Femtosecond Laser Microfabrication of Optofluidic Lab-on-a-Chip with Selective Chemical Etching

by

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A thesis submitted in conformity with the requirements for the degree of Doctor of Philosophy
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University of Toronto

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Abstract

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The three-dimensional (3-D) writing capability of a high repetition rate (1 MHz) fiber-amplified femtosecond laser with a wavelength of 522 nm was harnessed together with wet-chemical etching for laser-patterning of 3-D optofluidic Microsystems in fused silica glass, by the method of Femtosecond Laser Irradiation followed by Chemical Etching (FLICE). Selective chemical etching of laser irradiated glass with dilute hydrofluoric acid (HF) enabled micro-fabrication of high aspect-ratio embedded micro-channels and fine-period 3-D glass meshes in a 3-D inverted woodpile (IWP) arrangement that permitted high density lab-on-a-chip (LOC) integration of flow channels, reservoirs, glass chromatography columns, and optical circuit devices.

Optical waveguides, reservoirs, micro-channels, and IWP structures were first laser patterned and followed by selective wet etching controlled by the polarization orientation of the writing laser. With the laser polarization perpendicular to the scanning direction, the volume nanogratings were aligned perpendicular to glass surfaces to facilitate HF etching and thus created designer shaped micro-channels with the smoothest sidewall surfaces measured at present and terminated with open reservoirs. An array of vertical access holes spaced periodically apart facilitated etching of continuous and highly uniform buried channels of unrestricted length in the glass to interconnect flow channels
and reservoirs. Alternatively, laser polarization parallel to the scan direction provided low-loss optical waveguides with nanograting walls resisting the acid etching, providing a convenient one-step laser scanning process of optofluidic microsystems prior to wet etching. For the first time, dual-channel capillary electrophoresis was demonstrated by simultaneous fluorescent detection of separating dyes in a 3-D microsystem having over- and under-passing crossed channels in fused silica. In addition, an on-chip particle counting device based on capillary force to drive analytes through an embedded micro-channel into a calibrated reservoir for particle counting was designed and demonstrated. Further, a new type of glass mesh structure is presented where a 3-D IWP micro-channel array with diamond-like symmetry was integrated inside a micro-channel for capillary electrophoretic chromatography. The FLICE technique thus enables rapid prototyping of fully integrated 3-D optofluidic systems in bulk fused silica glasses for numerous applications, and these provide the groundwork and open new 3-D design approaches for advanced microsystems in the future.
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Appendix A G-code: Particle Counting Cytometer

Appendix B G-code: Multi-Level Capillary Electrophoresis Device

Appendix C G-code: IWP/Chromatographic/Waveguide Probing Device

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Bibliography
Acronyms

µ-TAS micro-total-analysis-system. 1, 10
µCP microcontact printing. 12
2-D two-dimensional. 2, 4, 6, 9, 12, 17, 19, 31, 120, 124
3-D three-dimensional. 2, 7, 9, 11, 12, 14, 15, 18, 19, 31, 35, 36, 47, 48, 71, 79, 80, 90, 91, 93, 103, 110, 117, 119, 124, 127, 129
AFM atomic force microscope. 64, 66
AOM acousto optical modulator. 43, 44
AP aperture. 43
BR buffer reservoir. 104, 107, 108, 111, 117
BW buffer waste reservoir. 104, 107, 108
CE capillary electrophoresis. 69
CEC capillary electrochromatography. 71, 72
CPA chirped-pulse amplification. 39
CS capillary system. 20, 69
CSµCP controlled sagging microcontact printing. 12
DLW direct laser writing. 3, 18, 35, 80, 90
DNA deoxyribonucleic acid. 16
DOE diffractive optical element. 18
DRIE deep reactive ion etching. 72
EDL electric double layer. 25, 28
EM electromagnetic. 18
EOF electroosmotic flow. 26 28 30 104
FHD flame hydrolysis deposition. 16
FLICE femtosecond laser irradiation with chemical etching. 2 5 9 15 19 34 36 38
  47 48 69 71 119 121 123 124 127 129
fs femtosecond. 5 8 9 14 15 19 20 31 32 34 36 38 48 49 86 88 120 129
FWHM full-width at half-maximum. 39 40
G gas. 21
HF hydrofluoric acid. 2 15 33 34 36 38 40 48 50 52 55 57 62 64 66 67 70 73
  75 78 92 94 95 97 100 106 115 119–121
HM hot mirror. 44
HWP half-wave plate. 43
IR infrared. 13 31 44 79 80 86 122 127
IWP inverted-woodpile. 5 47 48 71 73 75 78 80 121 122 124 128
KOH potassium hydroxide. 3 15 36 38 48
L liquid. 21
LBO lithium triborate. 43 44
LOC lab-on-a-chip. 1 7 9 20 23 25 31 37 39 47 48 69 71 88 90 91 93 98 99
  103 104 119 120 122 130
NA numerical aperture. 33 34 36 43 49 72 73 75 79 80 93 129
NaOH sodium hydroxide. 107
ND neutral density. 44
O over-passing. 91 104 107
OL objective lens. 45
P polarizer. 43
PC photonic crystal. 3 4 9 10 18 45 48 71 121 122 126 129
PDMS polydimethylsiloxane. 11 12 17 93 104 106 107 116
PECVD plasma-enhanced chemical vapour deposition. 16
PMMA poly(methyl methacrylate). 11, 30, 92
PSO position-synchronized output. 43
Rh 123 rhodamine 123. 91, 107, 108, 111, 112, 114, 115, 117
rms root-mean-square. 65, 72, 93, 96, 120, 128
S solid. 21
SEM scanning electron microscope. 34, 51, 64, 66, 73, 78
SFC sample flow channel. 96, 98, 100, 101
SHG second harmonic generation. 44
SL stereolithography. 12
SR sample reservoir. 100, 101, 104, 107, 108
SVC stop-valve channel. 100, 103
SW sample waste reservoir. 104, 107, 108
TC tapered channel. 100, 102
TM turning mirror. 42
TR terminating reservoir. 100
U under-passing. 91, 104, 107, 108
UV ultraviolet. 11, 13, 17, 30, 72, 92, 127
WP woodpile. 45, 71, 73, 75
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Chapter 1

Introduction

The development of lab-on-a-chip (LOC) devices is very much based on the idea of miniaturization of a complete laboratory on a chip-sized platform incorporating the capabilities of sample preparation, transport, reaction, and analysis. This idea was first proposed and demonstrated by Terry et al. [1] in 1979, in which a complete gas chromatography system was integrated on a 2” silicon wafer. This work failed to capture the interest of the scientific community at the time due to the lack of insightful discussion of its significance for chemical sensors or analysis systems in general. In 1990, almost twenty years later, Manz et al. [2] introduced the concept of micro-total-analysis-system (µ-TAS) which presented the basic theory of miniaturized chemical systems providing higher chromatographic and electrophoretic separation efficiencies together with significant reduction in reagent consumption. This stimulated the strong growth of LOC research in the academic community and the subsequent commercialization by industry.

The vision of placing a full-size laboratory onto a single chip-size device was pursued with the combination of microfluidics, electronics and photonics, promising a remarkable miniaturization without sacrificing performance or even improving efficiency. This integration vision has not been fully developed, but continues to evolve pushing biophotonic LOC forward. Depending on the application, various components including
micro-optics, Bragg grating filters, optical circuits, photonic crystals, electrodes, and micro-channel networks can be integrated onto a thumb-sized chip with multiple configurations. The micro-fabrication advances underlying this development were centered on two-dimensional (2-D) LOC devices, while new challenges requiring improved patterning flexibility together with integrating higher density microfluidic networks and functionalities are forcing a shift toward three-dimensional (3-D) fabrication.

Three-dimensional LOC fabrication has been explored with many materials of choice including glasses [3–6] and polymers [7–10]. Polymers are low in cost and micro-channel fabrication can be easily done by molding and embossing [11]. However, 3-D devices fabricated with polymers are usually 2-D structures layered together requiring highly precise alignment and numerous fabrication steps [7,8]. Polymers have their merits with the potential of mass production. However, polymers have poor thermal conductivity which can lead to Joule heating when driven by electrokinetic means. On the other hand, glasses having advantageous properties for LOC consist of chemical inertness, high optical transparency, low autofluorescence, high wettability and thermal conductivity that collectively support electro-osmotic flow. However, glasses are more challenging to process, and more expensive.

Fused silica glass has a well established micro-fabrication process based on photolithography and wet/dry etching for 2-D LOC. The recent advent of femtosecond laser irradiation with chemical etching (FLICE) enables 3-D fabrication in fused silica to sub-micron resolution, by utilizing the formation of nanogratings to facilitate chemical etching. This combined process provides an etching contrast ratio of up to 280:1 of laser modification tracks over the native glass with a 2.5% hydrofluoric acid (HF) solution [4,12–14]. Good progress has been made on fused silica LOC with microsystems integrated with optics and micro-mechanics [3], or waveguides integrated with commercial LOC [15] or laser-fabricated single-track micro-channels [4]. In developing highly functional 3-D microfluidics, practical LOC applications would benefit from a chemi-
cally inert fused silica platform with good electrokinetic properties and minimal external contamination from embedded microfluidic structures of variable shapes with smooth sidewalls and unrestricted channel length.

While FLICE allows the formation of high aspect ratio micro-channels, the embedded channel length is often limited to less than 4 mm [16] before tapering and uncontrollable cross-sectional shape evolve to limit the application broadly within a LOC direction [4, 14, 17]. Osellame et al. introduced a conical wobbling technique to create long straight and uniform cylindrical micro-channels, but this technique is not flexible enough for creating other cross-sectional shapes or non-straight channels [16]. Recently, buried micro-channels of nearly uniform cross-section were formed up to 9.2 mm length, but required a high concentration (10 M) of aqueous potassium hydroxide (KOH) solution and elevated temperature of 80 °C to yield only a moderate etch rate of ∼1.7 µm/min [18]. An improved method for micro-channel shape control together with unrestricted length would further advance LOC development with flexible shaping of microfluidic networks.

The FLICE process can be extended to the fabrication of periodic, dielectric structures to 3-D stimulated widespread study of photonic crystals (PCs) [19, 20] that promised dramatic miniaturization and integration of photonic devices onto LOCs. PCs have been formed by a range of 3-D nanofabrication processes, including colloidal self-assembly [21] and holographic lithography [22] for large area fabrication and direct laser writing (DLW) for creating more flexible PC designs in photoresists [23] and chalcogenide glass [24]. Chalcogenide is toxic, and photoresists are hydrophobic, autofluorescent, and less chemically stable while also shrinking during development. While an indirect multi-step method of coating and inverting a 3-D PC photoresist template has been demonstrated [25], using DLW with post selective chemical etching to generate PCs allows direct integration into micro-channels for highly functional LOCs.

The 3-D FLICE enables unprecedented 3-D capability and flexibility for the fabrication of glass LOCs. This method provides a major opportunity in dense packaging
while opening new 3-D design approaches for highly advanced microsystems. The aim of this thesis, thus, is the development and optimization of the FLICE technique to fabricate and monolithically integrate optical components, namely waveguides and PCs, into highly compact 3-D microfluidic structures with reservoirs and embedded micro-channels in bulk fused silica. The significance of the 3-D integration to define a dense biophotonic LOC is the possible opening up of a wide range of microsystems possibilities with multiple functionalities for simultaneous optical probing, manipulation, and analysis of the sample content, while providing a platform for sample preparation, transport, and reaction.

1.1 Thesis Motivations and Objectives

To advance the original vision of LOC that builds a laboratory onto a single chip-size device through integration of microfluidics, electronics, and photonics, a platform substrate with excellent thermal and chemical properties, to avoid Joule heating and unwanted reactions, together with a technique for flexible 3-D micro- and nano-fabrication of dense micro-structures in highly integrated microsystems, are required. Further, a flexible fabrication method providing ease of reconfiguration is needed to truly realize the advantages of glass as a substrate material. This motivation is pursued in this thesis work by focusing on the chemically inert fused silica glass and the FLICE technique to provide the new means to accomplish the following objectives:

1. Demonstrate that the FLICE process can be optimized for the fabrication of LOCs. In particular, FLICE fabrication and optimization of essential LOC microfluidic components in fused silica, including reservoirs for holding sample solutions and wastes, and surface (2-D) and embedded (3-D) micro-channels with unrestricted length, variable cross-section and smooth sidewalls for fluid transport and optical analysis of bio-materials inside micro-channels, for building the 3-D LOC platform.
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2. Direct femtosecond (fs)-laser writing of buried optical waveguides operating in the visible spectrum for later integration with micro-channels, and optical probing of bio-materials with possible applications in optical analysis.

3. FLICE fabrication of dense meshes of micro-capillaries to form uniform inverted-woodpile (IWP) structure for later integration with micro-channels for possible chromatographic application and optical analysis of bio-analytes.

4. Monolithic integration of various combinations of optical and microfluidic components to demonstrate the capability and flexibility of FLICE for 3-D rapid-prototyping of highly customizable biophotonic LOCs in fused silica, thus paving the way for highly functional LOCs.

5. Demonstration of 3-D LOCs with electrokinetic and capillary force functions for showcasing potential diagnostic applications.

The research presents the investigation of innovative 3-D microfluidic designs and micro-fabrication of 3-D micro-structures and optical components to push the forefront of the LOC community.

1.2 Chapter-by-Chapter Outline

This dissertation starts with a literature review in Chapter 2, followed by the microfabrication experimental set-up in Chapter 3, the studying and structuring of novel 3-D LOC components with femtosecond laser micro-fabrication in Chapter 4, and finally the demonstration of various 3-D integrated LOC devices in Chapter 5. The significance of
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this work and the future of biophotonic LOC devices are discussed in Chapters 6 and 7, respectively.

The following is a chapter-by-chapter summary:

Chapter 2 (Background — Biophotonic Lab-on-a-Chip) provides an overview of the past, present, and future of the LOC technology. It is the basis of this research, and comprises a comprehensive literature review. This chapter is divided into six sections: Section 2.1 describes LOC technology and its development, Section 2.2 introduces the use of two most popular types of materials used for LOC namely, polymers and glasses, Section 2.3 presents the current designs and applications of LOC and the forthcoming LOC technologies, Section 2.4 offers a brief introduction of electrokinetics and capillary forces in LOC fluid control, Section 2.5 introduces femtosecond laser-glass interaction providing 2-D and 3-D fabrication capabilities, and Section 2.6 describes the use of chemical etching to harness the femtosecond laser 3-D writing capability in LOC fabrication.

Chapter 3 (Experiment Set-up) presents the two main systems used for the 3-D laser-direct patterning of various LOC components. Section 3.1 provides an overview of the femtosecond laser system used to generate the laser pulses, and Section 3.2 describes the beam delivery system used to deliver the laser beam.

Chapter 4 (Lab-on-a-Chip Component Designs and Fabrication) is focused on the 3-D structuring and optimization of various LOC components, with Section 4.1 presenting the designs, fabrication, and results of LOC components including buried micro-channels, reservoir, woodpile structure, and Section 4.2 presents the fabrication and results of optical waveguide.
Chapter 1. Introduction

Chapter 5 (Biophotonic Integrated Devices) furthers the work presented in Chapter 4 to design and fabricate three biophotonic devices by integrating different combinations of LOC components. Section 5.1 presents the experimental procedures and fabrication parameters. The results are presented in Section 5.2 for the waveguide probing device, Section 5.3 for the particle counting device, and Section 5.4 for the multi-level capillary electrophoresis device.

Chapter 6 (Significance of this Work) reflects on the significance of the present work. Section 6.1 discusses the significance of the fabricated microfluidic components, Section 6.2 examines the significance of photonic crystals fabrication in bulk glass, and Section 6.3 discusses the importance of this work in providing the means for rapid prototyping of 3-D LOC platforms. Section 6.4.1 presents the preliminary work in progress for a microphotonic chromatographic LOC device.

Chapter 7 (Conclusions) summarizes the present work, and further discusses the future work and the impact in the fabrication and application of LOCs.
Chapter 2

Background—Biophotonic Lab-on-a-Chip

The field of photonics has long been established in dealing broadly with the optical interactions of materials and the applications of light. Photonics is the science and technology of generation, manipulation and detection of photons that has played a central role in the field of information technology. By trading electrons with photons, the capacity and the speed of data transmission, processing and storage have improved dramatically, while extension to biology, medicine, and imaging has led to the development of biophotonics.

Biophotonics denotes a combined field of biology and photonics that encompasses all the techniques involving the interaction of photons and biological materials. State-of-the-art photonic technologies are not only used to modify biological materials, they are also employed to aid the analysis of biomaterials through photon absorption, transmission, reflection, and detection. The rapid growth of biophotonics is driven by the development of new photonic technologies. With the advent of fs-laser technologies, the high light intensity and ultra-short pulse duration provide new approaches and directions in laser-material interaction, which promise to further drive the growth of biotechnology.

One niche of biotechnology envisions the replacement of a full size laboratory into
a chip-size device, namely LOC. Current fabrication techniques are borrowed from the semiconductor industry where photolithography and wet etching provide the basic structure of LOC. However, new fabrication approaches are desired to further enable the integration of compact photonic devices for the biophotonic LOC. Laser micromachining is the alternate approach for direct micro-fabrication that readily provides submicron features in a wide variety of materials in 2-D planar structure. FLICE is a new approach to fabricate 3-D LOC with the potential of incorporating photonic capabilities including optical circuits and PCs with microfluidic structures.

In this chapter, a review of the LOC technology in the two most popular class of materials, namely polymers and glasses, is presented with the emphasis on glass materials. The basic fluidic control methods that form the basic understanding of microprocessing and operation of LOCs are described. Lastly, fs-laser processing of glass, and the FLICE technique for forming surface 2-D and embedded 3-D micro-structures are presented to provide an overview of the current state-of-the-art laser fabrication technology for the biophotonic LOC application direction.

2.1 What is Lab-on-a-Chip Technology?

The original concept of LOC dates back to 33 years ago. Terry et al. fabricated and demonstrated the first chromatographic analyzer on a 2” silicon wafer, which was able to separate a mixture in mere seconds [1]. This novel concept of miniaturization went unnoticed, and there was no immediate response from the scientific community largely because this work was mainly focused on the device fabrication rather than providing insights into the potential benefits of miniaturized chemical sensing and analysis devices. The effort continued to mainly direct toward the fabrication of silicon micropumps [26–29] and microvalves [28–31], whereas diagnostic and analysis components were a minor part of the objectives that neglected the higher impact advantages.
A major advance was driven by Manz et al. in 1990, who demonstrated a novel miniature open-tubular liquid chromatograph on a 5 mm × 5 mm silicon chip [32]. More significantly, they proposed the concept of µ-TAS which presented the basic theory of miniaturized chemical systems encompassing sample pretreatment, separation, and detection on a single chip that enhances LOC performance together with the benefits of reduced reagent consumption [2]. This vision of integrating numerous components and functionalities within a single miniature platform without sacrificing performance, and allowing reduction in time and cost with high portability stimulated the scientific community to fuel a tremendous research effort.

LOC revolutionized the design of laboratories by integrating capabilities from sample preparation to diagnostics and analysis in a miniature platform offering enhanced measurement performances by manipulating minute amounts of reagents (order of 10⁻⁹ litres or less) in channels with micron-sized dimensions. The LOC concept has been applied in biology, including DNA separation and analysis [33–37], detection of biological molecules such as virus [38, 39], and cell separation and sorting [40–42]. Integration of photonic components including optical waveguides [3, 4, 15, 17, 36, 43–52], Bragg gratings [53, 54], and PCs [50, 55, 56] promise to greatly improve the diagnostic capabilities of LOC. Hence, advances in photonics promise new means to enhance the functionality of LOC particularly in the biophotonic LOC direction. Furthermore, the versatility and portability of LOC offer advantages in point-of-care ability in the medicinal field, environmental monitoring and other industrial applications.

2.2 Lab-on-a-Chip Materials and Fabrication Techniques

The initial fabrication technologies of LOC were borrowed from the semiconductor industry following from the success of photolithography in silicon. Most of the early LOC
work was dominated by silicon as the substrate material. But due to the requirement for chemical compatibility and optical transparency, research development shifted towards the use of glasses and plastics. Further, silicon is an expensive material that is opaque in the visible and ultraviolet (UV) spectrum, thereby limiting silicon application from many on-chip optical detection and analysis applications. On the other hand, polymers and glasses, although imperfect in their own ways, offer several advantages over silicon as a LOC platform. Polymers have gained much support because of their ease of fabrication, low cost (for mass production), permeability to gases, and elasticity. Glasses are more difficult to process, but have advantages of high optical transparency, chemical inertness, thermal conductivity and wettability over polymers. Novel fabrication technologies are being developed to take advantage of the benefits of glass materials for the LOC platform.

2.2.1 Polymers

Polymers have recently gained popularity as a substrate material in the field of LOC. Polymers are low cost and are generally compatible with biological or chemical applications. Microfluidic networks with channels of micrometer size can be fabricated by soft-lithograph of elastomers such as polydimethylsiloxane (PDMS). The soft-lithograph technique is based on printing, molding, and embossing with an elastomeric stamp [11,57]. Soft-lithography is a simple technique that is cost-effective for mass production because the mold can be reused numerous times. Imprinting is another replication technique that is able to fabricate complex micro-channel arrays by imprinting plastic substrates such as poly(methyl methacrylate) (PMMA) with an inverse 3-D image of the device micro-machined on a silicon wafer [58]. Although this method is cost effective, the multi-step process is not suitable for rapid prototyping.

For rapid prototyping of custom designed LOC, conventional soft-lithography may not be the most suitable technique. A low-power laser (10 mW) operating at 532 nm has been applied to ablate a microfluidic pattern on PMMA which was then transferred onto
a PDMS master stamp for microcontact printing (µCP) and controlled sagging microcontact printing (CSµCP) [59]. Although this is a step toward a more rapid prototyping technique, µCP involves multi-step processing that requires first creating a master stamp. As part of the search for rapid prototyping techniques, flexible 3-D fabrication is becoming attractive due to the higher complexity of microfluidic networks and functionalities required on a LOC. Three-dimensional LOCs fabricated with polymers are usually 2-D structures layered together requiring highly precise alignment and numerous fabrication steps based mainly on lithography [7, 8, 60]. Three-dimensional 3-D PDMS microfluidic structure have been formed by stacking several micromold patterned PDMS layers [60]. Alternatively, photoresist (SU-8) has been used to create multi-layer structure based on photolithography and successive adhesive bonding and releasing steps [7]. A combination of PDMS and SU-8 requiring multi-steps of lithography and elastomer curing [8] have also been applied towards demonstration of 3-D polymer based LOC.

With the availability of commercial stereolithography (SL) systems, embedded 3-D micro-channels have been demonstrated by line-scan SL [9]. However, SL has not been considered as a reliable method for the fabrication of horizontal embedded micro-channels due to the penetration of the laser beam to cause crosslinking within the channel when building the down-facing surface. A more cumbersome SL approach therefore requires a multi-step process involving the insertion and removal of micro-wires to keep the buried channels open [10].

Many research efforts are still largely based on lithography to benefit from the low-cost mass production standard of industry. This benefit, together with the optical transparency and gas permeability of elastomers such as PDMS have largely been the determining factors for choosing polymers rather than glasses for LOC. However, for applications not requiring permeability to gases, which eliminate most work with living mammalian cells, polymers have some drawbacks. Polymers have poor thermal conductivity which can lead to Joule heating when driven by electrokinetic means, are often
difficult to clean, are not reusable due to their porous nature, and are incompatible with several organic solvents, particularly with nonpolar solvents. To this end, application specific materials are selected based on their desirable properties.

### 2.2.2 Glasses

Glasses are more difficult to process compared with polymers, but have advantageous properties of chemical inertness to most solvents, optically transparency for detection and analysis in biological applications, high wettability without surface treatments, low autofluorescence, high thermal conductivity suitable for electrokinetics, and surface chemistry appropriate for electroosmotic flow, making glasses highly preferable for LOC applications. The main research has been focused on amorphous fused silica glass which has the additional advantages of high purity and the ability to be doped with other materials such as germanium and boron. Moreover, silica has been used extensively in the opto-electronic industries \[61\] because of high transmission from the UV to infrared (IR) and low optical nonlinearity and dispersion. These favourable physical and optical properties combine to define a suitable substrate for fabrication of optical circuits together with biophotonics-enabled LOC structures.

