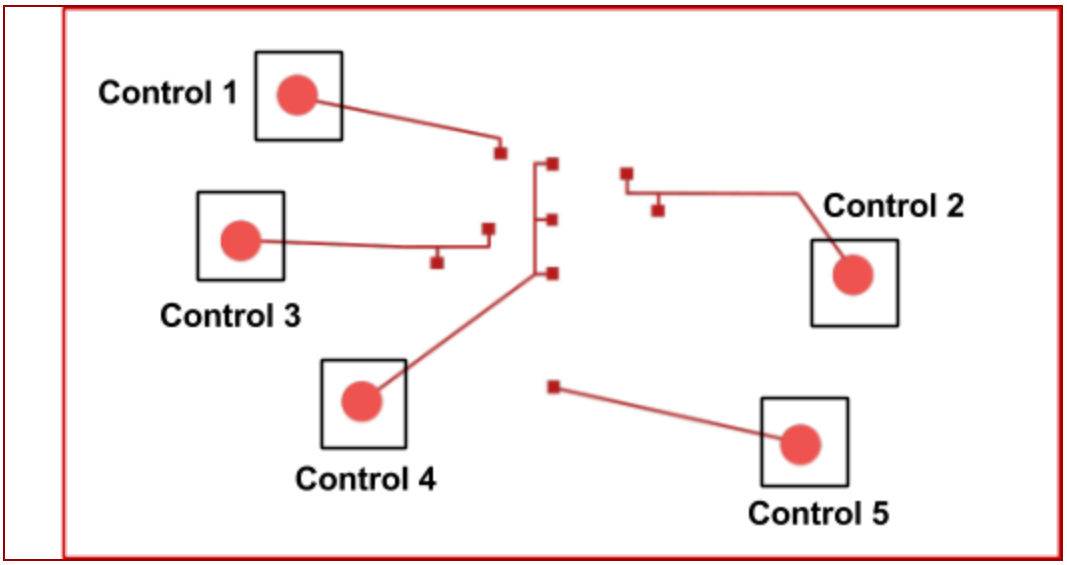
Flow Layer Setup



Inputs		
Name	**Liquid**	**Flow Rate**
A	Oil	0.2 mL/hour
B	Toehold	0.2mL/hour
C	Trigger RNA	0.2mL/hour
D	Master Mix	0.3mL/hour

Outputs	
Name	**Liquid**
a	Fluorescent Solution

Control Layer Setup



Testing the Chip

Setup

1. Prepare 4 syringes
    a. 3 filled with colored water
    b. 1 filled with mineral oil
    c. 5 empty control syringes
2. Attach the syringes containing coloured water to inputs B,C and D
3. Attach the syringe containing oil to input A
4. Attach the control syringes to all five control layer outputs

Running the chip

5. Open Controls 1 ,2 and 3; you should feel significant resistance while you open these control valves
6. Begin flowing Inputs A, B, and C
7. When the metered segment for each input is filled, stop the fluid flow and close their corresponding Control line
8. Open Control 4 and begin flowing Input A
9. When the oil is close to the final valve before entering the chamber, stop flowing Input A and close Control 4
10. Open Control 5 and flow Input D until the chamber is filled to a satisfactory level
11. Halt the flow of Input D and then close Control 5

Cleaning the Chip

12. Disconnect all syringes carefully
13. Clean the chip following the oil & water protocol listed here
14. Store your chip as detailed in the cleaning protocol