University of Illinois at Chicago

Doctoral Thesis

Devices for Biological Systems: On-Chip Horizontal Gene Transfer and 3D-Printed Microfluidic Applications

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Department of Bioengineering

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Declaration of Authorship

I, Martin D. BRENNAN, declare that this thesis titled, 'Devices for Biological Systems: On-Chip Horizontal Gene Transfer and 3D-Printed Microfluidic Applications' and the work presented in it are my own. I confirm that:

- This work was done wholly or mainly while in candidature for a research degree at this University.

- Where any part of this thesis has previously been submitted for a degree or any other qualification at this University or any other institution, this has been clearly stated.

- Where I have consulted the published work of others, this is always clearly attributed.

- Where I have quoted from the work of others, the source is always given. With the exception of such quotations, this thesis is entirely my own work.

- I have acknowledged all main sources of help.

- Where the thesis is based on work done by myself jointly with others, I have made clear exactly what was done by others and what I have contributed myself.

Signed:

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Date:

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"Thanks to my solid academic training, today I can write hundreds of words on virtually any topic without possessing a shred of information, which is how I got a good job in journalism."

Dave Barry

Abstract

Faculty Name

Department of Bioengineering

Doctor of Philosophy in Bioengineering

Devices for Biological Systems: On-Chip Horizontal Gene Transfer and 3D-Printed Microfluidic Applications

by Martin D. BRENNAN

The Thesis Abstract is written here. The page is kept centered vertically so can expand into the blank space above the title too. . .

This is a test

Acknowledgements

The acknowledgements and the people to thank go here, don't forget to include your project advisor. . .

Contents

List of Figures

List of Tables

Abbreviations

LAH **L**ist **A**bbreviations **H**ere

Physical Constants

$$\text{Speed of Light} \quad c \quad = \quad 2.997\ 924\ 58 \times 10^8 \text{ ms}^{-S} \text{ (exact)}$$

Symbols

a	distance	m
P	power	W (Js^{-1})
ω	angular frequency	rads^{-1}

For/Dedicated to/To my. . .

Chapter 1

Introduction and Background

1.1 Main Section 1

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1.2　Main Section 2

1.2.1　Subsection 1

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1.3　Horizontal Gene Transfer

1.3.1　History and discoveries

In 1928 Griffith reported on the discovery of the 'transforming principal' where mice that were injected with a non-virulent strain resulted in a lethal infection when injected along with a heat killed virulent strain[? , Griffith1928] Both virulent and non-virlent strains could then be isolated from the blood of the dead mouse. Somehow the presence of the virulent strain, although dead, passed information that allowed the non-virulent strain to transform and become virulent. A subsequent study by Avery, McLeod and McCarty demonstrated that the presence of an overlooked substance, DNA, was responsible for re-programing strains[? , Avery1944] At the time it was believed that some yet to be discovered protein complex was the carrier of genetic information, but after this result scientists began looking more closely at DNA. In 1951 Freeman demonstrated that a phage could re-program *Corynebacterium diphtheriae* from non-virulent to virulent[?]. Hershey and Chase found that the DNA is incorporated into the cell rather than the protein from the phage[?]. They confirmed this by producing two groups of phages, one with a radio labeled protein and one with radio labeled DNA. Watson, one of the scientist that discovered the structure of DNA was a phage scientist as well.

1.3.2   Mechanisms

Horizontal gene transfer (HGT) or (sometimes lateral gene transfer) describes the introduction of genes from an outside source which is distinct from vertical gene transfer where genes are passed from mother to daughter cells. The mechanisms of HGT are transformation, transduction and conjugation.

Transduction involves a phage which carries and infects the host cell by injecting the DNA or RNA into the cytoplasm.

In **conjugation** genetic material is passed directly during cell-to-cell contact.

1.3.3   Transformation

Transformation is the uptake of exogenous DNA from the environment by a cell[**?**]. Many species naturally transformable species of bacteria have been reported such as *Streptococcus pneumoniae*, Species such as *Streptococcus pneumoniae* undergo transformation naturally. Transformation was first described in *Streptococcus pneumoniae* [**?**]. In transformation the recipient cell controls the process and expresses all the proteins required

Chapter 2

Materials and Methods

2.1 3D Printed Microfluidic Devices

2.1.1 Fabrication of the 3D-printed Part

The fluidic distribution networks were designed in Autocad in 2D and then extruded to
3D with Blender. Because the channel path length to each well varied the flow resistance
in each channel was balanced by adjusting the width. Each distribution network services
six pillars that reach into the well of the plate leaving a 500 μm gap to the culture surface.
A pipe within a pipe design was used to allow a forward and return path in each pillar
that also created uniform flow across the diffusion membrane. The design was repeated
to create four sets that service six wells. Hose barbs were added to each inlet and outlet
port. The resulting STL file was printed by fineline prototyping with a Viper SLA
system in WaterShed XC, an ABS or PBT-like proprietary material.

2.1.2 Attachment of Membranes

Gas permeable membranes were made by compressing 10:1 mixed and degassed PDMS
between two glass plates that were spaced 100 μm apart with scotch tape. Molds were
baked at 50°C on a hot plate to avoid bubble formation. Membranes were transfered to
a transparency and placed over a cutting template and cut to size for the pillar bottoms.
To attach the membranes to the 3D printed part, a small amount of PDMS was applied
to the membranes and spread thin to act as a mortar. The membranes were left in place
to cure overnight.

2.1.3 Oxygen Characterization

PtOEPK (Pt(II) Octaethylporphine ketone) sensors were created by spinning thin films from a PtOEPK in polystyrene-toluene solution. Thin film sensors were cut and fixed to the bottom of each well with PDMS.

to each well in the network we One central input branches to central input that equalizes the flow along each path length by varying the channel width to the proximal, intermediate, and distal wells (Fig 1).

2.1.4 Subsection 1

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Chapter 3

Results

3.1 Main Section 1

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Chapter 4

Discussion

4.1 Main Section 1

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Appendix A

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Bibliography