The earliest glass LOC processing was based on photolithography borrowed from the well established semiconductor industry. In general, there are three types of lithography divided into optical lithography, x-ray lithography, and electron beam lithography. Photolithography is a multi-step technique involving the pattern transfer from a mask onto a layer of photoresist followed by wet-chemistry or dry etching \[62, 63\]. Upon the photoresist exposure, the resist either becomes insoluble (negative resist) or more soluble (positive resist) due to photochemical induced changes. With the current photolithography technology, submicron features (100 nm) can be easily generated in a cleanroom environment. Although such high resolution is an advantage, the time consuming multi-step process is not suitable for efficient prototyping purposes. Nevertheless, standard
photolithography has been widely applied to fabricate glass LOCs for electrophoresis separation [64–66] and micro-analytical applications [67].

Direct-laser photoablation, made available with the advancement of laser technologies, uses high energy photons to remove irradiated materials from a substrate. This technique defines a direct and flexible rapid prototyping method for creating desired patterns of microfluidic structures on the surface of the substrate. The F_2 laser, with a wavelength of 157 nm, provides high energy photon (7.9 eV) for driving strong interactions in glasses and has been used to demonstrate the micro-structuring of smooth and crack-free surface microfluidics [68]. Other lasers have also been used to fabricate surface micro-channels including the use of a frequency-doubled copper vapour laser with a wavelength of 255 nm and fs laser with a wavelength of 800 nm [69] and 1660 nm [70]. However, the technique of photoablation yields planar LOC structures and is not easily extendable to the fabrication of 3-D structures.

Extending the microfluidic chip layout to make full use of all three spatial dimensions is a significant objective to greatly improve the flexibility of glass LOC devices and the ability to integrate optical waveguides and other sensors. Three-dimensional microfluidic devices have become feasible with the development of fs-laser micromachining. The unique feature of fs-laser micromachining, that is based on photoablation, is caused by multiphoton absorption, is that the ablation is tightly confined to the focal volume. Regions surrounding the focal volume are almost completely transparent to the incident radiation and remain unmodified by the laser beam. This effect has been exploited to fabricate surface microfluidic channels and a subsurface tunnel connecting two surface chambers in a fused silica microscope slide [70]. The roughness of the channel sidewalls is readily visible under the optical microscope, suggesting a surface roughness of the order of a few microns. Further, the subsurface tunnel is limited to a length to ~50 µm with significant tapering from 40 µm to 15 µm from end-to-end. Micro holes [71] and channels [72] have been fabricated in silica glass with the liquid-assisted fs laser technique.
Nanofluidic channels in a glass coverslip have also been reported using the liquid-assisted technique \cite{73}, but the writing process is extremely slow, limited in length of hundreds of microns, and required ultrasonic agitation for this extended length. The ultrasonic agitation facilitates the flow of liquid to reduce the redeposition of debris, but the ultrasonic wave is detrimental to delicate structures. The use of fs-laser micromachining in the FLICE technique is an alternative approach, where the glass substrate is first laser modified within a small focal volume such that the modified fused silica became susceptible to a much higher etch rate in HF than the surrounding, unexposed material \cite{4,12-14}. Embedded micro-channels of lengths up to 4 mm have been demonstrated with HF etching \cite{16} and up to 9.2 mm with high concentration (10 M) of aqueous KOH at 80 °C \cite{18}. Custom designed photosensitive Foturan glass has also been used to produce 3-D microfluidic devices such as a nanoaquarium for observing microorganisms and LOC devices with photonic biosensing \cite{74}. But the resolution here is limited to a few microns due to the crystalline phase of lithium metasilicate formed as well as due to a low chemical etching contrast ratio of 43:1 compared with unmodified glass in 10% HF solution \cite{5,6}.

The benefits of the FLICE technique now available for 3-D fabrication of glasses make such materials more attractive for LOC. This allows us to take advantage of the glasses’ material stability, reliable hydrophilic surface without special surface coating, optical transparency, thermal conductivity, and re-usability that are crucial for many application directions.

### 2.3 The Present and the Future of Biophotonic Lab-on-a-Chip

Photonic technologies are finding biological applications in areas such as optical manipulation, diagnostics, analysis, and therapeutic treatments. The purposeful fusion of
photonics and biology emerges as biophotonics. This multi-discipline field not only draws expertise from engineers, biologists and chemists, it promises to improve the quality of life through increasing productivity — such is the aim of biophotonic LOC.

Biophotonic LOC provides a powerful tool that offers enhanced diagnostic sensitivity through integration of photonic functions with a minute sample volume. LOC seeks the miniaturization of large instruments to improve portability and analysis throughput of bio-samples to serve in wide spread laboratory purposes with eventual self administered use in home. Many early researches have been focused on electrophoretic and electrophoretic LOC applications that were combined with an external optical system for detection and analysis of various analytes by fluorescence excitation of deoxyribonucleic acid (DNA), protein and living cells.

The early development of biophotonic LOC employed a practical solution of conventional optical tools for detection and analysis. However, such external tools require precise alignment and mechanical stability of free-spaced optical systems that are not readily miniaturized for compact packaging. With this in mind, there is an increased research effort in micro-fabrication of LOC with micro-scale photonic components. This type of biophotonic LOC has the potential to revolutionize the way laboratories are designed, with less space and time required to perform the same analytical work.

Much progress has been made in the past decade in integration of photonic capabilities on LOC. With reactive ion etching and anodic bonding for sealing, a lithographically patterned chip has been integrated with planar waveguides fabricated with flame hydrolysis deposition (FHD) that offered a minimum measurable concentration of 20 pM of Cy5 fluorescent dye while further demonstrating its applicability as a biophotonic LOC for analytical measurements of fluorescent-labelled oligodeoxynucleotides. One of the advantages of FHD was its ability to scale to large substrate sizes. Unlike plasma-enhanced chemical vapour deposition (PECVD) there is no need for a vacuum system and it is possible to deposit on any size of substrate. Although this demonstration was
encouraging, the complex multi-step and multi-technology based fabrication was difficult to scale for rapid prototyping production.

For these reasons, researchers have been looking for simpler ways for photonic integration with microfluidic devices. Friis et al. presented an unique layout for monolithic integration of waveguides and micro-channels which prevented leakage and offered hermetical sealing of the channels. The device was used to demonstrate optical absorption spectroscopy and flow cytometry with fluorescent polymer beads [44]. A simpler design approach demonstrated by Cui et al. was based on the integration of pairs of optical fibers by fixing the fibers in grooves inside the micro-channel walls, thereby providing single particle detection with light scattering and fluorescence emission, as well as particle counting and flow velocity measurements [78]. These techniques were promising at the time. However, fabrication methods involving multi-step and multi-tool techniques together with the limitation of planar 2-D structure made this direction obsolete.

The next wave of biophotonic LOCs involved various methods of solid or liquid optical circuit integration on a polymer platform, which is a key component for chip-based controlling of light for detection and/or analysis. Polymers have been the material of choice since the initial development of LOC because of their ease of processing and low-cost for mass production. From the solid waveguides fabricated in SU-8 microfluidic device for fluorescence detection using photolithography [45,46], to the concurrent development of the soft-lithography technique for fabricating UV-laser-written optical adhesive [50] large core waveguides and PDMS based solid [79] and liquid-core waveguides [36] for fluorescence filtering and detection, these polymeric biophotonic LOCs have been gaining popularity. The benefit of a liquid core waveguide is the ability to adjust the waveguide properties by simply changing the liquid within. However, such waveguides rely on the stability of the continuous liquid flow [49,80]. Recently, Fei et al. demonstrated a simple and robust method to monolithically integrate liquid core waveguides with PDMS chip by utilizing a dead-ended channel for cytometry [52], thereby eliminating the need for
the continuous flow of liquid.

Optical component integration in biophotonic LOC goes beyond optical waveguides or circuits for guiding electromagnetic (EM) waves in the optical spectrum. In recent years, the widespread 3-D development of PCs [19,20], periodic dielectric nanostructures that affect the propagation of photons creating photonic bandgaps, promised dramatic miniaturization of photonic devices offering possible integration into LOC. A range of 3-D micro- and nano-fabrication techniques have been developed to form large area PCs including colloidal self-assembly of silica microspheres [81], self-organized nano-void array [82] formed by laser-induced microexplosion, and holographic lithography [22] using a multi-level diffractive optical element (DOE). These methods are not sufficiently flexible for generating more complex patterns beyond simple periodic structure. DLW of PCs, which is a highly flexible writing method, has been demonstrated in photoresists [23] and chalcogenide glasses [24]. Chalcogenide glasses are toxic, and photoresists are hydrophobic, autofluorescent and shrink on development, which may not be suitable for direct LOC integration. With silica being the superior LOC material, silica chemical vapour deposition has been used to coat over photoresist template to form a silica inverted PC template [25]. However, this method requires multi-step processing to remove the polymer template, which can limit the integrability of this type of PC in LOC. Clearly, a method with direct and highly integrable fabrication of glass PC into microfluidic LOC devices is desired.

DLW technique for optical component integration has its own merits for rapid prototyping glass materials with the advantages as were previously discussed for LOC fabrication in Section 2.2.2 above. By exploiting the high 7.9-eV photon energy of the F2 laser that drives strong absorption in fused silica glass for ablation of smooth micro-channel walls and strong photosensitive response for optical waveguide fabrication, an integrated biophotonic sensor was demonstrated [48]. This 157-nm laser writing method provided a rapid prototyping method for biophotonic LOC but once again confined in the realm
of forming only planar 2-D structures.

In the last decade, fs-laser technology has become a popular tool for biological applications not only in diagnostics and analysis, but also in biophotonic LOC fabrication. This laser technology provides a means to produce 3-D microfluidic devices in glasses as discussed above in Section 2.2.2 and its unique processing qualities offer direct-writing of 3-D optical circuits [83]. Taking advantage of the offerings of fs laser, micro-channels fabricated with the FLICE technique can be made to intersect with optical waveguides on a single fused silica substrate [4]. Photosensitive Foturan glass modified by the fs laser have been used to produce biophotonic LOC with hollow micro-channels and micro-optics followed by direct-writing of optical waveguide for fluorescence analysis and absorption measurement [84]. These are only a small sample of the significant research activity into what the future holds for creating highly functional biophotonic LOCs that extend into the third dimension.

There are several main driving forces behind the ever-advancing LOC technology including improvement in molecular analysis in molecular biology to achieve higher sensitivity and throughput from device miniaturization, biodefence requirements for detecting chemical and biological threats with field-deployable portable systems, and the further hope for transferring the success of high volume microelectronic fabrication techniques to the LOC [85]. Amongst these varying driving forces are the common need for high versatility and increased integrative capacity in LOC. These continual challenges of incorporating an increasing number of components for added functionalities in even more compact devices dictate the future direction for commercialization success of biophotonic LOCs.

At one end of the spectrum, low-cost large-scale production is the current main stream thrust for biophotonic LOC development. But this direction impedes the flexible and rapid adoption of new applications requiring insertion of additional microfluidic or photonic components. At the other end of the spectrum is the need of rapid prototyping for
custom devices or devices required in the research phase where short turnaround time for facile component integration over many prototyping iterations are necessary. With the ever-changing world of biomedical research, the future in LOC production may evolve towards the combinations of multi-step mass-produced devices where add-on functions in rapid prototyping steps will use other more flexible fabrication techniques. For high reconfigurability in the case of today’s commercial fused silica LOC production, standard wet-lab technique may be combined with fs-laser post-processing to insert optical waveguides for fluorescence excitation in pre-existing micro-channels [15]. The future of biophotonics is looking brighter with the advent in new LOC fabrication technologies, and the continual advancements will further facilitate the growth of this field.

2.4 Fluid Control

The microfluidic channel is the main vessel for fluidic transport in most LOC designs. It is important to understand the basics of fluid control in these devices. In the well-developed microfluidic field, micro-channels of various size and shape have been explored for harnessing mass transport in LOCs. There are two main driving mechanisms of continuous on-chip fluidic transport which have been studied extensively, namely, capillary forces and electrokinetics. The following sections provide the basic theories and understanding behind each of the driving force of fluidic flow through micro-channels.

2.4.1 Capillary Forces

Capillary forces are found throughout nature. As an example, plants transport water up from their roots against gravity by means of capillary action in which the binding forces between water molecules (cohesion and water tension) and the adhesion forces are greater than the gravitational force. These same forces are responsible for driving autonomous capillary system (CS) in which capillary effects dominate in microfluidics which have a
large surface-to-volume ratio. Capillarity or capillary action is the phenomenon, which encompasses adhesive and cohesive forces and surface tension, where liquid wetting of the interior of capillaries serves to minimize the surface area of a given volume.

Surface tension, often called interfacial tension, is the force along a line of unit length parallel to the surface but acting perpendicular to the line, and is represented by $\gamma$. It is a property of the surface of a liquid which resist an external force and can be seen as behaving like a membrane at the interface between a liquid and a vapour under isotropic tension. This is the result of the cohesion force of the liquid molecules. The lack of liquid molecules at the surface in the direction of the gas phase creates an unbalanced force resulting in the tendency of the surface molecules being pulled back to the bulk. This leads to the reduction of the surface area as much as possible under given boundary conditions and the constant enclosed volume.

For a droplet of water at equilibrium as depicted in Figure 2.1, the forces and tensions at the boundaries of liquid (L), solid (S) and gas (G) are balanced according to the Young-Dupré equation,

$$\gamma_{SG} - \gamma_{SL} = \gamma_{LG}\cos\theta,$$

(2.1)

where $\theta$ is the equilibrium contact angle measured in the liquid phase between the solid surface and the tangent to the liquid surface. The contact angle depends on the surface tensions. The contact angle $\theta$ is between $0^\circ$ and $90^\circ$ if $\gamma_{SG}$ is greater than $\gamma_{SL}$, and $\theta$ is between $90^\circ$ and $180^\circ$ if $\gamma_{SG}$ is less than $\gamma_{SL}$. The wettability increases when the contact angle decreases, and $\theta = 0^\circ$ represents the limit of complete or perfect wetting. A hydrophilic surface provides a low contact angle, whereas a hydrophobic surface has a large contact angle.

How does a liquid get drawn in a capillary? “Wicking” is used to describe the spontaneous transport of a liquid through a micro-channel by capillary forces which are caused by wetting. This implies that wetting is a prerequisite for wicking. For the spontaneous
displacement of the solid-gas interface with the solid-liquid interface, the work of penetration has to be positive, meaning that free energy has to be gained. This process is achieved when the solid-gas interfacial energy, \( \gamma_{SG} \), is greater than the solid-liquid interfacial energy, \( \gamma_{SL} \), as described by the equation:

\[
W_p = \gamma_{SG} - \gamma_{SL}
\]  

For capillary flow, the work of penetration \( (W_p) \), or \( \gamma_{SG} - \gamma_{SL} \), must be positive, which further requires the quantity \( \gamma_{LG}\cos\theta \) from the Young-Dupré Equation 2.1 to also be positive. This suggests that \( \cos\theta \) must be positive, yielding a contact angle \( \theta \) to be \(<90^\circ\). Therefore, spontaneous transport of liquid can be easily generated in a micro-channel with hydrophilic walls.

A common approach in characterizing capillary action is to relate capillary penetration to capillary pressure. The meniscus formed between the liquid-gas interface, due to the wetting of the liquid on the capillary walls, creates a pressure difference \( (\Delta P) \) across the liquid-gas interface called the Laplace pressure or the capillary pressure, defined by
the Laplace equation:

\[ \Delta P = \gamma_{LG} \left( \frac{1}{R_1} + \frac{1}{R_2} \right). \]  

(2.3)

Here \( R_1 \) and \( R_2 \) are the radii of the curved interfaces. Because many micro-channels in the literature have a circular cross section, the radii of the curvature \( R_1 \) and \( R_2 \) will be identical causing the Laplace equation to be simplified as follow:

\[ \Delta P = \frac{2\gamma_{LG}}{R}. \]  

(2.4)

For a circular micro-channel with a radius \( r \), the radius can be related with \( R \) according to

\[ \frac{r}{R} = \cos \theta, \]  

(2.5)

which when substituted into Equation 2.4 yields the following Laplace pressure equation:

\[ \Delta P = \frac{2\gamma_{LG}\cos \theta}{r}. \]  

(2.6)

In addition to circular cross-section micro-channels, rectangular cross-section channels have gained popularity in the field of LOC. To derive the equation governing the pressure difference in a rectangular channel, the hydraulic radius needs to be defined first,

\[ r_{hyd} = \frac{2A}{P}, \]  

(2.7)

where \( A \) and \( P \) are the cross-sectional area and perimeter, respectively. The hydraulic radius for a circular channel is

\[ r_{hyd} = r, \]  

(2.8)
and for a rectangular channel is

$$r_{hyd} = \frac{1}{\frac{1}{w} + \frac{1}{h}},$$

(2.9)

where $w$ and $h$ are the width and height of the rectangular cross section as shown in Figure 2.2 with a cartesian coordinate system. Therefore, the capillary pressure for a rectangular channel is given by:

$$\Delta P = 2\gamma_{LG}\cos\theta \left( \frac{1}{w} + \frac{1}{h} \right).$$

(2.10)

The basic understanding of capillary forces for spontaneous liquid transport in micro-channels is essential for building autonomous capillary systems. This section presents a brief overview of capillarity fundamentals, and is not intended to fully account for the various wetting and wicking processes. Further details can be obtain in standard textbooks discussing capillarity and wetting phenomena [86, 87].
2.4.2 Electrokinetics

The LOC technology has been growing rapidly, where liquids remain as the most important media of transport for enabling biomedical analysis and diagnostics. In general, LOCs consist of a network of micro-channels integrated with reservoirs, electrodes and other functionalities depending on the application. As more complex devices are built, multi-step manipulation of reagents and analytes become more difficult under autonomous flow, and automation through external controlled signals is required. Such signals are generally provided in the form of applied electric field through the integrated electrodes, enabling controlled liquid transport within the micro-channels. This mode of liquid transport, called electrokinetics, offers precise manipulation of microfluidic processes and enhances the performance of the LOC. The objective of this section is to provide a basic understanding of electrokinetics. For a more detailed conceptual understanding of electrokinetic phenomena, there are many textbooks, such as the book by Dongqing Li.

Electrokinetic phenomena are driven by the existence of a net charge at the liquid-substrate interface of the micro-channels, affecting the oppositely-charged ion (or counter ion) distribution in the surrounding interfacial region close to the surface, when under the influence of an applied external electric field. For silicate glasses, the surface will form an anionic charge resulting from the ionization of the surface silanol (Si–OH) group to negatively charged silanoate (Si–O\(^-\)) group after the passing of a basic solution through the micro-channel. The counter ion concentration, which are positively charged for silicate surfaces, increases close to the surface creating an electric double layer (EDL) charge distribution adjacent to channel walls with an electric potential called the zeta potential (\(\zeta\)). The EDL can be separated into two main layers: the inner layer, called the stern layer, having ions firmly bound, and an outer layer, called the diffuse layer, with ions less strongly attached.

There are four distinct effects, collectively called electrokinetic effects, when an elec-
Electrophoretic Flow

Electroosmotic Flow

Figure 2.3: Schematic showing the principle of capillary electrophoresis involving a combination of two principal factors called electroosmotic flow and electrophoretic mobility under an electric field applied across the micro-channel.

The electric field is applied through such channel with electrical charged surfaces: electroosmosis, electrophoresis, streaming potential, and sedimentation potential. Electroosmosis or electroosmotic flow (EOF) is the bulk flow of liquid in a micro-channel resulting from the surface charge on the interior walls of the channel, whereas electrophoresis is the movement of a charged particle relative to the bulk liquid under an applied electric field. Figure 2.3 illustrates these two concepts schematically. Streaming potential and sedimentation potential are the electric fields generated when a liquid flows past a stationary charged surface and when charged particles move relative to a stationary liquid, respectively.

EOF is the liquid flow caused by the migration of the hydrated counter ions in the diffuse layer, under an applied electric field, which in turn drags the electrically neutral bulk liquid \[89\]. The diffuse ions in the EDL induces motion of the liquid due to the electric body force,

\[
f_E = \rho_E E. \tag{2.11}
\]

Here \(\rho_E\) is the electric charge density and \(E\) is the electric field. The liquid motion due to the electric body force in the EDL provides a viscous drag force on the bulk liquid.
In micro-channels, this EOF can be described by the Navier-Stokes equation (equation 2.12), which provides the velocity field derived from the conservation of momentum in a continuous fluid,

\[
\rho \left( \frac{\partial \mathbf{v}}{\partial t} + \mathbf{v} \cdot \nabla \mathbf{v} \right) = -\nabla p + \mathbf{f}_E + \mu \nabla^2 \mathbf{v},
\]

(2.12)
together with the Poisson’s equation describing the charge distribution relating to the spatial variation of the electric field,

\[
\nabla^2 \psi = -\frac{\rho E}{\epsilon},
\]

(2.13)

where \( \rho \) is the fluid density, \( \mathbf{v} \) is the fluid velocity, \( p \) is the pressure, \( \mu \) is the viscosity, \( \psi \) is the electrostatic potential, and \( \epsilon \) is the permittivity of the liquid given by

\[
\epsilon = \epsilon_r \epsilon_0,
\]

(2.14)

where \( \epsilon_r \) is the relative permittivity and \( \epsilon_0 \) is the vacuum permittivity. The Poisson’s equation can be simplified using the Debye-Hückel approximation, which combines Poisson’s equation and Maxwell-Boltzmann distribution followed by truncating this combined Poisson-Boltzmann equation through Taylor series, to obtain the following:

\[
\nabla^2 \psi = \frac{\psi}{\lambda_D^2},
\]

(2.15)

where \( \lambda_D \) is the Debye length which is the characteristic thickness of the EDL and is given by

\[
\lambda_D = \left( \frac{\epsilon R T}{F^2 \sum_i z_{v,i}^2 c_{0,i}} \right)^{\frac{1}{2}}
\]

(2.16)

where \( R \) is the gas constant, \( T \) is the temperature, \( F \) is the Faraday’s constant, \( z_{v,i} \) is the ion valence of the \( i \)th species, and \( c_{0,i} \) is the bulk concentration of the \( i \)th ion species.
For the case of uniform zeta-potential (i.e. uniform electric charge on channel walls) with no pressure (i.e. $p = 0$), the steady state EOF velocity is found to be

$$v_{\text{eof}}(y) = -\frac{\epsilon\zeta E}{\mu} \left(1 - \frac{\psi(y)}{\zeta}\right).$$

(2.17)

The EOF velocity is proportional to the magnitude of the electric field. For the characteristic length of the micro-channel, $L$, to be much bigger than the Debye length (typically $<10$ nm in micro-channels), the flow field can be separated into the near-wall region and bulk region where the flow is dominated by viscous and electrostatic forces, and inertial and pressure forces, respectively [90]. The bulk flow, which is the EOF outside the EDL, separated by a slip velocity condition determined by the Helmholtz-Smoulochowski equation is

$$v_{\text{eof}} = -\frac{\epsilon\zeta E}{\mu}.$$

(2.18)

For a rectangular micro-channel with a cross-section of width $w$ and height $h$ as shown in Figure 2.2, the velocity profile with uniform zeta-potential for a transient EOF can be solved by Navier-Stokes equation (equation 2.12) under the slip velocity condition as [91]

$$v(x, y, t) = -\frac{\epsilon\zeta E}{\mu} + \frac{16\epsilon\zeta E}{\mu\pi^2} \sum_{m=1}^{\infty} \sum_{n=1}^{\infty} \frac{(-1)^{m+n} \cos \left[\frac{(2m-1)\pi x}{w}\right] \cos \left[\frac{(2n-1)\pi y}{h}\right]}{(2m-1)(2n-1)}$$

$$\times \exp \left[-D_h^2 \left(\left(\frac{2m-1}{w}\right)^2 + \left(\frac{2n-1}{h}\right)^2\right) \frac{tv}{D_h^2}\right]$$

(2.19)

where $\nu$ is the kinematic viscosity, and $D_h$ is the hydraulic diameter which is defined as follows:

$$D_h = 4r_{\text{hyd}}.$$

(2.20)

For such flow under the slip velocity condition, EOF can be considered as Couette flow which is a laminar flow of a viscous liquid between two surfaces that have tangential relative motion.
The other electrokinetic effect of interest is electrophoresis, which is the electromigration of charged molecules and particles suspended in the electrolyte toward the electrode of the opposite charge under an applied electric field. The electrophoretic velocity, \( v_{ep} \), then is the equilibrium velocity reached between the attractive and the viscous forces acting on the molecule or particle, and is proportional to the electric field as given by

\[
v_{ep} = \mu_{ep} E
\]  

(2.21)

where \( \mu_{ep} \) is the electrophoretic mobility.

For a particle with radius, \( r_{par} \), much smaller than the Debye length, \( r_{par} \ll \lambda_D \), which is the case for most ions and molecules, the electrophoretic velocity can be derived through Stoke’s law and expressed as

\[
v_{ep} = \frac{q}{6\pi\mu r_{par}} E
\]

(2.22)

where \( q \) is the particle charge. However, for particle radius larger than the Debye length, \( r_{par} > \lambda_D \), the electrophoretic velocity of such particle is expressed as

\[
v_{ep} = \frac{2\epsilon\zeta f(K_a)}{3\mu} E
\]

(2.23)

where \( f(K_a) \) is Henry’s function that depends on the ratio of the particle radius to the Debye length \( \left( \frac{r_{par}}{\lambda_D} \right) \). There are two values, 1.5 or 1.0, generally used for \( f(K_a) \) as approximations. For large particles in polar media, i.e. particles having radius larger than \( \sim 0.2 \ \mu m \) dispersed in electrolytes with more than a concentration of \( \sim 10^{-3} \ M \), \( f(K_a) \) is approximated to the maximum value of 1.5. This is referred to as the Smoluchowski approximation, and equation 2.23 becomes the Helmholtz-Smoluchowski equation (equation 2.18). For particles in non-polar media, \( f(K_a) \) is approximated to the minimum value of 1, and this is referred to as the Hückel approximation.
In a micro-channel under an applied electric field, the electrophoretic flow of the suspended particles is superimposed on the EOF of the liquid. This combined particle velocity is the superposition of the electroosmotic and electrophoretic velocities giving

\[ v_{\text{par}} = v_{\text{eof}} + v_{\text{ep}}. \] (2.24)

The schematic illustration of such flow is shown in Figure 2.3.

2.5 Femtosecond Laser Processing in Glass

Laser, an acronym for Light Amplification by Stimulated Emission of Radiation, is a term first published by Gordon Could in 1959 [92]. In 1960, Maiman demonstrated the first functioning ruby laser [93], from which the laser has emerged as a new energy source for processing of materials. With this novel source, a collimated beam of light can be focused to a small spot size producing a high power density to drive efficient and novel material interactions.

Direct laser processing of materials has become an important tool in many industrial fields, and remains an exciting research field in which new application directions and new laser systems are being developed. Traditional machining methods involve direct contact with the surface which can cause damage or contamination on the substrate surface. Various laser systems are typically tailor designed for specific applications. The advantages of laser processing that drive its increasing importance can be generalize as follows: non-contact micro- and/or nano-machining with minimal contamination and machine wear; mass production capability or high flexibility for custom fabrication and rapid prototyping; machining hard-to-process materials including glasses; and ability to internally modify the material properties.

In 1987, Küper and Stuke first demonstrated the use of a 300-fs UV excimer laser at 248-nm to ablate PMMA with improved surface morphology [94]. This had since
led to processing of various materials using fs-lasers including metals, semiconductors, dielectrics, polymers, and biological cells and tissues. Femtosecond lasers offer several advantages due to the high intensity in the range of $\sim 10^{13}$ W/cm$^2$ driving highly nonlinear absorption processes in materials that do not normally absorb at the laser wavelength and ultrafast pulses (ps/fs) depositing energy on a timescale that is short compared to atomic relaxation processes. These benefits are particularly attractive for LOC fabrication and processing, including their versatile ability to fabricate micro- and nano-structures inside optically transparent materials through non-linear absorption, and low collateral damage due to short pulses.

### 2.5.1 Laser-Glass Interaction

Glass is difficult to process simply due to its high optical transparency to the laser radiation. However, glass has material properties ideally suited for fabrication of biophotonic LOC devices due to its inert nature to most chemicals, high thermal conductivity and transmission at the visible and near-IR spectrum. Femtosecond-laser processing of glass is of particular interest because of the laser’s ability to micromachine and modify such material in 2-D and 3-D. Together, these qualities make them an excellent combination for integration of microfluidics with optical components. Other materials such as polymers can be used for LOC fabrication, but this thesis will be focused on laser fabrication in glass, and in particular, fused silica glass which has superior optical and chemical properties as well as fabrication versatility as discussed in Section 2.2.2.

Femtosecond-laser energy is highly absorbed in materials such as metals and semiconductors due to their large density of free carriers at room temperature available for strong laser-material interaction. In contrast, transparent materials such as glasses have wide bandgaps that exceed the laser photon energy, and are weak for linear optical absorption. The dominating laser-glass interaction therefore involves a nonlinear process, where valence electrons are promoted to the conduction band through nonlinear photoionization
such as multiphoton \textsuperscript{95} and/or Zener tunneling \textsuperscript{96} ionization depending on the laser intensity and frequency. At sufficient intensity and/or pulse duration, the subsequent free-carrier absorption results in collisional ionization leading to avalanche ionization.

At sufficient laser intensity and high optical frequency, which is still below the frequency needed for single photon absorption, multiphoton absorption is the dominant mechanism for nonlinear photoionization. At high laser intensity, nonlinear photoionization proceeds through Zener tunneling \textsuperscript{97}. Multiphoton ionization is frequently the dominant excitation process for waveguide writing with fs lasers due to the threshold level of laser interaction required inside a wide bandgap material \textsuperscript{98}. Once the energy of the fs laser is absorbed inside the focal volume through the optical breakdown processes described above, electrons transfer this energy to the lattice. However, the exact physical mechanisms for material modification and subsequent refractive index change are not fully understood. When laser energy is deposited inside the material, it is locally heated to a very high temperature. As the heated volume cools down, the density is modified locally through stress redistribution that alters the refractive indices \textsuperscript{99}. Another mechanism that is believed to induce refractive index change is attributed to laser-induced color centers through the Kramers-Kronig relation \textsuperscript{100}. One other possible mechanism proposed is the rearrangement of the network of chemical bonds in the glass matrix leading to a density increase \textsuperscript{101}. The combined effects from these three mechanisms are widely accepted to contribute to fs-laser modification of the refractive index.

2.5.1.1 Laser-Induced Modifications

In general, the laser-induced modification of fused silica glass can be classified into three regimes of structural change from the observed morphology depending on the laser pulse duration and energy \textsuperscript{102}: formation of nanograting-free smooth modification, self-ordered nanogratings, and disruptive modification. In the formation of nanograting-free
modification using low pulse energies (e.g. \( \sim 100 \) nJ for 800-nm, \( \sim 200 \)-fs laser pulses at 100-kHz repetition rate focused with a 0.65 numerical aperture (NA) lens) \cite{102}, Chan et al. found a significant increase of 3 and 4 member ring structures, indicating the presence of densification and attributing this to yield isotropic refractive index change \cite{101}. In this regime, the structural modification takes the form of bond rearrangement without destroying the overall integrity of the glass, forming smooth and uniform refractive index modification suitable for low-loss optical waveguide fabrication.

The regime that produces ordered sub-wavelength nanogratings is defined with exposure at higher pulse energies (e.g. \( \sim 100-300 \) nJ for 800-nm, \( \sim 200 \)-fs laser pulses at 100-kHz repetition rate focused with a 0.65 NA lens) \cite{102}. The laser-formed nanogratings align perpendicularly to the laser polarization direction to provide a high HF-etching rate and selectivity that follows the grating direction \cite{14}. Shimotsuma et al. proposed a model based on the interference between the incident light field and the electric field of the bulk electron plasma wave that results in the periodic modulation of electron plasma concentration and the structural changes in fused silica glass \cite{103}. An alternative model was proposed to explain the nanograting formation by Bhardwaj et al. based on the transient nanoplasmonic theory \cite{104}. In this model, the nanograting forms over a three-step process that takes place over many laser pulses. In the first step, inhomogeneous dielectric breakdown from multiphoton ionization begins with intrinsic inhomogeneity, metastable color centers or defects to form into nucleation centers that lead to the formation of localized spherical nanoplasmas over successive laser pulses due to a memory effect. In the second step, the field enhancement at the plasma boundary results in an asymmetric growth of the underdense spherical nanoplasmas, forming into nanoplanes aligned in the direction perpendicular to the laser polarization. Finally, the randomly distributed nanoplanes, having plasma electron density exceeding the critical density, affect the laser light propagation similar to that by a planar metallic waveguide. The parallel nanoplanes are expected to separate by \( \lambda_0/2n \), where \( \lambda_0 \) is the laser wavelength.
Figure 2.4: SEM images of nanogratings, formed 65 µm below the fused silica surface with linear polarization (a) parallel and (b) perpendicular to the scanning direction, etched for 20 min in 0.5% HF. (c) Top-view optical images of HF etched (8 hours @ 2.5% HF) micro-channels demonstrating the highly selective etching with laser polarization parallel (top), linear 45° (middle), and perpendicular (bottom) to the scanning direction [14].

and $n$ is the refractive index of the material. Hnatovsky et al. observed these periodic sub-wavelength nanoplanes after HF etching the fs-laser irradiated tracks as shown in the scanning electron microscope (SEM) and optical images of Figure 2.4.

This nanograting-forming regime can be used for fabrication of Bragg filters and attenuators in optical fibers or waveguides [105]. Moreover, the produced nanogratings allow high etch rate (for perpendicular polarization or circular polarization) [14] that is suitable for FLICE microfluidic channel fabrication as discussed in Section 2.6.

For high laser pulse energies (e.g. > 300 nJ for 800-nm, ~200-fs laser pulses at 100-kHz repetition rate focused with a 0.65 NA lens) [102], disruptive modification produces a less dense or hollow cavity surrounded by a region of compacted or densified material [106]. Such a structure is a result of immense pressure driving shockwave formation that propagates radially outward. During cooling, the material does not anneal, and the denser phase is frozen in. Although, the onset of this regime may be detrimental to
waveguide or microfluidic channel fabrications, it can be used for 3-D memory storage as demonstrated by Glezer et al. [107].

### 2.5.2 Laser-Glass Patterning

The technique of DLW provides 3-D microstructuring by scanning a tightly focused laser beam into the bulk of the glass substrate. Through nonlinear multiphoton [95] and/or Zener tunneling [96] photoionization and avalanche ionization [95,96], local modifications are induced in the focal volume that will carve out the desired pattern by either scanning with lens movements or stage movements as shown in Figure 2.5. Modification tracks form freely in the transverse (x- and y-axis) and longitudinal (z-axis) directions to provide 3-D micro- or nano-structures in arbitrarily shaped patterns.

For microscope objective lenses, the working distance of several millimeters is very limited and scanning in the longitudinal direction poses a length restriction. A laser writing technique using an axicon for focusing femtosecond pulses provide an alternative
to longitudinal writing. An axicon is a conical lens that produces a Bessel beam having a long depth of field and a sharp central spot, thereby distributing the focus over a long line (> 1 cm) with a width of few microns without requiring translation of the sample [108].

In the transverse writing geometry, such working distance (i.e. several millimeters) is sufficiently flexible for writing 3-D optical circuits and modification tracks for optofluidic chip applications. Further, the length limitation is alleviated, offering unrestricted patterning over the entire area of substrate. However, this writing scheme produces elliptical cross-sectional modification zones, which may be undesirable for applications such as waveguide writing. To overcome this shortcoming, various approaches have been explored including the use of astigmatic focusing with a slit [109] and asymmetric focusing with a cylindrical lens [110] to extend the beam waist to match the depth of focus. In exchange of poorer resolution with these methods, laser tracks with circular cross-section are formed. Also, spherical aberration at the air-glass interface becomes significant at higher scanning depth, and depth-corrected exposure is required to fabricate structures with consistent cross-section. Approaches including the use of high-NA oil immersion lens [111] and objective lens with a correction collar [102] have been demonstrated to effectively control the size of the modification zone and compensate for spherical aberration at varying depths.

2.6 Femtosecond Laser Irradiation with Chemical Etching

Femtosecond laser irradiation with chemical etching (FLICE) is a technique that uses the benefits offered by a focused laser beam to provide permanent micron to sub-micron modifications through nonlinear absorption, followed by the chemical etching of the laser-modified regions that are susceptible to a higher etch rate than the unexposed material by using dilute HF [12, 14, 112] or KOH [18]. Hnatovsky and coworkers demon-
strated that laser exposure of fused silica glass with a laser polarization perpendicular to
the scan direction offered a 70-fold increase in HF etching rate compared with parallel
polarization [14]. They associated this high differential etch rate with the formation of
volume gratings with nano-planes aligned orthogonal to the polarization direction (refer
to Section 2.5.1.1) that opens the flow of HF solution along the laser modification track.
Circular polarization offered a similar high contrast in etch rates to those achieved with
perpendicular polarization due to the disordered nature of the nanostructures formed
which in turn do not restrict the flow of HF acid along the laser modification track.
Differential etch rates as high as 280:1 between laser modified and unmodified glass have
been reported [102] for exposure with 40 fs pulses with an energy of 200 nJ, followed by
2.5% HF solution.

While this FLICE technique permits the formation of high aspect ratio micro-channels
in fused silica glass, the embedded channel length is often limited to less than 4 mm before
tapering and uncontrollable cross-sectional shape evolve to limit this application direction
for the LOC community [4,14]. OSELLAME et al. introduced a conical wobbling technique
to create long straight and uniform cylindrical micro-channels but this technique may
not be flexible for creating other cross-sectional shapes or non-straight channels [16]. Re-
cently, buried micro-channels of nearly uniform cross-section were formed up to 9.2 mm
length, but required a high concentration (10 M) of aqueous KOH and elevated temper-
ature of 80 °C to yield only a moderate etch rate of ∼1.7 μm/min [18].

The post fs-laser irradiation enhancement in chemical-etching rate of fused silica may
be due to the combination of two mechanisms [51]. The first mechanism is related to
the densification of silica in which the number of 3 and 4 member silica ring structures
increase at the expense of the 5 or 7 member rings [113]. The reduction of the bridging
bond angle in the resulting densified silica increases the reactivity of the oxygen atoms
due to the deformed configuration of their valence electrons [12]. This in turn results
in increased etching in the laser-modified zones compared with the unmodified zones.
This mechanism is also suited to explain the low intensity modification regime, which corresponds to the formation of optical waveguides.

The chemical process that takes place in the dissolution of silica by aqueous HF is due to the F\(^{-}\) ion attacking the silicon atom, while the H\(^{+}\) ion attacks the oxygen atom according to the following chemical equation

\[
\equiv \text{Si–O–Si} \equiv + \text{HF} \rightarrow \text{Si–OH} + \equiv \text{SiF}.
\]  

Therefore, the formation of the strained Si–O–Si bond in the planar three-member rings are more susceptible to the HF attack compared with six-member rings, which is ascribed to the enhanced dissolution of the laser-modified zone. As for aqueous KOH, the hydroxide ion, OH\(^{-}\), attacks the \(\equiv \text{Si–O–Si} \equiv\) groups, and the bonds are cleaved to give \(\equiv \text{Si–OH}\) and \(\equiv \text{Si–O}^{-}\). Although densification of silica after fs-laser irradiation is a reasonable explanation for the greater susceptibility to the HF etching, the exact mechanism at the molecular level is still not completely understood.

Laser patterning by this FLICE technique has also been employed in photostructurable glass-creamics such as Foturan. This has led to several photonic biosensing demonstrations in photosensitive Foturan glass [5,6,74]. However, the resolution is limited to a few microns due to a completely different laser-material interaction in which the Ce\(^{3+}\) ions donate an electron to Ag\(^{+}\) ions upon laser irradiation and a post-annealing process precipitates colloidal Ag phase. This favours the growth of lithium metasilicate which can then be dissolved in a HF solution to form microstructures with an etching contrast ratio of 43:1 (in 10\% HF solution) compared with unmodified glass.
Chapter 3

Experimental Setup

In this chapter, the central tools for carrying out the LOC fabrication in this thesis, the femtosecond laser along with the beam delivery system, are reviewed in section 3.1 and 3.2 respectively.

3.1 IMRA Fiber-Amplified Femtosecond Laser

The femtosecond fiber-amplified laser system (IMRA America µJewel D-400-VR) provided \(~440\) fs to \(~690\) fs pulses (full-width at half-maximum (FWHM) with Lorentzian fit) at a variable repetition rate of \(100\) kHz to \(5\) MHz, respectively, with output powers ranging from \(~320\) mW to \(~1300\) mW, respectively. This fiber laser is ideal for both a research laboratory and industrial site because of its compact footprint (excluding the free-space compressor) and ease of turn-key operation with waterless cooling. In addition, the variable and moderately high repetition rate and a high average power are well suited for driving strong nonlinear interactions for refractive index modification \(^{114}\) and nanograting formation \(^{103}\) in glasses under flexible conditions appropriate for research purposes.

The fiber-amplified femtosecond laser employs a chirped-pulse amplification (CPA) scheme for the amplification as shown schematically in Figure 3.1. The 300-fs pulses at
Chapter 3. Experimental Setup

Figure 3.1: Schematic showing the CPA in the IMRA µJewel laser. The image is reproduced from [114].

10-mW average power from an Ytterbium-fiber oscillator are stretched to 200 ps pulses that can be amplified at peak intensities below the self-focusing threshold for avoiding excess nonlinearities during propagating and damage within the fiber in the subsequent amplification stages [114]. Following the fiber-amplification, the pulses are recompressed with a bulk diffraction grating to provide a range of pulses with an average power of ∼320 mW at 100 kHz to ∼1.3 W at 5 MHz having pulse durations of ∼440 fs to ∼690 fs (Lorentzian FWHM), respectively. The compressor schematic is shown in Figure 3.2.

The input amplified beam passes under mirror M3 to reflect from mirror M1 to the diffraction grating, where the beam is diffracted and directed to the prism. The beam reflects inside the prism and directs back to the grating the second time. The beam then propagates to mirror M2 (i.e. the end mirror), and reflects back to the grating (third time). The beam reflects internally in the prism again and directs back to the grating (fourth time). Finally, the compressed beam is reflected by mirrors M3 and M4, and exits the compressor. The prism position is adjusted accordingly for varying laser repetition rate. For the majority of this thesis, the laser was operated at a repetition rate of 1 MHz with ∼540-fs pulses providing an average power of ∼1 W. The chosen operating parameters provided smooth micro-channel walls after HF etching and visible waveguide writing which will be described in Chapter 4.
Figure 3.2: Schematic showing the external compressor unit and the beam paths of the IMRA μJewel laser. M = mirrors.
Figure 3.3: Schematic showing (a) the top view and (b) the front view of the beam delivery system for the IMRA µJewel laser. A = Aperture, AOM = Acousto Optical Modulator, HM = Hot Mirror, HWP = Half-Wave Plate, L = Lens, ND = Neutral Density Filter, OL = Objective Lens, P = Polarizer, TM = Turning Mirror.

3.2 Beam Delivery System

The schematic of the beam delivery system is shown in Figure 3.3 depicting the beam path from the fiber laser to the sample on the air bearing motion stages. All the turning mirrors (TMs) in the beam path were oriented at 45° and designed for high reflectivity
at $\lambda = 1045$ nm (CVI Y1-1037-45) and/or $\lambda = 522$ nm (CVI HM-1037-45). The 1045-nm mirrors and 1045/522-nm mirrors were used before and after the lithium triborate (LBO) crystal, respectively. The 200-ps pulses at a wavelength of $\lambda = 1045$ nm generated from the laser head (IMRA $\mu$Jewel D-400-VR) was directed into the external compressor (Figure 3.2) with two turning dielectric mirrors TM1 and TM2. The bulk diffraction grating inside the external compressor recompressed the pulses to the range of $\sim 440$ fs to $\sim 690$ fs at 100 kHz to 5 MHz, respectively. The half-wave plate (HWP) (Thorlabs WPH05M-1053), used to control the laser power, was mounted on a computer controlled Aerotech rotational stage (Aerotech ART310). After passing the HWP, a polarizing beam splitter (CVI PBS-1047-050) was used to ensure the laser polarization was parallel to the optical table surface.

Following the polarizer (P), the beam could be directed through two paths. One beam path went through the acousto optical modulator (AOM) (Neos 23080-3-1.06) for writing complex structures requiring beam modulation, and the second bypassed the AOM for writing simple structures, and both paths reconvened at aperture AP5. All the apertures (APs) mentioned hereafter were used for beam alignment along each beam path. For the first beam path (i.e. AOM beam path), the laser was reflected by turning mirrors TM3, TM4, and TM5 and focused through lenses L1 and L2 into the AOM for beam modulation as shown in Figure 3.4. A converging lens (L1) of focal length 250 mm and a diverging lens (L2) of focal length -50 mm were used to provide a 1-mm diameter focal spot at the AOM for an optimal response time of $\sim 150$ ns. The AOM was digitally modulated by a radio frequency driver (Neos 21080-2DS), which was synchronized with the output from a position-synchronized output (PSO) from the Aerotech A3200 motion controller based on the x, y, or z coordinates of the Aerotech stages. The first diffraction order, with a maximum diffraction efficiency of 60%, was directed through lenses L3 and L4, which were identical, but reversed in order as L1 and L2, to expand and re-collimate the laser back to its original size. Turning mirrors TM6 and TM7 directed the beam
through aperture AP5. For the second beam path (i.e. non-AOM beam path), turning mirrors TM3 and TM7 were removed, permitting the laser to reflect on turning mirror TM14, which directed the laser through apertures AP10 and AP5.

After passing aperture AP5, the laser was reflected by TM8 and directed through aperture AP6, a neutral density (ND) filter, and the telescope assembly consisting of the LBO crystal (Newlight Photonics LBO1663) placed in between L5 and L6. The purpose of the ND filter was to further reduce the laser power for processing low threshold materials such as polymer, but was not used for the current work on fused silica glass.

The schematic of the LBO assembly is shown in Figure 3.5. Second harmonic generation (SHG) was achieved through the LBO crystal from the fundamental wavelength of $\lambda = 1045$ nm with laser polarization parallel with the table surface to the frequency doubled wavelength of $\lambda = 522$ nm with laser polarization perpendicular to the table surface. The LBO crystal, which was placed on a railing system for easy adjustment, was positioned at the focus of L5 and heated inside an oven to $170^\circ$C for optimal conversion efficiency. The hot mirror (HM) (Thorlabs FM01), inserted before L6, was used to reflect the IR wavelength of $\lambda = 1045$ nm, while transmitting the visible wavelength.
of $\lambda = 522$ nm.

The frequency doubled laser beam passed through AP7, and TM9 directed the beam upward toward TM10. The beam was then aligned through AP8 and AP9 before being reflected downward by TM11 toward the Aerotech z-axis stage (Aerotech ALS130). Two parallel turning mirrors, TM12 and TM13, placed on the Aerotech z-axis stage, directed the laser through an objective lens (OL) which was mounted onto an objective holder (Newport LP-1A) with 2-axis translation and tilt control for ease of alignment. For the majority of this work, a $0.55\text{-NA}$ aspherical lens (New Focus 5722-A-H, $40\times$ with $f = 4.5$ mm) was used for focusing the laser beam to pattern optical waveguides, microfluidics and woodpile (WP) structures, while a $0.90\text{-NA}$ objective lens (Nikon 333387 BD Plan 100×) was used for writing scaled-down WP structures meant for PC fabrication.

For increased laser-writing stability, the air-bearing motion stage (Aerotech ALS130) for vertical z-positioning, the linear motion stages (Aerotech ABL1000), one of each for x and y horizontal positioning, together with the sample holder were all mounted on a granite with an arch for the z-axis stage. The motion stages had a 2-nm resolution and 200-nm position repeatability for high resolution scanning of laser tracks. All the
motion stages, including the rotational stage (Aerotech ART310), were connected to a motion controller (Aerotech A3200) that was interfaced to a computer equipped with the NView software program (HMI v2.21) running G-code scripts (refer to Appendices). The average power required for an experiment was measured with a thermal power meter (Ophir 12A) placed on the sample holder.

Prior to laser writing, the sample was first firmly mounted with magnets, and the laser focus position was found with the aid of the CCD camera (Sony XCD-X710) with an \( \infty \)-conjugate-focusing zoom-lens (Computar L5Z6004) overhead. A weak laser beam (below the threshold of modification) was focused and positioned onto the sample surface by observing the CCD image of the Fresnel reflection that leaked through mirror TM11 and directed toward the CCD camera by reflecting off mirrors TM12 and TM13. To ensure the sample was mutually orthogonal in the x-direction and y-direction with respect to the optical axis (z direction), the tilt stage (Newport TTN80) attached below the sample holder was used to correct the angular deviation until back reflections were made collinear with the incoming laser beam path. Sample adjustment was completed when the smallest reflected laser spot was observed on the CCD image while the xy-motion stages translated the sample over the entire writing area. Such an iterative correction procedure enabled the laser focus to be placed within \( \pm 2 \) \( \mu \)m of the sample surface over the entire sample. This configuration was only possible with a collinear laser beam. Any divergence in the incoming beam required an offset correction of z-focus position that could be calibrated by observing the ablation threshold at smallest laser power at assumed best focus. The sample surface non-uniformities was found to be negligible and did not affect the quality of the microfluidic and optical structures that were fabricated.
Chapter 4

Lab-on-a-Chip Component Designs and Fabrication

In this chapter, the micro-fabrication procedures, characterizations, results and discussions of the individual LOC components (i.e. micro-channels, reservoirs, 3-D periodic structures, and optical waveguides) are discussed.

In traditional wet-lab processes for LOC fabrication, microfluidic structures are produced in a planar geometry, which limits the routing of isolated channels across the surface without cross contamination. The FLICE technique enables the fabrication of 3-D microfluidic layouts that greatly improve the flexibility of LOCs and the ability to integrate optical waveguides together with novel 3-D structures for enhanced capabilities. Section 4.1 presents the 3-D fabrication of buried microfluidic channel with variable cross-section, reservoir, and IWP structure. Section 4.2 presents the fabrication of optical waveguides for operation in the visible spectrum.

4.1 Three-Dimensional Microfluidic Structures

For LOC devices, the most fundamental components are microfluidic channels and reservoirs. Reservoirs offer continuous supply of analyte or serve in waste storage, whereas
micro-channels connected to reservoirs provide a pathway for fluid transport. Further, to enhance the diagnostic and analytical capabilities of LOCs, IWP and PC structures may be developed for chromatography and optical sensing purposes. Using the FLICE technique, exposure recipes were developed here for the fabrication of the aforementioned components for LOC devices. A comprehensive study to optimize the laser exposure for fabricating cross-sectional controllable micro-channel with smooth walls and unrestricted length is presented in Section 4.1.1. The reservoir is one of the largest components on a LOC, and a novel method to dramatically reduce the laser exposure time is presented in Section 4.1.2. To extend on the fabrication of micro-channels, 3-D interlacing micron-sized capillaries to form stress-free and high-density meshes of IWP structures is presented in Section 4.1.3.

### 4.1.1 Buried Micro-Channel

The FLICE technique permitted the formation of high aspect ratio cylindrical micro-channels embedded in bulk fused silica, but the literature review (Section 2.6) revealed the current limitation of channel length of up to 4 mm \[4\] and 9.2 mm \[18\] with HF and KOH etching, respectively. To overcome these restrictions in an effort of opening up applications of LOC devices, the FLICE technique with fs-laser processing of fused silica was modified to produce large buried channels with controllable shaping of the cross-sectional profile by forming multi-scan modification tracks with the fiber-amplified fs laser. A comprehensive study of the differential etch rates as a function of laser polarization, laser energy, sample scanning speed, sample scanning angle, and multi-line scanning defined optimized exposure conditions to form exceptionally smooth-walled rectangular micro-channels in addition to circular and elliptical shape of varying size and aspect ratio, demonstrating the control of the cross-sectional shape of the buried channel. In addition, a new technique of incorporating vertical access ports was developed to extend the reach of HF etching and form channels of unlimited length, thereby removing the need
Figure 4.1: Schematic showing the multi-scanned laser modification zones for elliptical/circular and rectangular cross-section patterns scanning in y-axis ($\theta = 0^\circ$) direction where $d_s$ is the depth from the surface, $d_T$ is the transverse line-to-line separation along the x-axis, $d_L$ is the layer-to-layer separation along the optical z-axis. Vertical laser tracks were also formed to provide multiple access ports for HF etching.

for an ultrasonic bath in the post-etching process to facilitate the formation of delicate microstructures without inducing cracks in the substrate.

For the laser exposure optimization and fabrication of micro-channels, the fs laser was applied at 522 nm wavelength and 1 MHz repetition rate with pulse energy ($E_p$) ranging from 50 to 200 nJ to expose fused silica (Corning 7980) substrates. The laser beam was focused with a 40× aspherical lens of 0.55 NA into the transparent glass substrate at depths ($d_s$) varying from 75 to 210 $\mu$m below the surface. Laser modification tracks were formed with parallel, perpendicular, and circular polarized beams scanned transversely at speeds ($v_s$) ranging from 0.2 to 10 mm/s with the 3-axis air-bearing translation stage (Aerotech ABL1000 motion stages). The details of the beam delivery and second harmonic setup are described in Section 3.2.

To shape the cross-section of embedded micro-channels, single- and multi-scanned laser modification tracks were written inside the glass plate under variable exposure con-
Figure 4.2: Schematic showing the laser-scanned fan structure for investigating the nanograting formation and etch-rate dependence on laser scanning angle with fixed polarization orientation of the laser.

Rectangular cross-sections were studied by forming a $5 \times 5$ array of modification tracks with layer-to-layer separations ranging from $d_L = 1$ to $12 \, \mu m$ and track-to-track separations ranging from $d_T = 1$ to $3 \, \mu m$. Elliptical and circular shaped micro-channels were also formed with various array geometries as shown in Figure 4.1. All laser tracks were scanned layer-by-layer from bottom to top to avoid propagation of the laser through modified scattering volumes.

After laser patterning, the end facets were ground and optically polished before wet etching the substrate in 2.5 to 5% concentration of HF for 1 to 3 hours at room temperature without the use of an ultrasonic bath. In order to fabricate embedded micro-channels with unrestricted length and minimize the tapering effect seen over long micro-channels, small access ports were periodically formed as depicted in Figure 4.1 by scanning the laser vertically from the surface at a speed of $v_s = 1 \, mm/s$ to the transverse modification tracks with a pulse energy of $E_p = 100 \, nJ$ and thus provide multiple access points for the dilute HF acid to reach the buried transverse tracks. The spacing of the vertical access ports were optimized together with laser exposure condition to form uniform channel cross-section over long lengths.
In order to investigate the possibility of creating uniformly shaped micro-channels in circular or arc-shaped pathways, etch rates were examined for a variety of polarization conditions (parallel, perpendicular, and circular) and scan directions in 1° increments over $\theta = -45^\circ$ to $+45^\circ$ relative to the y-axis ($\theta = 0^\circ$ in Figure 4.1) shown schematically in Figure 4.2. The single tracks were written at a speed of 0.5 mm/s with energies in the range of $E_p = 75$ to 150 nJ and depths of 75 to 210 $\mu$m from the surface. The laser-induced nanograting structure was also examined on polished sample facets with a SEM (Leo 1540 SEM) prior to HF etching.

4.1.1.1 Nanograting Formation

For the range of laser exposures applied to write microfluidic channels, the orientation of the nanogratings were aligned parallel with respect to the scanning direction when the laser polarization (E-vector) was perpendicular to the scanning direction. Nanogratings aligned perpendicular to the scanning direction were also examined for the purpose of writing optical waveguide, which is discussed in Section 4.2. Circular polarization was also examined, yielding randomly oriented nanogratings. Figure 4.3 shows the cross-sectional SEM (LEO (Zeiss) 1540XB) images of laser tracks written at $E_p = 100$ nJ and 125 nJ with perpendicular (Figure 4.3(a)), parallel (Figure 4.3(b)) and circular (Figure 4.3(c)) polarizations having the nanogratings clearly formed parallel, perpendicularly, and randomly with respect to the scanning direction. For perpendicular polarization, the nanograting period was observed to decrease from $\sim180$ to $\sim120$ nm as the pulse energy increased from 75 to 150 nJ (Figure 4.4), which is contrary to the observation by Shi-motsuma et al. for their condition of linear polarization with static exposure and longer 800 nm wavelength [103], but consistent with Kazansky and coworkers for linear polarization with wavelengths of 522 nm, 800 nm and 1045 nm [115]. The nanoplasmonic model developed by Bhardwaj et al. [104] predicts an energy-independent nanoplane spacing of $\lambda_0/2n = 180$ nm for the present $\lambda_0 = 522$ nm wavelength, which is only consistent
Figure 4.3: SEM images showing the formation of nanogratings at $E_p = 100 \text{ nJ}$ and $125 \text{ nJ}$ with polarization (a) perpendicular, (b) parallel, and (c) circular with respect to the scanning direction, written at a scanning speed of $v_s = 0.5 \text{ mm/s}$ and a depth of $d_s = 150 \mu\text{m}$ with the laser focusing from the right.

with the exposure of $75 \text{ nJ}$ pulse energy. More advanced models are clearly necessary to explain these disparate observations, but is out of the scope of this thesis.

4.1.1.2 Etch Rate Dependence on Polarization, Pulse Energy, Scanning Speed and Scanning Angle

An average HF etch rate was assessed for single-scan modification tracks formed in fused silica over a large range of laser exposure conditions. Perpendicular (x-axis in Figure 4.1), parallel (y-axis), and circular (xy-plane) polarization of the laser were tested for laser scanning along the y-axis to form tracks perpendicular to the facet ($\theta = 0^\circ$). The average etching rates are shown in Figure 4.5 as a function of pulse energy for a scanning speed of $v_s = 0.5 \text{ mm/s}$ and a focal depth of $d_s = 150 \mu\text{m}$ after etching for 1 hour in 5% HF.
Figure 4.4: Nanograting period versus pulse energy plot for nanogratings formed at a scanning speed $v_s = 0.5$ mm/s and a perpendicular polarization with respect to the scanning direction.

Figure 4.5: Etch rate (5% HF for 1 hour) of single laser modification tracks ($v_s = 0.5$ mm/s, $d_s = 150$ µm) with three different polarizations (perpendicular, parallel, and circular) versus pulse energy.

The etch rate for the perpendicular polarization was $\sim 5.5$ µm/min over a wide exposure.
range of 50 to 200 nJ. A slightly lower etch rate of $\sim 5.0 \, \mu m/\text{min}$ was observed for circular polarization for pulse energies above 75 nJ. In sharp contrast, the etch rate for the parallel polarization was $3 \times$ to $28 \times$ lower, varying from $\sim 0.2 \, \mu m/\text{min}$ at 50 nJ pulse energy to $\sim 1.5 \, \mu m/\text{min}$ at 200 nJ pulse energy. The considerable contrast of $\sim 28:1$ in differential etch rate between perpendicular and parallel polarizations at $E_p = 50$ nJ follows the expected outcome where the orientation of the nanograting perpendicular to the scanning direction for parallel polarization yields stop-layers that hinder the progress of the etchant along the modification track while nanograting planes running along the laser track for perpendicular polarization are effectively open to direct the etchant rapidly along the laser modified track. Higher differential etch rates of 70:1 were reported by Hnatovsky et al. [14, 102] using very different exposure conditions of laser wavelength (800 nm vs. 522 nm), pulse duration (40 to 500 fs vs. 220 fs), and scanning speed (0.03 to 0.11 mm/s vs. 0.2 to 10 mm/s). Nevertheless, the overall etch rate for the perpendicular polarization case here is similar to the $\sim 5 \, \mu m/\text{min}$ etch rate reported by Hnatovsky et al. [14, 102]. The relatively constant etch rate of $\sim 5.5 \, \mu m/\text{min}$ for perpendicular polarization throughout a broad processing window of $E_p = 50$ to 200 nJ indicates formation of high contrast nanograting planes aligned along the scanning direction. However, the channel cross-section increased from $5 \, \mu m \times 12 \, \mu m$ to $7 \, \mu m \times 22 \, \mu m$ over this pulse energy range. For all polarizations, chemical etching was not observed for laser pulse energy of 25 nJ or smaller.

The polarization and pulse energy dependence on etch rates for other scanning speeds were similar to the trend shown in Figure 4.5 but with etch rate decreasing as the scanning speed increased as shown in Figure 4.6. At a 100-nJ pulse energy, the etch rate remained constant at scanning speeds from 0.2 to 1.0 mm/s for perpendicular and circular polarizations, but decreased $\sim 50\%$ for parallel and circular polarizations, and decreased $\sim 10\%$ for perpendicular polarization as scanning speed increased from 1.0 to 10 mm/s. The reduced etch rate can be attributed to an expected lower volume of material modi-
Figure 4.6: A representative graph of etch rate (5% HF for 1 hour) of single laser modification tracks ($E_p = 100$ nJ, $d_s = 150$ µm) versus pulse energy with three laser polarizations (perpendicular, parallel, and circular).

The scanning depth was also found to affect the HF etch rate. Using a constant pulse energy $E_p = 100$ nJ, an increase in the focusing depth from $d_s = 150$ to 210 µm produced different etching trends, decreasing the etch rate by $\sim 3\%$ for perpendicular polarization, and increasing by $\sim 50\%$ for parallel polarization and increasing by $\sim 5\%$ for circular polarization. The increasing spherical aberration produced by the glass surface at increased focusing depth [102] presumably weakens the nanograting formation by spreading out the focused energy, resulting in diminished nanogratings. Therefore, the less well defined parallel nanogratings cause a nearly insignificant drop in etch rate for the perpendicular polarization but a large increase for the case of parallel polarization as the stop-layers become less effective.
Figure 4.7: Laser scanned fan structures written at $v_s = 0.5 \text{ mm/s}$ with (a) perpendicular polarization, (b) parallel polarization, and (c) circular polarization at $E_p = 100 \text{ nJ}$ and $d_s = 210 \mu\text{m}$, followed by 5% HF etching for 1 hour. The darker lines show the laser modification tracks that were opened by the HF acid while the lighter tracks were unetched.
Figure 4.7 shows optical images of laser modification tracks written with 100 nJ pulse energy, 210 µm focal depth, and three different polarizations (perpendicular, parallel, and circular) obtained over a range of scanning angles $\theta = -45^\circ$ to $+45^\circ$ to the facet normal and following one hour of etching in 5% HF solution. For the circular polarization, an inferred etch rate of $\sim 4.7$ µm/min from an average hole depth of $\sim 280$ µm was found that was independent of the scan angle ($\theta$) from all observed angles of $\theta = -45^\circ$ to $+45^\circ$. This etch rate invariance, which is shown in Figure 4.7(c) (darker lines = etched tracks), follows from the anticipated random orientation of grating nanostructures for circularly polarized light and is attractive for directionally independent formation of uniform micro-channel shapes in any laser scanning direction (xy-plane) within the substrate.

A similar analysis for linear polarization (Figure 4.7(a) and (b)) yielded etch rates that were strongly dependent on the angle, $\phi$, measured between the laser polarization and the channel scan direction ($\theta$), yielding the plots shown in Figure 4.8(a) for various writing depths and Figure 4.8(b) for various laser pulse energies. The full angle range of $\phi = 0^\circ$ to $180^\circ$ was obtained by stitching together data from laser exposures scanned from $\phi = 45^\circ$ to $135^\circ$ (Figure 4.7(a)) with polarization perpendicular to the y-axis (Figure 4.1) and laser exposures scanned from $\phi = 0^\circ$ to $45^\circ$ and $\phi = 135^\circ$ to $180^\circ$ (Figure 4.7(b)) with polarization fixed parallel to the y-axis. A peak etch rate of $\sim 5.5$ µm/min was observed for all pulse energies $E_p = 50$ to 200 nJ and focal depths $d_s = 75$ to 210 µm shown here for perpendicular polarization ($E_\perp, \phi = 90^\circ$), but seen to decrease strongly by up to 5 fold for $E_p = 75$ nJ and $d_s = 75$ µm over only a narrow $10^\circ$ change in polarization angle ($\phi = 90^\circ$ to $80^\circ$ or $90^\circ$ to $100^\circ$). In order to take advantage of an etch rate within 96% of the maximum value, the laser polarization must be accurately oriented within a narrow angle range or $\phi = 89^\circ$ to $91^\circ$ to the scanning direction that may be challenging when forming looped or curved paths for the micro-channels. The strong etching rate contrast of $28\times$ between perpendicular ($E_\perp, \phi = 90^\circ$) and parallel ($E_\parallel, 0^\circ$ and $180^\circ$) polarizations is also clearly evident in Figure 4.8(a) and Figure 4.8(b) and consistent with the values
The etch rate discontinuities at $\phi = 45^\circ$ and $135^\circ$ in both Figure 4.8(a) and Figure 4.8(b) arise from the orthogonal orientation of nanogratings formed either parallel (y-axis polarization) or perpendicular (x-axis polarization) to the facet when scanning at either $\theta = \pm 45^\circ$ direction to the surface. For both parallel and perpendicular polarization at this scan angle, nanogratings are aligned at $45^\circ$ to the modification track that should yield identical etch rates in the bulk. However, there is an anomaly where the initial etching rate is inhibited at the facet for parallel polarization which positions nanograting stop-layers parallel with the facet, while nanogratings oriented perpendicular to the facet would enhance the initial etching for perpendicular polarization.

A geometric consideration of the orientation of nanoplanes with the track direction (subtending angle, $90^\circ - \phi$) suggest that the slow etch rates found for parallel polarization, $R_\parallel = 0.2 \ \mu\text{m/min}$ ($E_p = 75 \ \text{nJ}, d_s = 75 \ \mu\text{m}$) at $\phi = 0^\circ$ or $180^\circ$, will scale up trigonometrically as follows:

$$R(90^\circ - \phi) = R_\parallel \csc(90^\circ - \phi) = R_\parallel \sec(\phi).$$

(4.1)

Here, dramatically faster ($28 \times$) etching along the nanoplanes enhances the slow $R_\parallel$ etch rate as the apparent stop-layers rotate with $\sec(\phi)$ to align with the channel direction. The secant lines (dashed lines) in Figure 4.8 closely match the experimental etch rates for exposure conditions of $E_p = 75 \ \text{nJ}$ with $d_s = 75 \ \mu\text{m}$ and $150 \ \mu\text{m}$. The secant fit is capped at the peak etch rate of $R_\perp = 5.5 \ \mu\text{m/min}$ for perpendicular polarization in the angle range of $\phi = 87.8^\circ$ to $92.2^\circ$. As the etching depth (Figure 4.8(a)) and pulse energy (Figure 4.8(b)) increase, the observed etch rates deviated from the secant fit where the uniformity and quality of nanograting stop-layers were apparently degraded by higher optical aberration expected when focusing deeper into the glass [102,116], as well as by

reported above.
Figure 4.8: Average etch rate (5% HF after 1 hour) of single-scan modification tracks written at \( v_s = 0.5 \) mm/s versus angle between laser polarization (linear) and scanning direction for exposures of (a) \( E_p = 75 \) nJ and depths of \( d_s = 75 \) µm, 150 µm and 210 µm, and (b) \( d_s = 150 \) µm and pulse energies of \( E_p = 75 \) nJ, 100 nJ, 125 nJ and 150 nJ. The black dashed lines are secant fits according to Equation 4.1.
higher pulse energy. Hence, the strongest contrast in parallel and perpendicular polarization etch rates were found in channels formed at shallow depth of 75 µm and low pulse energy of 50 nJ to provide the most uniform and highest contrast nanograting structures for etching straight channels. However, the angle-invariant etch rate found for circular polarization (Figure 4.7(c)) is generally more attractive for fabricating circular or arc-shaped micro-channels (as will be shown in Chapter 5) whereas linear polarization requires the rotation of polarization to follow the changing scan direction to facilitate rapid etching along curved laser tracks.

In summary for single track laser writing, a wide laser processing window was found for enabling fast HF acid etching of 5.5 µm/min for perpendicular polarization and 5.0 µm/min for circular polarization over a wide range of focusing depth, from 75 to 210 µm, and scanning speeds ranging from 0.2 to 1.0 mm/s with pulse energy in respective ranges of $E_p = 50$ to 200 nJ and 75 to 200 nJ. The combination of the results from Figure 4.5 and Figure 4.8 indicate a new fabrication window of low laser pulse energy for efficient nanograting generation to provide high polarization contrast etching of micro-channels. Further, the high etching contrast is attractive for integrating optical waveguides with such channels; however, rectangular cross-sectional shaped channels with smooth side-walls and without cracks would be highly desirable.

### 4.1.1.3 Flexible Cross-Sectional Shaping

In this section, a method is presented for creating arrays of multiple laser exposure lines that are optimized in their vertical, $d_L$, and transverse, $d_T$, track-to-track spacing to control the cross-sectional profile of microfluidic channels with the objective of forming crack-free and open channels with smooth morphology.

Figure 4.9(a) provides a reference showing a backlighting microscope image of an end view of a single laser track written with a pulse energy of 50 nJ observed before (top) and
Figure 4.9: Cross-section microscope images of laser exposed and etched (5% HF after 1 hour) (a) single track and (b)–(f) rectangular shaped micro-channels laser written with perpendicular polarization at depth $d_s = 150 \mu m$, scanning speed $v_s = 0.5 \text{ mm/s}$ and pulse energy (a)–(e) $E_p = 50 \text{ nJ}$ and (f) $E_p = 125 \text{ nJ}$. The line-to-line spacing in rectangular channels were (b), (f) $d_T = 2 \mu m$ and $d_L = 3 \mu m$, (c) $d_T = 3 \mu m$ and $d_L = 3 \mu m$, (d) $d_T = 3 \mu m$ and $d_L = 6 \mu m$, and (e) $d_T = 3 \mu m$ and $d_L = 12 \mu m$. The laser was incident from the bottom.
after (bottom) HF etching. The etched single track micro-channel appears dark under backlighting after 1 hour in 5% dilute HF and increased in cross-sectional area compared with the original laser modification size from $2 \mu m \times 6 \mu m$ to $5 \mu m \times 11 \mu m$. The elliptical cross-sectional shape approximately follows the laser profile of 1-µm focal spot diameter and 6-µm depth-of-focus expected with a 40× aspherical lens and 522-nm laser light.

After sufficient etching time, arrays of single-line modification tracks would open up to each other to form into a large single channel over long lengths as shown in the images of Figures 4.9(b) to (f). These representative samples of multi-track arrays examine a range of horizontal spacing of $d_T = 2$ to $3 \mu m$, and vertical spacing of $d_L = 3$ to $12 \mu m$. With a large spacing of $d_T = 3 \mu m$ and $d_L = 12 \mu m$ as shown in Figure 4.9(e), the top and bottom walls are strongly corrugated as channels in horizontal rows have opened, while etching was not completed between these layers, creating a stack of thin and parallel micro-channels of interest for parallel channel cytometry. A single open channel with improved sidewall smoothness was found as the vertical offset was decreased to $4.5 \mu m$ and the horizontal spacing was decreased to $2 \mu m$ as shown in trend from Figures 4.9(e) to (b) with $d_T$ from 3 to $2 \mu m$ and $d_L$ from 12 to $3 \mu m$. A trade-off balancing the formation of relatively smooth wall surfaces against longer laser scanning time was found over a large range of line-to-line separation of $d_T = 1$ to $2 \mu m$ and $d_L = 1.5$ to $6 \mu m$.

One limitation with the multi-scan cross-section shaping of micro-channels is the high stress accumulated at the corners of the laser-modification-track arrays, as seen from birefringence observations under an optical microscope (Figures 4.9(b) to (f)). Under sufficient laser exposure, such stresses induce micro-cracks as shown in Figure 4.9(f) that result in the finger-like open structure seen after HF etching. For perpendicular polarization, crack formation was not evident in the present array geometries written at depth $d_s = 150 \mu m$, energy of $E_p < 125$ nJ, and scanning speeds ranging from 0.2 to 10 mm/s. Micro-cracks only become apparent as the depth increased to $d_s = 210 \mu m$ at speeds of
$v_s < 1 \text{ mm/s}$. On the other hand, for parallel and circular polarizations, cracks were observed at depths of $d_s = 150 \mu\text{m}$ and $210 \mu\text{m}$ with $E_p > 100 \text{ nJ}$ and scanning speeds of $v_s < 1 \text{ mm/s}$. Parallel and circular polarizations are more susceptible to initiate cracks as the nanogratings forming perpendicularly and randomly along the scanning direction, respectively, build up a higher stress along the channel compared with the case where the nanogratings align along the scanning direction.

The laser exposure in ranges applied here are known to generate a positive refractive index change that is associated with densification of the glass substrate \[101\] and manifests in the stress fields seen as the bright regions in the glass surrounding the micro-channels as observed in Figure 4.9. The cracks were mostly concentrated along sharp low-radius corners of rectangular structures where stress fields are seen to flare out diagonally as observed in Figure 4.9(b) to (f). The finger-like structure formed after chemical etching of the micro-cracks also suggested the region surrounding the micro-crack is densified and this zone etched at a similar rate as the modified glass. Spontaneous cracking after exposure and HF etching is undesirable and limits the total exposure acceptable, favouring exposure at pulse energies of $E_p < 125 \text{ nJ}$ for perpendicular polarization, and $E_p < 75 \text{ nJ}$ for parallel and circular polarizations at moderate scanning speeds of $v_s = 0.5$ to $1 \text{ mm/s}$. In order to fabricate high contrast and crack-free micro-channels of flexible cross-sectional size and in a rapid etching time, laser polarization perpendicular to the scanning direction is preferred to produce nanogratings aligned parallel with the channel with exposure conditions in the following ranges: scanning speed of $0.5 \text{ mm/s} \leq v_s \leq 1.0 \text{ mm/s}$, pulse energy of $50 \text{ nJ} \leq E_p \leq 125 \text{ nJ}$, and focusing depth of $75 \mu\text{m} \leq d_s \leq 150 \mu\text{m}$.

Figure 4.10 demonstrates the advantage of the multi-scan method in flexibly shaping the cross-section of micro-channels. The microscope images show various rectangular and nearly circular shaped micro-channels laser written with the laser parameters of
Figure 4.10: Cross-section microscope images of laser exposed and etched (5% HF after 1 hour) (a),(b) rectangular and (c),(d) circular/elliptical shaped micro-channels laser written with $E_p = 75$ nJ, $v_s = 0.5$ mm/s and $d_s = 150$ µm. Rectangular channels and circular/elliptical channels were written with perpendicular laser polarization and circular laser polarization, respectively. The line-to-line spacing was (a) $d_T = 1$ µm and $d_L = 3$ µm, (b) $d_T = 2$ µm and $d_L = 3$ µm, and (c),(d) $d_T = 2$ µm and $d_L = 1.5$ µm. The laser was incident from the top.

The rectangular shaped micro-channels in Figures 4.10(a) and 4.10(b) were laser written with a perpendicular polarization, and the circular/elliptical shaped micro-channels in Figures 4.10(c) and 4.10(d) were written with a circular polarization.

The above optical microscope assessment provided a broad laser exposure window (0.5 mm/s $\leq v_s \leq$ 1.0 mm/s, 50 nJ $\leq E_p \leq$ 125 nJ, $d_s < 150$ µm, 1 µm $\leq d_T \leq$ 2 µm and 1.5 µm $\leq d_L \leq$ 6 µm) for creating crack-free and smooth surfaces in the rectangular micro-channels, with perpendicular polarization appearing to yield the smoothest surfaces as shown in the optical microscope images (Figure 4.11) with single and multi-scanned micro-channels written at $E_p = 75$ nJ. Further optimization based on SEM analysis (Hitachi S-4500) and atomic force microscope (AFM) (Veeco Dimension 3000) yielded the best surface smoothness as shown for a micro-channel fabricated with perpendicular polarization in Figure 4.12(a) for the channel bottom and Figure 4.12(b) for the channel.
sidewall. In contrast, micro-channels fabricated with parallel (Figure 4.11(c) and (d)) and circular (Figure 4.11(e) and (f)) polarizations presented rougher surfaces as the nanograting planes were not aligned with any of the channel walls as in the case of perpendicular polarization. The smoothest walls were found for perpendicular polarization and laser exposure of pulse energy $E_p = 50 \text{ nJ}$ and scanning speed $v_s = 0.5 \text{ mm/s}$, yielding a bottom surface roughness of $\sim 200 \text{ nm (rms)}$ based on $50 \mu\text{m} \times 50 \mu\text{m}$ area) in Figure 4.12(c), and a sidewall surface roughness of $\sim 10 \text{ nm (rms)}$ based on $3 \mu\text{m} \times 3 \mu\text{m}$ area) in Figure 4.12(d). The limited viewing area ($3 \mu\text{m} \times 3 \mu\text{m}$) available for the sidewall may suggest a larger overall wall roughness, however, the AFM image encompasses two laser tracks that were very precisely positioned (2 nm resolution) by the motion stages. The $20\times$ smoother sidewalls are attributed to the parallel alignment of nanogratings which otherwise form orthogonally to the bottom/top channel wall and lead to corrugation of $\sim 200 \text{ nm}$ period that matches the $180 \text{ nm}$ period that was found above for the bulk nanograting. This highly smooth sidewall with near optical quality promises the integration of optical waveguides crossing the micro-channel with relatively
Figure 4.12: SEM images of the (a) bottom and (b) sidewall of an etched micro-channel laser written with perpendicular polarization, $E_p = 50$ nJ, $v_s = 0.5$ mm/s, $d_s = 75$ µm, $d_T = 1$ µm, and $d_L = 1.5$ µm and the corresponding AFM images (c) and (d), respectively.

low scattering loss from each sidewall facet.

The SEM and AFM assessment for smoothest channel walls favoured the lowest laser pulse energy ($E_p = 50$ nJ) at the threshold of HF etching where one expects the lowest stress accumulation together with the fewest defects in generating parallel nanogratings. Further, this low exposure level may facilitate a smooth extension of nanogratings into new exposure zones as the laser scans forward as reported by Taylor et al. [117]. A slightly higher exposure of $75$ nJ $\leq E_p \leq 100$ nJ was used hereafter to ensure the rectangular channels would be completely opened at this above threshold exposure with only slightly less smooth sidewalls and without cracking over long channel lengths.

In brief, a rapid prototyping technique in cross-section shaping was presented that permits flexible and arbitrary patterning by merely changing the laser scanning path-
way to form the desired cross-sectional shape while also reducing tapering along the micro-channel length in otherwise single laser scanning modification tracks.

4.1.1.4 Micro-Channel Length

The multi-scan arrays (Section 4.1.1.3) assist in holding the shape of the resulting open channel over long lengths unlike the distortions and tapering effects reported previously for single track lines [12,13,17,102,112]. However, the HF etching was limited as in prior reports [4,14,17] to producing micro-channels of millimeters in length. Figure 4.13(a) shows a schematic of a buried rectangular micro-channel where vertical holes have been introduced to open to the channel periodically to the glass surface to facilitate HF penetration over unlimited channel length. Here, $\theta_T$ indicates a tapering angle anticipated for a buried channel. Figures 4.13(b) to (e) are the top view microscope images of a section of rectangular micro-channels of $8\,\mu\text{m} \times 26\,\mu\text{m}$ cross-sectional area formed over $30\,\text{mm}$ length and have $\sim 7\,\mu\text{m}$ diameter vertical holes positioned periodically with $200\,\mu\text{m}$ (Figures 4.13(b) and 4.13(c)) and $100\,\mu\text{m}$ (Figures 4.13(d) and 4.13(e)) separations. The channel was written at a depth of $d_s = 75\,\mu\text{m}$ using a pulse energy of $E_p = 100\,\text{nJ}$ and etched for 3 hours in $5\%$ dilute HF.

For the $200\,\mu\text{m}$ separation of access holes, a slight tapering angle of $\theta_T = \sim 2^\circ$ is noted in Figure 4.13(b), while no taper or width variation is discernible by the optical microscope within the $< 1\,\mu\text{m}$ resolution for the $100\,\mu\text{m}$ hole separation case shown in Figure 4.13(d). Such access holes are clearly advantageous for unlimited extension of the micro-channel length while only covering an insignificant $0.6\%$ of the wall surface area with occasional access ports. Such holes can easily be sealed on the glass surface, and further investigation ensuring minimal disturbance of the flow dynamics that would otherwise limit potential microfluidic applications is presented in Section 5.3. In many applications, opened holes would effectively act as sealed due to strong surface tension of...
Figure 4.13: Schematic of a micro-channel (a) with vertical access holes, and the top view microscope images of completely etched (5% HF after 3 hours) multi-scanned tracks, laser written with perpendicular polarization at $E_p = 100$ nJ and $d_s = 75 \mu m$ with vertical holes laser written at $E_p = 100$ nJ and separations of (b),(c) 200 $\mu m$ and (d),(e) 100 $\mu m$. Imaging was aligned with the buried channel in (b) and (d) and the top glass surface in (c) and (e).
Figure 4.14: Schematic of a reservoir volume (dimensions $L \times W \times H$) laser-scanned and divided into multiple cubes which were to be released by chemical etching.

the liquid reaching the small diameter (7 $\mu$m) hole opening. But the additional capillary forces from numerous access holes may modify the flow dynamics in applications such as autonomous CS and capillary electrophoresis (CE) that need to be tested.

4.1.2 Reservoir

Reservoirs are essential structures in LOC devices to provide a volume for waste storage or a supply of liquids or analytes for the micro-channels. Since the reservoir is one of the largest structures, and usually multiple reservoirs are required on a single device, fabrication of this structure could be very time consuming if the full volume were required to be laser scanned in glass substrates. This section describes the use of the FLICE technique for the fabrication of open-top reservoirs by drawing from the comprehensive study results of the differential etch rates found over optimized laser exposure conditions as presented in Section 4.1.1.

In an effort to reduce the exposure time, an unique scanning method was developed as shown in the schematic illustration of Figure 4.14. The reservoir was divided into 100-$\mu$m
Figure 4.15: Microscope images of a reservoir having dimensions of 0.5 mm × 0.5 mm × 0.2 mm (a) after laser scanning and (b) after chemical etching for 3 hours in 5% HF.

cubes and double layered arrays of scanning tracks were written along the cube walls at a pulse energy of $E_p = 125$ nJ and a scanning speed of $v_s = 1$ mm/s with layer-to-layer spacing of $d_L = 5$ µm and transverse line-to-line spacing of $d_T = 2$ µm. The double layer improved the etching completion in comparison with etching a single array of laser tracks at the cube walls. Figure 4.15 shows the microscope images of the laser scanned reservoir prior to etching (Figure 4.15(a)) and after chemical etching in 5% HF for 3 hours (Figure 4.15(b)). The release of the laser patterned cubes during HF chemical etching opened the full reservoir volume with the advantage of reducing the scanning time ∼7-fold (for a 0.5 mm × 0.5 mm × 0.2 mm reservoir) or higher (for larger reservoir) in contrast with multi-line scanning of the entire reservoir volume. The large laser-track spacings produced higher wall roughness and corrugations that typically are not as significant for the reservoir in contrast with the narrow micro-channels. Nonetheless, this efficient fabrication method of reservoirs with rougher walls will serve the purpose of sample and waste storage volumes without affecting the micro-channel flow dynamics.
4.1.3 Inverted-Woodpile and Photonic Crystal Structures

In previous sections, FLICE has been developed to harness the nanogratings that readily form in fused silica for highly flexible writing of micro-channels that in turn can be integrated with laser-formed photonic structures to underpin new approaches in 3-D optofluidic microsystems. Previous studies (Section 4.1.1) focused on writing parallel channel arrays to form a cross-sectional shaped micro-channel, but has yet to address the 3-D extension of interlacing micron-sized capillaries to form stressfree and high-density meshes.

Laser exposures were optimized for the writing of small sized and orthogonally connected open microcapillary (single-track micro-channel) arrays, forming a high-density stable network in the bulk fused silica. The woodpile (WP) structure was chosen for the network template as a well-known and desired example in 3-D PC studies owing to favourable templating structure for generating 3-D stopbands. A highly uniform 3-D inverted woodpile (IWP) micro-channel array with diamond-like symmetry is presented for the first time by the 3-D direct-write and HF etching method in fused silica. This prototyping technique provides a rapid means to structure a robust 3-D structure directly in fused silica, which also permits ease of integration with networks of embedded micro-channels fabricated with the same technique. Moreover, such robust 3-D IWP structure integrated in a micro-channel can offer a capillary electrochromatography (CEC) capability on chip that permits differential partitioning and electrophoretic migration of the solutes under applied electric field.

In liquid chromatography, the standard approach relies on packed particles for separation. The particle size of a packing is important in determining the column efficiency and limiting the pressure drop. The majority of packing materials used today are porous particles with average diameters between 2 to 30 µm and are typically a silica-based or alumina-based material with pore sizes ranging from tens to hundreds of nanometers. For LOC, packing of particles in micro-channels often encounter challenges such as non-
uniform packing and affixing the particles in the channels [118]. For use of monoliths as the stationary phase, the monolith structures are simple to chemically bond onto micro-channels and are more porous and more permeable than packed beds, offering the advantage of lower pressure for separation. However, monolith formation is a multi-step process that involves monolith polymerization under UV irradiation and flushing out of non-polymerized monomers in a pre-fabricated microfluidic platform [119]. Uniform array of pillars with a mean diameter of 4.45 to 4.68 μm and a centre-to-centre separation from 5.93 to 8.22 μm have been fabricated by deep reactive ion etching (DRIE) for separation to offer a reproducible method to pack uniform structures [120]. However, DRIE is time consuming and limited only to patterning of surface structures. A highly uniform structure directly embedded in a micro-channel is desirable for on-chip CEC.

Extending the procedure outlined in Section 4.1.1, the laser beam was focused with a 0.55 NA aspherical lens to a ~1 μm focal spot diameter, or with a 0.90 NA objective lens to a ~0.6 μm focal spot at positions varying from the surface to a depth of 60 μm or 30 μm, respectively. This near surface exposure minimized the spherical aberration and provided direct access of HF to etch the embedded structures. Although linear polarization aligned perpendicular with the laser scanning direction provides smoother micro-channel sidewalls of ~10 nm rms roughness (Section 4.1.1), circular polarization was selected to randomize the orientation of the nanogratings and facilitate uniform HF etching of orthogonal micro-channels to yield an anticipated wall roughness of <200 nm rms. A sample scanning speed of 0.5 mm/s provided a high etch rate of ~5 μm/min. The laser tracks were scanned laterally, layer-by-layer, from the bottom to top as shown schematically in Figure 4.16 to form a WP pattern with a period, a, in the x and y transverse directions and vertical period, c, tested in a range from 2 to 7 μm and 2 to 14 μm, respectively, for the 0.55 NA lens, and 2 to 4 μm and 2 to 5 μm, respectively, for the 0.90 NA lens.
Figure 4.16: Schematic illustration of the 3-D laser scanning of the WP structure in fused silica glass.

The effects of pulse energy, lens NA and focal depth were explored to form closely packed, isolated, and very small diameter channels (capillaries) that become partially opened to their neighbouring crossed channels while retaining a rigid and stable WP structure. A comprehensive mapping of minimum channel sizes and periodicities was completed for both the 0.55 NA and 0.90 NA lenses in laser exposure windows of 25 to 75 nJ pulse energy ($E_p$) and 30 to 37 nJ, respectively.

After laser patterning, the sample was immersed in 5% dilute HF for 10 to 30 minutes without ultrasonic agitation to avoid destroying the delicate structure. The fabricated structures were cleaved and viewed with a tabletop SEM (Hitachi TM-1000) to reveal the top view and open WP cross-sections.

Stable WP structures with completely isolated microcapillaries were found to form for capillary array periods as small as $a = 5 \, \mu m$ and $c = 14 \, \mu m$ with the 0.55 NA lens as shown in Figure 4.17. The SEM top and cross-section views (Figure 4.17) show eight
Figure 4.17: Cross-sectional SEM views of the etched WP structures fabricated with a 0.55-NA lens and pulse energies of (a) 40 nJ, (b) 45 nJ, (c) 50 nJ, (d) 55 nJ, (e) 60 nJ, (f) 65 nJ, (g) 70 nJ, and (h) 75 nJ.
representative examples of etched WP structures written with uniform pulse energy that was applied over the 25 to 75 nJ range in 5 nJ steps. Examining the energy-depth dependence, one finds the width and height of individual micro-channels are significantly non-uniform, increasing linearly with increasing pulse energy and decreasing depth position. Figure 4.18 plots the width (Figure 4.18(a)) and height (Figure 4.18(b)) of holes against the layer depth at representative pulse energies of $E_p = 40$ to 75 nJ in 5 nJ steps, revealing a linear dependence of the hole sizes with the layer depth. Hole sizes were also found to increase linearly with the pulse energy. Incomplete etching is noted at deeper positions for lower energy exposure, while over-etching was observed near the surface at energies $E_p > 80$ nJ. The lower etch rate of the deeper structures arises from a combination of spherical aberration that increases the laser spot size (reduced intensity) and a short 30 minutes etching time that is insufficient for acid to penetrate to the deeper layered exposure tracks. In contrast, prompt HF exposure to the topmost laser tracks led to over-etching at higher pulse energy.

Similar trends were observed for WP structures formed with the 0.90-NA lens. However, more densely packed microcapillaries were possible to $a = 2 \ \mu m$ and $c = 5 \ \mu m$ periodicity as shown in Figure 4.19. Here, a low $E_p = 33$ nJ exposure yielded capillaries having widths and heights of less than 1 $\mu m$ and 3 $\mu m$, respectively, after 10 minutes of 5% HF etching. However, densely packed IWP structure could not be written deeper than 30 $\mu m$ due to spherical aberration, which blurred the focus to underexpose deeper structures. Therefore, the 0.55 NA lens was preferred to form uniform IWP structures deeper into the glass (>30 $\mu m$).

To compensate for the slower or delayed etching of deeper modification tracks for the case of the 0.55 NA lens, a procedure for increasing laser exposure in deeper layers was developed with the objective of forming $\sim 2 \ \mu m$ wide capillaries in the tightest possible
Figure 4.18: The observed (a) width and (b) height of capillaries formed at various depths in fused silica with laser pulse energies from 40 to 75 nJ in 5 nJ steps and 30 minutes of etching in 5% HF solution. The dashed curves indicated the energy compensated exposure for a uniform WP.
Figure 4.19: SEM images showing the (a) top view and (b) cross-section view of the IWP structure, laser written with circular polarization at $E_p = 33$ nJ and $v_s = 0.5$ mm/s using a 0.90-NA lens, following 10 minutes of etching in 5% HF solution.
Figure 4.20: SEM images showing the (a) top view and (b) cross-section view of the highly uniform IWP structure in fused silica glass, laser written at $v_s = 0.5 \text{ mm/s}$ with circular polarization and energy decreasing linearly from 87-nJ at 60 $\mu\text{m}$ scanning depth to 63-nJ at the top layer, using a 0.55-NA lens and following 30 minutes of etching in 5% HF solution.

packing geometry of $a = 5 \mu\text{m}$ and $c = 14 \mu\text{m}$ found above. Various linear and logarithmic representations of the depth dependent energy data from Figure 4.18 were applied to find an optimum exposure recipe. The most uniform IWP structure was formed with energy decreasing linearly from 87 nJ at 60 $\mu\text{m}$ scanning depth to 63 nJ at the first layer on the surface. SEM images of the top view (Figure 4.20(a)) and the 100 $\mu\text{m} \times 60 \mu\text{m}$ cross-section (Figure 4.20(b)) of the 5-mm long IWP channel reveal a highly uniform structure. The observed hole width, plotted along the horizontal dashed line in Figure 4.18(a), is seen to decrease only slightly from $2.31 \pm 0.07 \mu\text{m}$ to $2.09 \pm 0.10 \mu\text{m}$ as the depth increased to 60 $\mu\text{m}$. Hence, pulse energy compensation is a promising direction to follow in the fabrication of uniform periodic structures over varying depth, but limited here to $\sim$90 $\mu\text{m}$ due to saturation of etch rates at $E_p = 100 \text{nJ}$ (Figure 4.5).

The differing slopes in Figures 4.18(a) and 4.18(b) manifest as a varying aspect ratio of the capillary cross-sections with increasing depth, arising from spherical aberration and HF etching dynamic effects that have been partly compensated with the present depth-
corrected exposure (dashed curves). However, further balancing of both the hole width and height is a remaining challenge that may be augmented with methods of high-NA oil immersion \[111\], astigmatic focusing \[110\], asymmetric focusing \[109\], or spherical aberration correction \[102\] in order to fabricate a highly uniform IWP structure with consistent motif and filling fraction over large depth. Although oil immersion lenses and objective lenses with correction collar would provide aberration corrected structures at higher depth, they were not tested due to the potential for laser burning of oil at the surface and automation of dry lens collar position for varying depth could not be immediately implemented. Moreover, the two approaches to control and correct the elliptical capillary cross-section to obtain an ideal IWP structure by using astigmatic beam focusing or inserting a slit before the focusing lens greatly reduce the cross-section aspect ratio, but at the expense of sacrificing spatial resolution for closely packed tracks. Alternatively, much higher packing density may be possible if laser methods to etch nanofluidic channels \[117\] could be extended to form such delicate 3-D structures.

The uniformity of the IWP structure was assessed optically and found to cause significant optical scattering. When the IWP structure was filled with isopropanol, four first-order diffraction beams were observed at the expected \(\sim 6.0^\circ\) diffraction angle for a 532-nm laser beam probing in the \(\Gamma - Z\) direction (vertical in Figure 4.20(b)). A \(\Gamma - Z\) stop-band expected at 30 \(\mu\)m lies within the IR absorption bands of fused silica, and further efforts are required to scale down the size of the microcapillaries to submicron diameters with a closer spacing of below \(\sim 0.7\) \(\mu\)m (for first order stopband) or \(\sim 1.5\) \(\mu\)m (for second order stopband) to achieve a stopband below 2 \(\mu\)m wavelength where fused silica is transparent.
4.2 Visible Waveguide Fabrication

In Section 4.1, nanograting orientation controlled by the laser-writing polarization was exploited for writing buried micro-channel, reservoir and IWP structure. The integration of low insertion loss optical waveguides may serve as a device for probing analytes or forming a new platform for 3-D optofluidic microsystems. To integrate buried optical waveguides within the microfluidic substrate that would be single mode and low loss in the visible spectrum, laser exposure conditions were tuned from those reported previously for IR guiding conditions reported by Eaton and coworkers [114, 121]. Since the buried optical waveguides would be integrated directly with the micro-channels during the DLW step before chemical etching, the range of laser exposures applied here to write optical waveguides had the nanograttings aligned to be perpendicular with respect to the scanning direction when the laser polarization (E-vector) was parallel. Such perpendicular nanograting alignment served as strong chemical stop-layers, as demonstrated with the high polarization etching contrast (Section 4.1.1) between parallel and perpendicular polarizations, thus hindering the etching of such laser-formed waveguides during the etching step.

For waveguide writing, the laser beam was focused inside the fused silica with an 0.55-NA aspherical lens in a range of 75 to 150 µm depth from the surface. It was necessary to keep the pulse energy at the surface below the ablation threshold. However, the focusing beam created an intensity inside the sample to locally modify the refractive index at exposures found to be optimized between the smooth modification and the nanograting formation regimes presented in Section 2.5.1.1. A range of $E_p = 25$ to 125 nJ pulse energies was focused into the fused silica for waveguide writing. A range of constant scanning speed from $v_s = 0.2$ to 5.0 mm/s was also tested to generate 25-mm long uniform lines of refractive index modification with sufficient nanograting formation for hindering chemical etching to avoid high scattering losses by the guided light. The waveguide containing glass substrate was polished at the end facets after laser fabrication to obtain a lower
insertion loss. The following procedure outlines the polishing process by hand. The sample was first ground using 320 and 400 grit size silicon carbide papers with a drop of water to remove \( \sim 400 \, \mu \text{m} \) on each facet, followed by finely grinding another \( \sim 20 \, \mu \text{m} \) off each facet with 500 and 600 grit size papers. The final polishing using 1200, 2400, and 4000 grit size papers in sequential order provided a high quality optical surface at the end facets.

The near-field modal profile of light emerging from a buried waveguide can be used to obtain information about the refractive index modification. Hence, a diagnostic station placed on top of a floating optical bench for vibration control was constructed specifically for waveguide characterization as shown in Figure 4.21. The single mode optical fiber (SMF-28) was mounted onto a 3-axis stage (Luminos I3000) with 100-nm precision and used to couple the red laser source (Thorlabs Inc. S1FC635) of 635-nm wavelength into the optical fiber. The waveguide sample was placed on a 5-axis mechanical alignment stage (Luminos I5000). Together, both stages enabled effective alignment of the fiber source into a desired waveguide of the sample. A CCD camera (Sony XCD-V50) equipped with a 0.75 to 3\( \times \) zoom lens (Edmund VZM 300i) was connected to a computer and was positioned on top of the diagnostic setup to aid in alignment through the captured images or videos observed on a monitor. Refractive index matching oil (Cargille; \( n = 1.4420 \) at 589.3 nm) was applied to fill the air gap between coupling fiber with the waveguide surface. The near-field modal profile was captured through a 60\( \times \) aspherical lens which imaged the output mode shape onto a Spiricon CCD, and a calibration factor was used to correct the difference between the CCD pixel size and the true image size through the Spiricon software. A schematic of the diagnostic setup is shown in Figure 4.22(a).

For insertion loss measurement, the diagnostic setup was similar to the mode-profiling
Figure 4.21: Photograph of the waveguide diagnostic station showing the input and output fibers aligned with a sample positioned on mechanical stages (Luminos). The overhead CCD camera system connected to a computer was used for verifying visual alignment of optical fibers to the sample waveguide.
Figure 4.22: Schematic illustrations of the diagnostic setup used to launch a laser source into a waveguide under test inside the sample to (a) capture the near-field modal profile exiting the waveguide, or to (b) measure the insertion loss of the waveguide.
setup, but the CCD camera with the aspherical lens (mode profile assembly in Figure 4.22(a)) was replaced with a SMF-28 fiber butt-coupled to the waveguide sample and connected to an optical power detector (Newport 818SL) with a multi-function optical meter (Newport 1835-C) for observing the output. Figure 4.22(b) illustrates the diagnostic setup for the insertion loss measurement. The power output from the waveguide sample was compared to a reference output acquired by coupling the input and output fibers closely together with index matching oil filling the gap.

One of the largest loss components in the insertion loss is the waveguide propagation loss. The waveguide propagation loss can be measured in many ways. The method employed here used the diagnostic setup shown in Figure 4.22 to find the best input optical coupling through the waveguide under test. The overhead CCD camera system viewing the waveguides from the top was used both for aiding the alignment process and for capturing the gray-scale images of scattered waveguide light. Waveguide propagation loss was inferred from the exponential decrease in this scattering light with distance along the waveguide.

The waveguides written at pulse energies of \( E_p > 50 \, \text{nJ} \) were found to be multi-mode when formed at depths of 75 \( \mu \text{m} \) and 150 \( \mu \text{m} \), while \( E_p = 25 \, \text{nJ} \) exposure at scanning speeds of \( 0.2 \, \text{mm/s} \leq v_s \leq 5 \, \text{mm/s} \) generated faint modification lines that did not offer optical guiding. However, the waveguides written with \( E_p = 50 \, \text{nJ} \) provided single mode guiding for all the investigated scanning speeds from 0.2 to 5 mm/s and depths of 75 \( \mu \text{m} \) and 150 \( \mu \text{m} \). At the fastest scanning speed of \( v_s = 5 \, \text{mm/s} \), the insertion loss was too large to obtain modal profiles for analysis. Several modal profiles are shown in Figure 4.23 in 635 nm with waveguides written at \( E_p = 50 \, \text{nJ} \), \( d_s = 75 \, \mu \text{m} \) and \( v_s \) from 0.2 to 1.0 mm/s. The mode field diameter (\( \frac{1}{e^2} \) intensity) were determined by the Spiricon software to be \( \sim 7 \, \mu \text{m} \) in red.

The insertion losses of the buried waveguides were obtained from measuring the differ-
Figure 4.23: Near-field modal profiles of buried waveguides written in fused silica glass with pulse energy of 50 nJ, depth of 75 µm, an aspherical lens with 0.55 NA, laser polarization parallel to the scanning direction, and scanning speeds of (a) 0.2 mm/s, (b) 0.5 mm/s and (c) 1.0 mm/s in red (635 nm).

Figure 4.24: A CCD image in gray-scale illustrating the exponential decrease of the scattering light (right to left) from a waveguide due to propagation loss at a wavelength of 635 nm.

ence in transmitted power through the fiber-to-fiber coupling reference and the waveguide for fiber butt coupling. At a depth of $d_s = 75$ µm, the lowest insertion loss of 12.0 dB was obtained with the scanning speed of $v_s = 0.2$ mm/s, which increased to a loss of 17.6 dB at the scanning speed of 1.0 mm/s for a 25.4 mm waveguide sample.

Observation of the scattered light intensity from the waveguide provided an alternate estimation of the propagation loss. Figure 4.24 shows an image of a waveguide scattering 635-nm guiding light for measurement of the waveguide propagation loss. Buried waveguides written with a scanning speed of 0.2 mm/s, 0.5 mm/s and 1.0 mm/s had propagation loss values of approximately 1.1, 1.2 and 1.2 dB/cm for 635 nm, respec-
The waveguide \( (v_s = 0.2 \text{ mm/s}) \) with the lowest propagation loss of 1.1 dB/cm at 635 nm was also tested for 522 nm, and a value of 5.8 dB/cm was obtained. Figure 4.25 shows two representative semi-log graphs of the scattered light intensity plotted along the 2.3 cm long segment of waveguides, where the slopes of the data yielded a loss estimate for the fabricated waveguides.

The \( \frac{1}{r^2} \) distances of the near-field modal profiles were used to estimate the refractive index modification profile change of the single-mode buried waveguides with the RSoft simulation program (RSoft Photonics CAD). With a step-index cylindrical model having a diameter of 2 µm, which was chosen to be the same as the width of the laser modification line, a refractive index contrast of \( \Delta n = \sim 4 \times 10^{-3} \) was inferred from the transverse near-field mode distance of 7 µm by the simulation. This refractive index contrast value is in the same order of magnitude as the results from the fabricated buried waveguides for 1550 nm wavelength guiding as reported in previous studies by Shane Eaton [114] for fused silica glass. For the waveguides written with higher pulse energies \( (E_p > 50 \text{ nJ}) \) with multi-mode, the refractive index change were higher \( (\delta n > \sim 4 \times 10^{-3}) \), which are as expected [114].

The fs-laser written buried waveguides for guiding IR light [114] and visible light [15] were previously demonstrated by various groups. The present results for near-field modal profile, insertion loss, propagation loss and refractive index contrast extend this to form waveguides guiding in the visible spectrum with a high repetition rate (1 MHz) ultrashort pulsed (220 fs) laser here with faster scanning speed of 0.2 mm/s and lower pulse energy of 50 nJ versus the 1 kHz and 150 fs pulses [15] previously studied at exposures of 4 µJ and a scanning speed of 0.02 mm/s. For fabricating single-mode waveguides, the lowest propagation loss of 1.1 dB/cm was observed here with a laser scanning speed of 0.2 mm/s and a pulse energy of 50 nJ at 635 nm guiding. The results for insertion loss indicated
Figure 4.25: Semi-log plot of scattered intensity as a function of the distance from the end facet of waveguides formed at scanning speeds of (a) 0.2 mm/s and (b) 0.5 mm/s in bulk fused silica with $E_p = 50$ nJ and parallel laser polarization. The slope of the least squares fit of the data (solid line) provides a propagation loss estimate of (a) 1.1 dB/cm and (b) 1.2 dB/cm.
Figure 4.26: Top view optical images of waveguides written in fused silica at a depth of 75 µm, pulse energy of 50 nJ and scanning speeds of (a) 0.2 mm/s, (b) 0.5 mm/s and (c) 1.0 mm/s with a 0.55-NA lens.

A weak dependence on laser scanning speed. Figure 4.26 shows the top view microscope images of the waveguides written with various speeds at $d_s = 75$ µm and $E_p = 50$ nJ. As the scanning speed increased, more imperfections were observed in the laser modification tracks which contributed to the higher insertion loss found at higher writing speeds. The propagation losses of these fs-laser written waveguides of approximately 1.1 dB/cm at 635 nm are very promising for integration in LOC in comparison with losses of 1.4 dB/cm at 633 nm reported in SU-8 polymer waveguides [45] and 1.1 dB/cm at 633 nm as reported for liquid core waveguides [37].

For guiding at a shorter wavelength of 522 nm, the waveguide propagation loss of 5.8 dB/cm reported here is comparable to or better than waveguides fabricated with SU-8 polymer (~6 dB/cm at 522 nm [45]) and liquid core (8.2 dB/cm at 532 nm [37]), but remains significantly higher than the 0.9 dB/cm loss at 543 nm demonstrated by Osellame and coworkers in fused silica written at $v_s = 0.02$ mm/s and $E_p = 4$ µJ [15]. The significant increase of waveguide propagation loss from 1.1 dB/cm at 635 nm to 5.8 dB/cm at 522 nm can be attributed to stronger Rayleigh scattering loss ($\lambda^{-4}$ scaling).
as well as the secondary Bragg stop band identified at 540 nm that in [122] was attributed to the nanogratings formed perpendicular to the scanning direction. Further improvement in propagation losses in the visible spectrum will require further optimization of the laser repetition rate, pulse duration, pulse energy, polarization, scanning speed, and the laser wavelength.
Chapter 5

Biophotonic Integrated Devices

The previous chapter presented a multi-line laser scanning procedure for controlling the cross-section shape of embedded channels and further introduced periodic access ports to extend such channels over long lengths that underpin flexible formation of 3-D LOCs of any shape and size in fused silica. A novel laser scanning method to significantly reduce the reservoir fabrication time and DLW of optical waveguides for guiding visible light were also presented to complete the set of building blocks essential for creating fully functional biophotonic LOCs. In this chapter, the novel attributes of direct femtosecond laser writing together with selective chemical etching in fused silica are extended to the fabrication of 3-D microfluidic system that demonstrate the basic LOC functions of electrokinetic and capillary forces. In addition to extending the chip layout to make full use of all three spatial dimensions that greatly improves the flexibility of LOC devices, the ability to integrate optical waveguides and other optical components is also a significant objective.

In the following sections, the general experimental procedures together with three LOC devices are presented. Section 5.1 presents the preparation and experimental procedures for the devices. Section 5.2 takes advantage of the nanograting orientation for selective etching to combine buried micro-channels with optical waveguides for probing
analytes. Section 5.3 describes a 3-D passive particle counting device by writing channels that retain strong capillary force together with accurately formed reservoirs and stop-valves, enabling precisely calibrated particle counting. Section 5.4 furthers the LOC design to create under-passing (U) and over-passing (O) channel networks for parallel channel capillary electrophoresis (CE) separation in a multi-level microfluidic device. These microsystems demonstrate the utility of the precise laser patterning of high density 3-D microfluidic and optical structures when followed by chemical etching to obtain highly functional LOCs.

5.1 Experimental

The following experimental sections (Sections 5.1.1, 5.1.2 and 5.1.3) describe the general experimental procedures used for fabricating waveguide probing device (Section 5.2), particle counting device (Section 5.3) and multi-level capillary electrophoresis device (Section 5.4).

5.1.1 Preparation of Buffer, Fluorescent, Fluorosphere, and Etching Solutions

The buffer used for electrophoresis was prepared by dissolving 40.24 mg of sodium tetraborate (Sigma-Aldrich) in 10 mL of type I reagent-grade water to produce 0.020 M sodium tetraborate solution with a pH of 9.15. The dye solutions used for electrophoresis and waveguide excitation were prepared by dissolving 3.8 mg of rhodamine 123 (Rh 123) (Sigma-Aldrich) and 4.8 mg of rhodamine B (Rh B) (Fluka) powders each in 10 mL of type I reagent-grade water to produce 0.001 M solutions of Rh 123 and Rh B, respectively. Rh B is neutral at a pH of 9.0 because of the presence of the carboxyl group (COOH), while Rh 123 has this functional group esterified and therefore carries a positive charge. All solutions were filtered with Acrodisc® syringe filter with
0.2 \textmu m Supor\textregistered membrane from Pall Corporation. For particle counting, stock solution of fluorescent polystyrene beads conjugated with pycoerythrin (Flow-Count\textsuperscript{TM} Fluorospheres) with a nominal diameter of 10 \textmu m was used undiluted, and verified to contain 987 beads/\textmu mL with the BD FACSCalibur flow cytometry system. The fluorosphere solution was mechanically shaken and allowed to settle for 10 minutes before application. The glass etching solution was a 5\% dilute HF solution prepared by mixing 10 mL of ACS grade HF (48.0–51.0\%) with 90 mL of type I reagent-grade water at room temperature.

5.1.2 Preparation of SU-8, PMMA and PDMS for Device Sealing

Glass coverslip (Fisherfinest Premium) of dimensions 25 mm \times 25 mm with a thickness of 0.13 to 0.17 mm was pre-scribed \sim 2 mm from each of the four edges on bottom surface prior to spin-coating with either epoxy-type photoresist (MicroChem SU-8 2050) or PMMA (MicroChem 2200 PMMA A 11). With a similar viscosity, \sim 1 mL of SU-8 or PMMA was spin-coated onto the top surface of the prescribed coverslip using spin cycles of 500 rpm for 5 seconds with 100 rpm/s\textsuperscript{2} acceleration, followed by 3000 rpm for 50 seconds with 324 rpm/s\textsuperscript{2} acceleration. Following the spin-coating of the 35 to 45 \textmu m thick polymer layer, the pre-scribed coverslip was cleaved at the previously scribed lines at all four edges to provide a uniform coating thickness for reliable sealing to laser-fabricated chips. After spin-coating the SU-8 or the PMMA, the coated coverslip was immediately brought in contact with the laser patterned and chemically etched glass substrate. After contacting, The PMMA sealed device was then baked at 180 \degree C for 2 minutes to first harden and prevent the PMMA to flow into the micro-channels through the filled vertical access ports. For complete curing of SU-8 or PMMA the sealed device was illuminated with a UV spot curing system (Exfo Novacure N2001-A1) at a wavelength of 365 nm for 10 minutes with 4 W of power. This coverslip method enabled sealing of the vertical access ports, but the possibility of the polymer or epoxy resin inflow prior to curing
and the autofluorescence of SU-8 had prompted an alternate sealing method by using pre-cured sheet of PDMS.

For the sealing with PDMS, a 3.2-mm thick PDMS sheet was prepared by curing a 1:10 mixture of 4 g of Sylgard® 184 silicone elastomer curing agent and 40 g of base (Dow Corning) in a 15 cm diameter circular petri dish. The mixture was stirred with a glass rod for 10 minutes and was degassed for 1 hour at 100 kPa vacuum prior to curing in an oven for 1 hour at 60 °C. Different sized PDMS sheets were used for sealing the waveguide device and electrophoresis device. For reversible sealing, the PDMS sheet was contacted with the device surface. For permanent sealing, the PDMS sheet was oxygen-plasma treated to form an irreversible bond upon contact with the device surface.

5.1.3 Laser Patterning and Chemical Etching

For all the LOC devices to be demonstrated, a 40× aspherical lens of 0.55 NA was used to focus the 522 nm laser, operated at 1 MHz, into the transparent fused silica at depths varying from the sample surface of \( d_s = 0 \) to 140 µm. Laser modification tracks were written with a laser polarization perpendicular to the scan direction or a circularly polarized beam for shaping the micro-channels and reservoirs, and parallel polarization for scanning waveguides. The scanning speed range of \( v_s = 0.5 \) to 1.0 mm/s was used for scanning microfluidic structures and waveguides.

The laser exposure recipes to pattern the waveguide probing device, particle counting device and multi-level electrophoresis device were based on multi-line laser scanning procedures developed in Section 4.1.1 to minimize crack formation and offer smooth surfaces of ~10 nm (rms) and ~200 nm (rms) roughness respectively for the sidewalls and upper/lower surfaces. However, the assembly of such channels into close-packed 3-D microfluidic networks resulted in significant stress accumulation at channel crossings and corners that induced fractures and cracks. Minimal vertical and lateral offsets of 30 µm and 50 µm, respectively, were found necessary to be applied between neighbouring
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channels to mitigate the formation of such high stresses.

5.1.3.1 Micro-Channel Fabrication

Straight, curved and tapered micro-channels were all written by multi-line laser scanning with a speed of \( v_s = 0.5 \text{ mm/s} \), layer-to-layer spacing of \( d_L = 2 \mu\text{m} \), transverse line-to-line spacing of \( d_T = 1.5 \mu\text{m} \), and a pulse energy of \( E_p = 75 \text{ nJ} \). The average etching rate was \( \sim 5 \mu\text{m/min} \) in 5% dilute HF for both perpendicular and circular polarizations. Straight and curved micro-channels were written with perpendicular polarization and circular polarization, respectively. Perpendicular polarization aligned nanogratings parallel with the channel sidewalls to provide the smoothest sidewalls for straight channels, whereas the formation of randomly oriented nanogratings with circular polarization was preferred for writing curved channels due to the isotropic etching possible in any scanning direction.

5.1.3.2 Vertical Access Ports

To facilitate chemical etching along unlimited lengths of the embedded channels, arrays of vertical access ports with 8.3 \( \mu\text{m} \) transverse spacing for the particle counting device and electrophoresis device, and a single vertical port array for waveguide probing device, were positioned periodically at 100 \( \mu\text{m} \) spacing along all channels. The vertical ports were exposed with a double overlapping collinear exposure scan of \( v_s = 1.0 \text{ mm/s} \) speed and \( E_p = 100 \text{ nJ} \) pulse energy.

5.1.3.3 Reservoir Fabrication

The reservoirs for each device were of different size. All reservoirs were defined by the method of dividing the volume into 100-\( \mu\text{m} \) cubes. The walls of the cubes were defined by scanning double-layered arrays of laser tracks with layer-to-layer spacing of \( d_L = 5 \mu\text{m} \) and line-to-spacing of \( d_T = 2 \mu\text{m} \) at \( v_s = 1.0 \text{ mm/s} \) speed and \( E_p = 125 \text{ nJ} \) energy, as described in Section 4.1.2. This innovative scanning method reduced the laser exposure
time dramatically by \sim 7\text{-fold} (for a $0.5 \text{ mm} \times 0.5 \text{ mm} \times 0.2 \text{ mm}$ reservoir) or higher (for larger reservoir) in contrast with multi-line scanning of the entire reservoir volume.

### 5.1.3.4 Chemical Etching

The laser patterned glass plate was submerged in a 5\% dilute HF solution for 4 to 5 hours depending on the complexity of the device design to completely etch out the appropriate laser-modification tracks. This was followed with rinsing in distilled water and drying with $N_2$ gas flow before the device was ready for testing.

### 5.2 Waveguide Probing Device

The waveguide probing device (Figure 5.1) aims to exploit the nanograting orientation to integrate buried optical waveguides within the microfluidic substrate that would be single mode in the visible spectrum. The laser exposures applied here to write optical waveguides and microfluidic channels, the orientation of the nanogratings aligned to be perpendicular (for waveguides) or parallel (for channels) with respect to the scanning direction when the laser polarization (E-vector) was parallel or perpendicular, respectively. From the studies in Section 4.1.1, the high polarization contrast etching ($28:1 = E_\perp:E_\parallel$) was attractive for integrating chemical etching resistant optical waveguides with the fast
Polarization contrast is demonstrated to be highly beneficial in optofluidic integration. A single laser scanning step was applied to form a buried rectangular micro-channel and two open reservoirs with perpendicular polarization and defined nearly optically smooth sidewall surfaces through which an optical waveguide was made to intercept. The G-code program used for patterning this device is presented in Appendix C. For this device, a high purity synthetic fused silica glass plate of dimensions 50.8 mm × 25.4 mm × 1 mm was used. The 25-mm long waveguide was laser written at a depth of $d_s = 75$ µm from the surface, scanning speed of $v_s = 0.2$ mm/s, and $E_p = 50$ nJ pulse energy. Parallel polarization was used to form perpendicular nanogratings that aligned parallel with the channel wall and thus serve as strong chemical stop-layers preventing the etching of the waveguide precisely at the channel wall as shown in the optical microscope image of Figure 5.2. The buried sample flow channel (SFC) of 10 µm × 20 µm rectangular
Figure 5.3: An optical image of a buried optical waveguide (a) guiding a 522 nm light and (b) exciting Rhodamine B fluorescent dye in an embedded micro-channel. (a) The scattered light in the micro-channel is due to the diverged laser light scattering off the channel walls. (b) The vertical and horizontal dashed lines indicate the positions of the channel and waveguide, respectively.

cross-section and 20 mm length was fabricated with \( d_s = 65 \, \mu \text{m} \) and a series of vertical access holes. This buried SFC connects to two waste reservoirs (WRs), with one WR at each end having dimensions of 0.5 mm \( \times \) 0.5 mm \( \times \) 0.2 mm. After laser exposure, the sample was immersed in 5\% dilute HF for 4 hours.

The optical waveguide has a mode field diameter of \( \sim 7.0 \, \mu \text{m} \) and a propagation loss of approximately 1.1 dB/cm in 635 nm (Section 4.2). An average insertion loss as small as 2.00\( \pm \)0.07 dB was measured through a water filled channel. Considering an expected Fresnel loss of 0.02 dB and a mode mismatch loss of \( \sim 1.84 \, \text{dB} \), one infers a scattering loss of approximately 0.07\( \pm \)0.04 dB from each of the sidewall surfaces attesting to their
exceptional wall smoothness.

A fluorescent dye solution of $10^{-3}$ M Rh B fluorescent dye solution diluted to $10^{-5}$ M using type I reagent-grade water was prepared and injected to fill the SFC for probing fluorescence excitation from the crossing waveguide. Figure 5.3(a) shows the optical image of the scattering 522-nm wavelength green light guiding along the optical waveguide from the right side and exciting the Rh B fluorescence at the micro-channel in the SFC. The diverging light from mode-mismatch and uncoupled light in butt-coupling scattered off the micro-channel walls lighting up the SFC. A 532-nm long-pass filter was used to exclude the 522-nm excitation light and observe the emitted fluorescent light in the channel (Figure 5.3(b)). This example serves as a basic demonstration of optofluidic function, and provides evidence that the polarization-dependent etching offers a convenient means of laser patterning to integrate microfluidic and optical components in a single exposure step without chemical etching damage into the optical components. Further, this approach is advantageous compared with post-writing of waveguides [15] into pre-existing microfluidic channels that introduce extra process steps and optical aberrations in the laser focusing. The present method also eliminates the need for a thick stop-layer between the micro-channel and the waveguide [47] that possibly degrades the optical performance of the LOC.

5.3 Particle Counting Device

The particle counting device is an autonomous device that employs capillary force for driving fluid carrying particles. One aims in its design to serve as a particle counting chip in a portable hand-held analyzer that offers the potential for affordable and efficient blood testing. Such a particle counting device must provide a platform for particle flow and an optical reader with an appropriate software to count the fluorescent conjugated particles and displays the results on an LCD screen, all within a hand-held analyzer.
To construct a microfluidic platform for autonomous particle counting that harnesses capillary forces to draw fluid carrying particles, the following concepts were considered in the device design. The fabrication of a single long straight channel for particle counting would be trivial. However, a long microfluidic platform would be required to achieve a relatively large counting volume, which defeats the purpose of a miniaturized LOC. Moreover, the reduction of fluid flow speed in long channel lengths would make particle counting inefficient. Therefore, a design with a straight channel opening to deep reservoirs for waste storage was considered to permit larger volume counting. At the abrupt opening connecting the channel and the waste reservoir, the fluid meniscus must change its curvature in order to achieve the equilibrium contact angle $\theta$. This curvature disappears at an angle $\beta = 90^\circ - \theta$ where the capillary pressure would drop to zero \[125\]. Therefore, the angle between the transition from the fluid carrying channel to the waste reservoirs needs to be less than $90^\circ - \theta$, or less than an angle of $50^\circ$ for fused silica with a contact angle of $\theta = 40^\circ$ in water. In contrast, stop valves are needed to cease the flow of fluids once the waste reservoirs have been filled for accurate particle counting. Therefore, micro-channels can be connected at the end of the reservoirs with an abrupt opening at
an angle of $\beta \geq 90^\circ - \theta$ to flatten the meniscus and pin the filling front to cease the flow.

In this study, the microfluidic substrate for such a particle counting device is presented. A high purity synthetic fused silica glass plate with dimensions of 50.8 mm $\times$ 25.4 mm $\times$ 1 mm was used. Various iterative versions of the particle counting device were formed in fused silica over dimensions of 28.4 mm $\times$ 1 mm $\times$ 0.5 mm with embedded channels and open reservoirs evolving to the device shown schematically in Figure 5.4. The device consisted of six major components that were designed to harness strong capillary forces in the 20 mm $\times$ 0.2 mm $\times$ 0.04 mm SFC written at a depth of $d_s = 70 \ \mu m$. The SFC drew analyte solution from the sample reservoir (SR) of 2 mm $\times$ 1 mm $\times$ 0.5 mm volume at controlled modest flow speed ($\sim$2.5 mm/s) for enabling accurate particle counting. The flow filled into three calibrated WRs, each with dimensions of 2 mm $\times$ 0.2 mm $\times$ 0.5 mm, through a $d_s = 70 \ \mu m$ deep tapered channel (TC) where video imaging for particle counting was recorded in the 40-\(\mu\)m-thick buried flow channel as seen in the inset in Figure 5.4. The particle counting was done at the TC region where more accurate counting was possible due to the larger counting area and a longer flow path for the particles to ensure each particle flowing into the WRs could be recorded and counted. In order to minimize meniscus curvature that otherwise would stop the flow, the side walls of the TC were formed to meet the walls of the SFC and WRs at 45$^\circ$ while the bottom and the top surfaces followed a cylindrical surface with smooth curved radii of 100 \(\mu\)m and 50 \(\mu\)m, respectively. Stop-valve channels (SVCs) of dimensions 2 mm $\times$ 0.025 mm $\times$ 0.017 mm, written at a depth of $d_s = 30 \ \mu m$ and located in the middle of each WR ceased the flow at the terminating reservoir (TR) (2 mm $\times$ 1 mm $\times$ 0.5 mm). During scanning, laser polarization was aligned perpendicular to channel sidewalls to fabricate the smoothest channel walls for strongest capillary flow, and the G-code program used for patterning this device is presented in Appendix A. After laser scanning, the patterned fused silica was immersed in 5\% HF for 5 hours to completely etch out the microfluidic structures.
Before the flow characterization, a 24.4 mm × 10 mm piece of PDMS was removed from a petri dish and promptly contacted to the device surface to form a reversible seal over the WRs and the vertical access ports of all the micro-channel sections (Sections 2 to 6 on Figure 5.4) as shown in Figure 5.5(a). The particle counting device was tested within 24 hours of PDMS sealing.

The particle counting chip was placed in a cell counting analyzer prototype, and 10 µL of the 987 beads/µL fluorosphere solution was loaded into the SR. The solution was drawn through the SFC under capillary force toward the TC and into the WRs. A 532 nm diode laser excited fluorescence from the fluorospheres at the observation window shown schematically in Figure 5.4 (enlarged window) and in the microscope image of Figure 5.5(b). Video recordings of the flow carrying fluorospheres were captured, through an optical filter to remove scattered excitation light, with a CCD video camera at a frame rate of 10 fps. Manual counting was carried out until the TC, WRs and SVCs were fully filled to a combined calibrated volume of 0.612 µL.
Figure 5.6: Microscope images (left) of (a) a partially filled single large waste reservoir (WR) with an air bubble, and (b) a series of fully filled small WRs together with the respective images (right) of fully filled stop-valve channels (SVCs) terminated at terminating reservoirs (TRs).

The first generation particle counting device relied on a single large volume (2 mm × 1 mm × 0.5 mm) WR, where strong adhesion forces drew liquid along the reservoir walls, filling the SVC and stopping the flow prior to complete filling of the WR. This led to the formation of a large bubble as shown in Figure 5.6(a), preventing a calibrated particle count. A modified design of three narrower WRs of dimensions 2 mm × 0.2 mm × 0.5 mm, separated by 0.2 mm thick walls, provided complete filling to the calibrated 0.600-µL volume before SVCs stopped the flow as shown in Figure 5.6(b). Accounting for the TC and
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SVCs volumes, a particle count of 599 beads was made for the total counting volume of 0.612 µL, matching the flow-cytometer calibrated solution particle concentration within 1% error. Automated counting software is currently being developed for this autonomous particle counting chip. The strong capillary forces generated to draw the analyte solution indicates the laser written and chemically etched channels are relatively smooth, and the low counting error is evidence of the precision of this fabrication technique. These qualities are essential and fundamental to all microfluidic devices.

5.4 Multi-Level Capillary Electrophoresis Device

The novel formation of nanogratings in fused silica by femtosecond laser processing has enabled new directions for post selective chemical etching, spurring numerous demonstrations of 3-D patterning that open embedded micro-channels, micro-optics, micro-mechanics, and other micro- or nano-scale structures inside pure fused silica. However, the 3-D potential of this fabrication technique has not been widely explored towards standard LOC applications, especially where fluidic control by capillary forces or electrokinetics in multi-level networks of channels and reservoirs offer new opportunities for compact and highly functional microfluidic systems. To this end, Sections 5.2 and 5.3 demonstrated a waveguide probing device and a particle counting device, respectively, that provided smooth sidewalls for drawing liquids with strong capillary forces and low scattering loss waveguide integration. The multi-level electrophoresis device presented here examine the efficacy of such laser-formed structures for electrokinetic flow and the sources of fluidic disturbances from surface roughness and periodic vertical access ports.

Even though micro-capillary electrophoresis devices require shorter analysis time compared with traditional capillary electrophoresis, micro-capillary electrophoresis intrinsically remains a sequential technique in which the majority of analysis demonstrations
have been on single-channel LOCs offering only one analysis or separation at a time. To enhance analysis throughput, parallel separations are desirable for practical applications, especially for high-throughput analysis such as required in drug screening, genetic and proteomic analysis. The multi-level electrophoresis device with closely-packed micro-channels not only provides parallel separations of analytes for high throughput and density LOC but the close proximity of the channels with applied electric field can also serve as an external electric field controller. By changing the polarity and magnitude of the external electric field in the micro-channel, the EOF can be reduced or increased, which is equivalent of having a dynamic capillary length [126].

For the multi-level electrophoresis device, a 50.8 mm × 50.8 mm × 1 mm high purity synthetic fused silica was used. The layout design for demonstrating parallel electrophoresis in fused silica is shown in Figure 5.7(a). The combination of four embedded channels and eight open reservoirs was laid out over 21 mm × 21 mm area. The over-passing and under-passing channels connecting the sample reservoirs (SR-O, SR-U) with the sample waste reservoirs (SW-O, SW-U), or the buffer reservoirs (BR-O, BR-U) with the buffer waste reservoirs (BW-O, BW-U), consisted of a 5-mm long straight section centered between two S-curved sections of 7.85 mm length that entered the respective reservoirs through an additional 2.5-mm straight section. The S-bends were formed from two quarter circular arcs with a radius of 2.5 mm. All channels were 2.57 cm long. Figure 5.7(b) shows the expanded view of the intersecting buffer and sample channels and the vertical access port arrays where they under/over-pass at a position 10.55 mm downstream from BR-O and BR-U reservoirs. These junctions provided for plug formation. The under-passing buffer channel was written to slope upward at 8° from $d_s = 140$ to 70 µm channel depth over a 500-µm length in order to match the upper buffer channel depth. In this way, simultaneous observation of analyte separation from inside the same imaging plane became possible along the two parallel channels,
Figure 5.7: The (a) full schematic drawing of the 3-D microfluidic device with under-passing and over-passing channels for capillary electrophoresis separation, and (b) the enlarged view at the channel crossing for pinch and plug formation. Here, $r$ is the radius of the circular shaped channels. $U =$ Under-passing, $O =$ Over-passing, $SR =$ Sample Reservoir, $BR =$ Buffer Reservoir, $BW =$ Buffer Waste Reservoir, $SW =$ Sample Waste Reservoir.
Figure 5.8: Optical images of (a) a completed multi-level electrophoresis device together with (b) a magnified view of the multi-level micro-channels at the crossings prior to HF etching.

with center-to-center separation of 100 µm. The sample and buffer micro-channels had a rectangular cross-section with dimensions of 50 µm × 40 µm written by an array of 30 × 16 laser tracks. The eight reservoirs had dimensions of 500 µm × 500 µm × 200 µm that were opened by etching excising cubes (Section 5.1.3.3). For this device, circular laser polarization was used to provide isotropic etching through the curved and crossed channel directions, and the G-code program used for patterning this device is presented in Appendix B. The laser-patterned fused silica was immersed in a 5% HF for 5 hours to completely etch out all the microfluidic structures.

Before device testing, a PDMS sheet of 3.2-mm thickness was used to seal all of the access ports, while also hosting larger reservoirs (6 mm diameter, 3.2 mm depth) aligned over the glass reservoirs such that larger buffer or analyte volumes would minimize the meniscus Laplace pressure. The glass substrate and the PDMS sheet were washed thoroughly with methanol, blown dry with N₂ gas and processed in a radio frequency oxygen
Table 5.1: Sample loading and separating voltage programs for electrophoresis

<table>
<thead>
<tr>
<th>Flow Phase</th>
<th>Program Purpose</th>
<th>SR-U</th>
<th>SR-O</th>
<th>SW-U</th>
<th>SW-O</th>
<th>BR-U</th>
<th>BR-O</th>
<th>BW-U</th>
<th>BW-O</th>
</tr>
</thead>
<tbody>
<tr>
<td>Sample Finishing Loading</td>
<td>1. Sample Loading</td>
<td>400 V</td>
<td>400 V</td>
<td>0</td>
<td>0</td>
<td>300 V</td>
<td>300 V</td>
<td>350 V</td>
<td>350 V</td>
</tr>
<tr>
<td>Sample Injection and Separation Phase</td>
<td>2. Parallel Equal Potentials</td>
<td>60 V</td>
<td>70 V</td>
<td>100 V</td>
<td>130 V</td>
<td>200 V</td>
<td>250 V</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td></td>
<td>3. Parallel Unequal Potentials</td>
<td>70 V</td>
<td>80 V</td>
<td>150 V</td>
<td>200 V</td>
<td>300 V</td>
<td>300 V</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td></td>
<td>4. Anti-Parallel Equal Potentials</td>
<td>120 V</td>
<td>170 V</td>
<td>220 V</td>
<td>260 V</td>
<td>350 V</td>
<td>400 V</td>
<td>0</td>
<td>0</td>
</tr>
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</table>

Capillary electrophoresis was driven on the electrophoresis device by applying a sequence of voltages to the reservoirs with an eight-channel high voltage sequencer (Lab-Smith HVS448 3000), having each channel connected to each reservoir through a wire as an electrode. Table 5.1 shows several voltage programs that were developed to form plugs of fluorescent dye on which to assess the plug flow speed, plug dispersion and separation efficacy. Before sample loading, all reservoirs, except BW-O and BW-U were filled with 1.01 M of sodium hydroxide (NaOH) while a vacuum of 24 inHg was applied at the BWs for 10 minutes to draw the NaOH solution through all micro-channels and established a high surface charge density. The NaOH solution was then cleared out, and 0.020 M sodium tetraborate solution was introduced under similar vacuum conditions for 15 minutes of flushing that ensured all air bubbles were removed from inside the channels. Reservoirs SR-U and SR-O were then each filled with 20 µL of 0.001 M Rh 123 and Rh B diluted with 70 µL of 0.020 M sodium tetraborate buffer solution to obtain a total of 110 µL of solution mixture with 0.0002 M Rh 123, 0.0002 M Rh B and 0.013 M sodium tetraborate buffer. All other reservoirs (i.e. SW-O, SW-U, BR-O, BR-U, BW-U, BW-O)
and BW-U were filled with 110 µL of 0.020 M sodium tetraborate buffer solution.

The fluorescent dye and buffer solution were loaded through the embedded cross channels under a pinched injection flow by using the reservoir potentials of Program 1 in Table 5.1 where 400 V potential was applied to SRs, 350 V potential was applied to BWs, 300 V potential was applied to BRs, and grounding was applied to SWs. A fluorescent plug was then formed in each of the parallel separation channels by switching to Program 2, 3, or 4, providing fluorescent plugs that moved in parallel at similar speed (Program 2: Parallel Equal Potential), in parallel at different speeds (Program 3: Parallel Unequal Potential), and at similar speed but in opposite parallel directions (Program 4: Anti-Parallel Unequal Potential), respectively. To create short plugs, the cross channel voltages for all programs (2–4) were tuned to limit buffer flow-back in the under-passing and over-passing cross channels and thereby compensate for differences in the meniscus Laplace pressure at the reservoirs. In this procedure, plugs were driven at potential differences from -250 V to +400 V along the 2.57 cm long separation channels. Since the injected plug in each micro-channel contained two differently charged fluorescent molecules (neutral Rh B and cationic Rh 123), they would separate along the channel under an applied electric field. The separations were observed and imaged under an inverted fluorescence microscope at the cross-channel intersections, and at a position 4.8 mm downstream from the intersections. Images were later analyzed to obtain the electropherograms, plug flow speeds and diffusion coefficients.

The dense packing of laser modification tracks together with the close proximity of parallel or cross channels created significant stresses that could lead to micro- or macro-cracks, spoiling the electrical and fluidic isolation between the channels. Figure 5.9 shows the formation of a crack along the over-passing channel connecting the under-passing channel. Such cracks were avoided when a vertical offset of 30 µm was used between under-passing and over-passing channels and when a 50 µm lateral wall-to-wall
Figure 5.9: Optical images of a crack formed in fused silica when the under-passing and over-passing microfluidic channels were too closely spaced with vertical and lateral offsets of 20 µm and 50 µm, respectively.
5.4.1 Parallel Capillary Electrophoresis: Equal Potentials

To demonstrate the functionality of the 3-D microfluidic channel network as formed by femtosecond laser and chemical etching, parallel capillary electrophoresis was carried out on the under-passing and over-passing channels using Programs 1 and 2 of Table 5.1. Fluorescent microscope images (Figure 5.10) show the generation (Figure 5.10(a)) and separation (Figure 5.10(b)) of the fluorescent plugs in under-passing (bottom-left cross-
Figure 5.11: Electropherograms of Rh 123 and Rh B plugs flowing in (a) under-passing and (b) over-passing channels for various potentials at the BR. Dashed lines show the linear increase of migration time as the applied potential decreases.

The videos captured at a position 4.8 mm downstream from the intersection of the cross...
channels for both under-passing and over-passing channels were used to generate electropherograms that plot the fluorescent intensities at varying migration time, showing the electrophoresis separation. Figure 5.11 shows typical electropherograms of the cross channels for both under-passing (Figure 5.11(a)) and over-passing (Figure 5.11(b)) channels where Rh 123 and Rh B have been identified to separate into isolated plugs. The Rh 123 migration times varied from 5.5 to 10.7 s and 4.8 to 9.4 s, whereas the Rh B arrived at later times, varied from 7.5 to 14.4 s and 6.3 to 12.0 s, for various potential differences in the range of 200 to 400 V along the respective under-passing and over-passing separation channels. The spatial peak variance, \( \sigma^2 \), in the plug length was also determined by fitting a Gaussian function to the temporal peak profile of the fluorescent intensity and recorded at various separation potentials and positions along the channels. The spatial peak variance was then plotted against the migration time, \( t \), as shown in Figure 5.12 for the fluorescent dyes Rh 123 (Figures 5.12(a) [over-passing channel] and 5.12(b) [under-passing channel]) and Rh B (Figures 5.12(c) [over-passing channel] and 5.12(d) [under-passing channel]) and was found to be well represented by the standard diffusion equation [127],

\[
\sigma^2 = 2D_{\text{eff}}t + \sigma_o^2,
\]

(5.1)

where \( D_{\text{eff}} \) is the effective diffusion coefficient and \( \sigma_o^2 \) is the initial plug width.

The slope of each plot (Figure 5.12) was then used to obtain the effective diffusion coefficient since the slope of the linear regression through this data is equal to two times the diffusion coefficient. In following the E-field method [128] for dynamic measurement of the diffusion coefficient, values of \( D_{\text{eff}} = 6.6 \pm 0.2 \times 10^{-6} \) to \( 8.7 \pm 0.5 \times 10^{-6} \) cm\(^2\)/s and \( 1.25 \pm 0.02 \times 10^{-5} \) to \( 1.63 \pm 0.08 \times 10^{-5} \) cm\(^2\)/s were found for Rh 123 in the under-passing and over-passing channels, respectively, for applied potential in the 200 to 400 V range. For Rh B, values of \( D_{\text{eff}} = 4.2 \pm 0.1 \times 10^{-6} \) to \( 6.4 \pm 0.9 \times 10^{-6} \) cm\(^2\)/s and \( 6.0 \pm 0.1 \times 10^{-6} \) to \( 9.1 \pm 0.9 \times 10^{-6} \) cm\(^2\)/s were obtained in the under-passing and over-
Figure 5.12: The measured spatial variance of fluorescent plugs plotted against the migration time for (a),(b) Rh 123 and (c),(d) Rh B in (a),(c) over-passing and (b),(d) under-passing channels for various potentials at the BR. Solid lines show a linear representation of the data used to obtain the effective diffusion coefficients.
passing channels, respectively, for applied potential tested in the range of 200 to 400 V. There is optical microscope evidence of slight over etching around the access ports of the over-passing channels, leading to slightly non-uniform channel walls that we attribute to the $\sim 88\%$ and $\sim 43\%$ higher effective diffusion coefficients for $\text{Rh 123}$ and $\text{Rh B}$ respectively, found here for the over-passing channel. Nonetheless, the $D_{\text{eff}}$ values are relative small for the under-passing (over-passing) channels, falling in ranges of $\sim 1.5$ to $2.0$ ($\sim 2.8$ to $3.7$) times and $\sim 1.0$ to $1.5$ ($\sim 1.4$ to $2.2$) times the values reported for binary diffusion coefficient for $\text{Rh 123}$ and $\text{Rh B}$ [129], respectively. This low ratio begins to rise rapidly ($>>1.0 \times$) when velocity exceeds $1$ mm/s, which we attribute to flow disturbances caused by the channel wall roughness, the presence of vertical access ports, and the Laplace pressure imbalances between reservoirs through Taylor-Aris dispersion [127]. At larger electroosmotic velocities of $1$ mm/s to $5$ mm/s, Blom et al. [127] reported much larger diffusion ratios of $1.65 \times$ to $17.25 \times$, respectively, that were attributed to $3$ $\mu$m periodic perturbations formed on $100$ $\mu$m wide channel. In contrast, the very low wall roughness ($<1$ nm) in conventional fused silica capillaries (nominal diameter of $20$ to $100$ $\mu$m) yield a very small $<0.04\%$ increase in effective dispersion that was measured to scale with the sixth-power of the surface fluctuation as reported by Pu et al. [130]. The present sub-micron roughness of the laser-formed channels is capable to outperform devices such as reported by Pu et al. [130] and Ghobeity et al. [131] for powder-blasted channels with surface roughness of $1–5$ $\mu$m and $0.4–0.6$ $\mu$m, respectively. The laser-formed channels are therefore sufficiently efficient for many capillary electrophoresis applications and would benefit with further reduction in $D_{\text{eff}}$ through means to eliminate the reservoir pressure differences with precise control of reservoir filling, to automate the alignment of the laser polarization to follow the curved path of channels, and to reduce the size and the number of vertical access ports.

The migration time for the fluorescent dyes are seen to fall inversely with the poten-
Figure 5.13: The observed flow speeds of the Rh 123 and Rh B plugs observed through the under-passing and over-passing micro-channels of the device in Figure 5.8 together with separation speeds with different applied potentials at the BRs.

The present device showed a clear separation of the two fluorescent dyes in the electropherogram (Figure 5.11) that manifested in very similar separation speeds for the under-passing and over-passing channels, rising from \( \sim 110 \) to 230 \( \mu \text{m/s} \) with increasing potential as seen in Figure 5.13. The present \( D_{eff} \) values correspond to theoretical plates and resolution values as high as \( \sim 13000 \) and \( \sim 8.6 \), respectively. Further balancing of the
fluorescent dye and buffer concentrations together with optimization of the channel geometry, size and roughness promise additional improvement in reducing the diffusion and increasing the resolution of the present channels.

### 5.4.2 Parallel Capillary Electrophoresis: Unequal Potentials

When unequal potentials (Program 3) were applied in such closely separated (50 µm wall gap) channels, strong plasma-treated PDMS sealing of the vertical access ports was required in order to prevent cross flow of fluorescent dyes between channel sections held at different voltage bias. The flow speeds in the over-passing and under-passing channels could be independently controlled with different bias, generally following with the same speeds, separation speeds, and diffusion as reported for the parallel equal potential case in Figures 5.11 and 5.13. The flow dynamics for this biasing were only weakly affected by a moderately strong transverse field (<1.2 × 10⁴ V/cm) that formed between the nearby
channels. Such fields will modify the charge density on the inner channel walls to affect the electroosmotic flow as reported by Lee et al. \cite{132} and by Ghowsi and Gale\cite{133}. Figure 5.14 shows the resulting flow speeds of the fluorescent plugs with 250 V and 350 V applied potentials for the respective over-passing and under-passing channels. Different potentials of 250 V and 350 V applied to the over-passing and under-passing channels, respectively, led to 6.4% (8.4%) and 3.0% (4.8%) slower speeds for the Rh 123 (Rh B) dye in the respective channels in contrast with the case of equal potential biasing in Figure 5.13. While a slower speed was indeed expected in the over-passing channel, Lee et al. \cite{132} and Ghowsi and Gale\cite{133} predicted an accelerated speed in the higher biased channel. However, the small 3.0% (Rh 123) and 4.8% (Rh B) decrease here may be attributed to the unbalanced Laplace pressure developing as the BR volumes became depleted. These external field effects may be flexibly mitigated by further separating the channels during the 3-D laser-writing step.

In addition to independently controlling the plug flow speeds in the parallel channels,
the plugs could be directed in reverse or opposite flow directions as shown in Figure 5.15 when running Program 1 and 4 of Table 1. The device offered high repeatable injections with holding times of 0.5 s and 1.0 s for Programs 1 and 4, respectively. The generation of oppositely directed plugs in such closely separated channels verified their strong electrical and mechanical isolation up to $\sim 10^5$ V/cm tested here. Otherwise incomplete sealing of the vertical ports or micro-crack formation would lead to cross contamination and leakage in the presence of very strong transverse electric fields.
Chapter 6

Significance of this Work

The fabrication of biophotonic LOC devices with microfluidic structures and integrated bio-sensors has been broadly demonstrated in various materials using multiple micro fabrication techniques. New techniques and methods are constantly being developed to circumvent shortcomings and enhance functionalities in LOC for improved efficiency in numerous application directions. The technique of femtosecond laser irradiation with chemical etching (FLICE) is a rapid and efficient way to serve in a research environment for prototyping as discussed in Chapter 2. This chapter discusses the significance of the contribution made to FLICE in this particular work and the contribution thereof to the scientific community towards the fabrication of 3-D biophotonic LOC components and devices. Moreover, further FLICE work in progress is presented in this chapter.

6.1 The Indepth Study of Microfluidic Components

The present results in Chapters 4 and 5 provide an efficient and rapid prototyping technology for fabricating 3-D optofluidic devices. Polarization controlled orientation of laser generated nanogratings was instrumental for enabling single-step laser patterning of all buried microfluidic channels and optical circuits prior to the HF etching step, harnessing nanograting stop-layers to prevent degradation of optical devices at the microfluidic inter-
faces. In comparison with the waveguide integration method demonstrated by Bellouard et al. [47], which required the fabrication of a thick stop-layer between the micro-channel wall and the intersecting waveguide, the polarization control of waveguide writing eliminates the need for such thick stop-layer that possibly degrades the optical performance.

Very low laser exposure conditions with polarization perpendicular to the scanning direction were identified to generate very smooth (~10 nm) and crack-free channels buried in the glass that dramatically reduce optical scattering loss for crossing optical waveguides scanned with parallel polarization. Previously, fs-laser photoablation produced surface roughness in the micron scale [70], while the FLICE technique, with a 100-fs laser operated at a wavelength of 800-nm and a repetition rate of 250-kHz, improved the surface roughness to below 1 µm [17] and 0.25 to 0.50 µm [3] in fused silica glass. Even with post-annealing process, only a rms roughness of a few tens of nanometers roughness has been demonstrated [74]. The optically smooth channel sidewalls demonstrated here are attractive for further applications requiring internal reflection to probe analytes or to integrate micro-mirrors as demonstrated by Sugioka and coworkers [5] in Foturan glass. Polarization and scanning-angle control to create such flat and smooth surfaces would also improve the precision of integrated micro-mechanical and optical devices for optical sensing applications as reported by Bellouard et al. [3]. Otherwise, circular polarization with isotropic etching in all directions provided the most facile means for the fabrication of curved channels, but with the disadvantage of rougher channel walls.

Two-dimensional (2-D) or planar LOCs have the advantage of fabricating microfluidic channels in any desired length, while the limitation in 3-D microfluidic channel length fabricated in bulk glass has always been a challenge with the longest demonstrated length of 9.2 mm [18] using the FLICE technique. Here, with the formation of periodic vertical access holes for HF etching, this new approach significantly opens the domain for writing microfluidic channels that may be buried inside the bulk glass and extended over unlimited length in any desired 3-D routing direction. In this way, multi-level network
Chapter 6. Significance of this Work

6.2 Inverted Woodpile Structure in Bulk Glass

The advances made here to FLICE include fabrication of arbitrarily cross-sectional shaped channels by writing parallel capillary arrays and harnessing the laser-generated nanogratings to enable the etching of smooth-walled straight micro-channel and curved micro-channel without ultrasonic agitation. The ultrasonic agitation has been used to facilitate the flow of the solution during the etching process to enable the etching of deep structures with a high etch rate \[5\,6\,12\,14\,18\,112\]. However, such ultrasonic waves are detrimental to delicate structures. For the first time, the approach here without ultrasonic agitation was extended to form uniform inverted woodpile structure (IWP) directly inside fused silica glass by writing orthogonal laser tracks that open up to connect micro-capillary arrays after chemical etching, while maintaining an HF etch rate on par with other groups \[12\,14\,112\]. This high-density IWP not only demonstrated a demanding example of 3-D fabrication, it also facilitated the direct integration of waveguides with micro-channels fabricated in the laser exposure step. With the micron-size capillary, IWP could define a structured micro-channel providing chromatographic capability in a highly inert and chemically stable substrate of fused silica to facilitate separations of analytes.

With the fabrication of photonic crystal (PC) structures that must be constructed from optically transparent materials with high dielectric constant, fused silica glass is a natural candidate to serve as template compared with the toxic chalocogenide glasses \[24\] or hydrophobic photoresists \[23\]. Various groups have demonstrated techniques to form large area PCs that allow microfluidic capability with the superior silica properties over polymers, including colloidal self-assembly of silica microsphere \[81\] and multi-step silica chemical vapour deposition coating over a photoresist template \[25\]. The further scaling
down of the capillaries to sub-micron size of the 3-D IWP demonstrated here promises PC structures that can be directly integrated within the micro-channels where the bandgaps may shift into the near IR or visible band for new optical sensing direction. The highly flexible laser-writing step also permits more complex open capillary arrangements for controlled incorporation of pre-engineered defects to disrupt the periodicity of the PC and possibly create optical states within the otherwise forbidden bandgap frequencies. Such PC and IWP substrates will open up new directions in biophotonic LOC with even more compact designs employing new optical and chromatography flow capabilities for new applications.

6.3 Rapid Prototyping of Three-Dimensional Lab-on-a-Chip

By harnessing the 3-D writing capability of femtosecond lasers together with selective chemical etching along the laser-generated nanograting planes, fully integrated 3-D LOCs in glasses can be precisely fabricated without the need for a cleanroom environment or multi-step photolithography processes [7,8]. The laser-formed microfluidic channels, with periodic vertical access ports, were crack-free and had sufficiently smooth walls to harness capillary or electrokinetic force to demonstrate particle counting in the cytometer (1% counting error) or dual-channel parallel electrophoresis separation in the 3-D multi-level device, respectively.

Optical waveguides were often laser-written on a LOC with a glass layer between the micro-channel and the waveguide [17], after micro-channel fabrication [4], and after an annealing process to smoothen the channel wall roughness [74]. These processing steps increased the complexity of the fabrication process, but were required to prevent damage to the optical waveguides or to prevent waveguide degradation during the high temperature annealing. With the work presented in this thesis, the nanograting orientation
Chapter 6. Significance of this Work

aligned parallel with the micro-channel, written at low laser energy near the threshold of nanograting formation, and perpendicular with the waveguide enabled one-step direct laser patterning of the LOC prior to chemical etching to form channels with smooth side-walls for low scattering loss waveguide integration. The significant improvement of the channel-wall surface roughness demonstrated in this work compared with that demonstrated previously \[3,17,74\] advances the FLICE technique to become more attractive for optofluidic integration.

The incorporation of vertical access ports, allowing access for HF etching, to facilitate fabrication of unrestricted channel length proves to have only a minor effect on both the capillary and electrokinetic flows. During the electrophoresis separation in the multi-level device fabricated with circular polarization, fluorescent emission was not detected from inside the vertical access ports as the plug passed along the separation channel, suggesting little or insignificant mixing of the micro-channel flow into the stagnant port volume. The moderately small increase in the effective diffusion of less than 3-fold over the binary diffusion coefficient for electrophoresis indicates the present channels to be comparable or better than microfluidic devices fabricated with more traditional techniques \[130,131\]. Further improvement in separation efficiency is expected with more advanced laser procedures that promise to smoothen the channel walls or reduce the diameter size and number of vertical access ports.

The close proximity of the parallel channels demonstrated in this work, together with the new opportunity to under-pass or over-pass channels, offer significant advantages towards the high densification of microfluidic components in LOCs. Multi-level arrays of such channels promise the simultaneous observation of capillary electrophoresis separation with nearly independent control of isolated channels. Together, these advantages provide the means toward the 3-D integration of closely packed multi-level micro-channel networks for fast parallel capillary electrophoresis in highly functional LOCs in glasses for simultaneous analysis of many analytes without cross contamination. Although fem-
tosecond laser writing is a moderately slow process step, the highly flexible 3-D writing approach is especially desirable to serve as a value added tool to enhance the functionality of traditional 2-D LOCs fabricated with batch processing. The demonstration of many femtosecond laser processes for writing of optical components inside fused silica further broadens the prospects for seamless integration of optical and microfluidic devices for fabrication of optofluidic microsystems with parallel fluidic and optical analysis capability.

6.4 Future Work

Great strides have been made in the fabrication of microfluidic channel, reservoir and inverted woodpile structure (IWP), extending their functionality and viability for biophotonic LOC applications. Vertical access ports have been incorporated within the embedded micro-channels that permit unrestricted length for integration on 3-D LOCs such as demonstrated in the waveguide probe device (Section 5.2), particle counting device (Section 5.3), and multi-level electrophoresis device (Section 5.4). The development of this reproducible technique to fabricate 3-D structures opens the door to microphotronics and chromatography integrated LOCs. However, much work remains to be done.

The preliminary step to integrate 3-D microphotronics and chromatography onto LOCs is the seamless incorporation of IWP structure. The following section gives a small glimpse to the work in progress for realizing such device.

6.4.1 Microphotonic Chromatographic Lab-on-a-Chip

The microphotonic chromatographic LOC in fused silica is constructed in much the same way as the devices described in Chapter 5 using the FLICE technique. Figure 5.1 illustrates the microphotonic chromatographic device that incorporates the fruit of the researches presented above, including micro-channel, waveguide, IWP structure and reser-
Figure 6.1: The full schematic drawing (not to scale) of the microphotonic chromatographic device with an enlarged view (right) of the inverted-woodpile structure integrated with the micro-channel.

Figure 6.1: The full schematic drawing (not to scale) of the microphotonic chromatographic device with an enlarged view (right) of the inverted-woodpile structure integrated with the micro-channel.

The micro-channel with the robust glass IWP structure can serve as the stationary phase for facilitating the separation of analytes in chromatography. Moreover, the waveguides intersecting the IWP structure and the micro-channel after the IWP and outside the device function as the probing and reference waveguides, respectively.

The preliminary construct of the microphotonic chromatographic LOC device is shown in Figure 6.2 with truncated micro-channels. This initial attempt of the device was laser-scanned with energies of $E_p = 125$ nJ, 75 nJ, 75 nJ, and 50 nJ for the reservoirs, micro-channels, IWP structure, and waveguides, respectively. The channel integrated with the IWP structure has dimensions of $32 \text{ mm} \times 0.05 \text{ mm} \times 0.05 \text{ mm}$ which include the $8 \text{ mm} \times 0.05 \text{ mm} \times 0.05 \text{ mm}$ IWP channel, and the cross channel has dimensions of $16 \text{ mm} \times 0.05 \text{ mm} \times 0.05 \text{ mm}$. All reservoirs have dimensions of $0.5 \text{ mm} \times 0.5 \text{ mm} \times 0.1 \text{ mm}$ that connect the ends of the micro-channels. The G-code program used for patterning this device is presented in Appendix C.

The next stage of experimentation would be forming an uniform IWP channel and
testing the performance in electrophoresis separation to prove its efficacy. Further, the validation of integrating IWP structure together with the continual effort of scaling down the capillaries would greatly accelerate the direct PC integrated LOC devices in fused silica.

Figure 6.2: Optical image of the microphotonic device in progress with a magnified view (right) of the inverted-woodpile channel etched for 30 minutes.
Chapter 7

Conclusions

Femtosecond laser irradiation with chemical etching (FLICE) was targeted for study as an attractive technique for fabrication of 3-D biophotonic LOC components and microfluidic channel integrated with optical waveguide and IWP structure. Fused silica glass was selected due to its attractive material properties: hard, durable, low thermal expansion, high chemical resistance, dielectric constant and thermal conductivity. Further, the wide optical transparency from the UV to IR spectrum offer a suitable platform for formation of optical waveguides and components. The numerous flexible combinations of these microfluidic and optical components were examined, leading to the demonstration of a waveguide probing device, a particle counting device and a multi-level electrophoresis LOC.

A broad range of femtosecond laser exposure conditions were explored experimentally, and found to enable wide exposure window and high polarization contrast HF acid etching with perpendicular polarization (28:1) whereas circular polarization offered scanning-angle-invariant etching. These polarization approaches provided the means to fabricate straight channels and curved channels, respectively. Low energy laser patterning of buried opened micro-channels of flexible cross-sectional shape with extended length facilitated the fabrication of crack-free rectangular micro-channels with smooth sidewalls.
Chapter 7. Conclusions

(~10 nm rms), enabling low loss (0.07 dB/wall) integration via laser writing polarization control of micro-channels and optical waveguides in the waveguide probing device. This extensive study on channel shape optimization together with the integration techniques enabled an efficient prototyping technology for fabrication of complex 3-D LOCs.

Extending the multi-line fabrication technique in laser-scanning microfluidic structures, a femtosecond-laser exposure technique with an energy compensation algorithm had been developed to directly form a uniform 3-D IWP structure of diamond-like symmetry in fused silica with a close-packed transverse and vertical periodicities of 5 µm and 14 µm, respectively. The IWP structure further defines a micro-channel in which new microfluidic application directions in chemical assays and chromatography may be developed to take advantage of the high stability and chemical inertness of fused silica. By scaling to smaller capillaries, the present structure promises optical sensing.

To highlight the unprecedented 3-D capability and flexibility of femtosecond laser writing together with selective chemical etching, various fused silica glass LOC devices were demonstrated, concentrating on the capillary force driven particle counting device and the multi-layered parallel channel electrophoresis separation device of Sections 5.3 and 5.4, respectively. A detailed characterization demonstrated the microfluidic efficacy and the smoothness of the laser-defined channel walls for driving strong capillary force and moderately low-dispersion (<4× the binary diffusion coefficient) electrokinetic flows on par with devices formed by traditional fabrication techniques. The flexibility and accuracy of this fabrication technique presented a compact flow particle counting chip with a precise particle count of less than 1% error. Further, parallel channel capillary electrophoresis separation of fluorescent dyes was experimentally demonstrated in a multi-level arrangement of under-passing and over-passing channels.

This thesis work has contributed in many ways to advance the femtosecond laser fabrication of biophotonic LOC as discussed in Chapter 6. However, there are still improvements and open challenges remaining. The FLICE technique is a rapid prototyping
method that enables flexible fabrication of 3-D LOCs, but the laser scanning step is time consuming. With the low-power laser processing window found in this work, it is possible to split a single laser beam into multiple beams for parallel processing of multiple LOCs simultaneously, decreasing the overall processing time and improving the efficiency of exploratory work. Since the laser scanning step is one of the limiting factor for speeding up the FLICE fabrication process, exploring a new processing window with higher laser repetition rate can possibly improve the fabrication time by enabling higher scanning speed. To further exploit the 3-D fabrication capability of the FLICE technique that provided an unprecedented sidewall smoothness, micro-optical components such as micro-mirrors and micro-lenses, which were demonstrated in Foturan glass that required post-annealing process to achieve a smoothness of tens of nanometers, can now be integrated on LOCs formed in fused silica glass. The scaling of capillaries to submicron size with a higher NA lens is promising to integrate PC capable of optical sensing together with chromatographic separation of analytes that could be the next significant step forward in enabling a fully integrated microphotonic LOC in the chemically and optically superior fused silica glass. Moreover, the fluid flow disturbances can be further improved to lower effective diffusion by reducing the number of vertical access ports that facilitate chemical etching of embedded micro-structures. An approach is to improve the wet etching chemistry that would provide a higher etching contrast between the laser-modified volume and the unmodified glass, while maintaining a high etching rate.

The future work in progress presented in Section 6.4 is only a glimpse of what could be achieved, and the research currently underway to fabricate lab-on-a-fiber with the technique developed in our laboratory is pushing the forefront of LOC fabrication. Since silica can readily be formed on silicon electronics, optofluidic structures in silica glass may be explored for direct integration on electronic-ready platforms. As the field of LOC moves forward, the femtosecond laser work presented here provides a facile means for opening new 3-D design approaches in developing highly advanced and densely pack-
aged biophotonic LOCs.
Appendix A

G-code: Particle Counting Cytometer

The G-code used for controlling the xyz-stages in laser patterning the particle counting cytometer is presented below.

```plaintext
// Title: Particle Counting Cytometer
// Author: Stephen Ho
// Date Created: September 02, 2011
// Description: Writes an embedded cytometer having a tapered channel with curved opening and 3 waste reservoirs

////////////////////////////////////////////////////////////////////////////
// Parameters (USER INPUT REQUIRED)

#DEFINE NSPEED 1 ; NUMBER OF SCANNING SPEED
#DEFINE NLDEPTH 1 ; NUMBER OF LAYER TO LAYER SEPARATION
#DEFINE NLINESEP 1 ; NUMBER OF TRANSVERSE LINE TO LINE SEPARATION
#DEFINE CDEPTH 1 ; NUMBER OF CHANNEL DEPTH
#DEFINE HOLESEP 1 ; NUMBER OF VERTICAL ACCESS PORT SEPARATION

DVAR $SPEED[NSPEED] $LDEPTH[NLDEPTH] $LINESEP[NLINESEP]
$DEPTH[CDEPTH] $HSEP[HOLESEP]

#DEFINE NLAYER 10 ; NUMBER OF LAYERS FOR THE FIRST CHANNEL
#DEFINE NLAYER2 5 ; NUMBER OF LAYERS FOR THE SECOND CHANNEL
#DEFINE NLINE 130 ; NUMBER OF LINES IN EACH LAYER
#DEFINE NLINE2 8 ; NUMBER OF LINES IN EACH LAYER FOR THE SECOND CHANNEL
#DEFINE LENGTH 20.4 ; LENGTH OF THE FIRST CHANNEL + TAPERLENGTH
#DEFINE LENGTH2 2 ; LENGTH OF THE SECOND CHANNEL
#DEFINE TAPERLENGTH 0.4 ; LENGTH OF THE TAPER SECTION
#DEFINE ABOVES 0.5 ; THE DISTANCE ABOVE THE SAMPLE SURFACE
```
#DEFINE OVERSHOOT 0.005 ; THE OVERSHOOT DISTANCE INTO THE CHANNEL
#DEFINE CEXTRA 0.050 ; THE EXTRA CHANNEL LENGTH INTO EACH RESERVOIR
#DEFINE RWIDTH 2.0 ; THE WIDTH OF THE RESERVOIR
#DEFINE RLENGTH 1.0 ; THE LENGTH OF THE RESERVOIR
#DEFINE RLENGTHSMALL 0.2 ; THE LENGTH OF THE SMALL RESERVOIRS
#DEFINE RHEIGHT 0.5 ; THE HEIGHT OF THE RESERVOIR
#DEFINE NSHELL 2 ; NUMBER OF SHELLS AT EACH BOARDER
#DEFINE DRT 0.002 ; TRANSVERSE SPACING IN RESERVOIR
#DEFINE DRL 0.005 ; LAYER SPACING IN RESERVOIR
#DEFINE CURVERADIUS 0.100 ; THE RADIUS OF THE CURVE FOR THE TAPER BOTTOM
#DEFINE CURVERADIUS2 0.05 ; THE RADIUS OF THE CURVE FOR THE TAPER TOP
#DEFINE DELTAY 0.002 ; LINE TO LINE SEPARATION FOR THE CURVED TAPER
#DEFINE CHANNEL2DEPTH 0.02 ; THE DEPTH OF THE SECOND CHANNEL FROM THE SURFACE

DVAR $CIRPOS $CURVELINES $YSFT $XSFT $LZERO $LONE $LTWO $LTHREE $LFOUR $LFIVE $LSIX $LSEVEN $LEIGHT $LNINE $LTEN $LELEVEN $LTWELVE $LTHIRTEEN $YFLAG $XFLAG $COUNTER

METRIC SECONDS

; PRESET PARAMETERS

$HSEP[0]=0.1 ; HORIZONTAL SEPARATION FOR EACH VERTICAL HOLE
$SPEED[0]=0.5 ; SCAN SPEED FOR THROUGH CHANNELS
$LDEPTH[0]=0.002 ; LAYER SEPARATION FOR EACH LAYER IN EACH CHANNEL FOR BOTTOM UP
$LINESEP[0]=-0.0015 ; SEPARATION FOR EACH LINE IN EACH LAYER
$DEPTH[0]=-0.05 ; THE CHANNEL DEPTH (MACHINE) FROM THE SURFACE (MULTIPLY 1.4 TO GET THE CORRECT DEPTH FROM THE SURFACE)
$XSFT = 0 ; DEFINE OFFSITE LENGTH ON X-AXIS
$YFLAG = -1
$XFLAG = 1

;;;;;;;;;;;;;;;;;;;;;;;;;;;;;;;;;;;;;;;;;;;;;;;;;;;;;;;;;;;;;;;;;;;;;;;;;;;;;;;;;;;;;;;;;;;;;;;;
; START OF PROGRAM

; ***MAKING RESERVOIR 1 (SAMPLE RESERVOIR)***
Appendix A. G-code: Particle Counting Cytometer

G90 UU16.147 E1 ; DEFINE THE SCANNING POWER

$YFLAG = -1
$XFLAG = -1

FOR $LTEN = 0 TO ((RHEIGHT / 0.1)-1) ; LOOP FOR NUMBER OF LAYERS IN THE OPTICAL AXIS DIRECTION

G90 Z2 F10 ; MOVE ABOVE THE FOCAL PLANE
G90 X0 Y0 F10 ; MOVE BACK TO THE ORIGINAL REFERENCE POINT
G90 Z((RHEIGHT * -1 + $LTEN * 0.1)/1.4) F1 ; MOVE TO THE BOTTOM LAYER

FOR $LELEVEN = 0 TO (RLENGTH/DRT) ; LOOP FOR NUMBER OF LINES AT THE BOTTOM LAYER

G91 Y(RWIDTH * $YFLAG) F$SPEED[0] ; SCAN ONE LINE IN THE Y-DIRECTION
G91 X(DRT * $XFLAG) F1 ; MOVE TO THE NEXT LINE
$YFLAG = $YFLAG * -1 ; CHANGE DIRECTION

NEXT $LELEVEN

G90 LINEAR Z2 F100 ; MOVE ABOVE THE FOCAL PLANE
G90 X0 Y0 F10 ; MOVE BACK TO THE ORIGINAL POSITION
$YFLAG = -1
$XFLAG = -1

FOR $LEIGHT = 0 TO (RWIDTH/0.1) ; LOOP FOR NUMBER OF LAYERS IN THE WIDTHWISE DIRECTION

G90 Z((RHEIGHT * -1 + $LTEN * 0.1)/1.4) F1 ; MOVE TO THE BOTTOM OF THE LAYER
G90 Y(-0.1 * $LEIGHT) ; MOVE TO THE CORRECT POSITION IN THE Y-DIRECTION

FOR $LNINE = 0 TO (0.1 / DRL) ; LOOP FOR NUMBER OF LAYERS IN EACH CUBE

G91 X(RLENGTH * $XFLAG) F$SPEED[0] ; SCAN ONE LINE IN X-DIRECTION
$XFLAG = $XFLAG * -1 ; CHANGE DIRECTION
G91 Y(DRT * $YFLAG) F1 ; MOVE TO THE NEXT SHELL
$YFLAG = $YFLAG * -1 ; CHANGE DIRECTION
G91 X(RLENGTH * $XFLAG) F$SPEED[0] ; SCAN ONE LINE BACKWARD IN X-DIRECTION
$XFLAG = $XFLAG *-1 ; CHANGE DIRECTION
G91 Z((DRL)/1.4) F1 ; MOVE UP ONE LAYER

NEXT $LNINE
NEXT $LEIGHT

G90 LINEAR Z2 F100 ; MOVE ABOVE THE FOCAL PLANE
G90 X0 Y0 F10 ; MOVE BACK TO THE ORIGINAL POSITION

$YFLAG = -1
$XFLAG = -1

FOR $LEIGHT = 0 TO (RLENGTH/0.1) ; LOOP FOR NUMBER OF LAYERS IN
THE LENGTHWISE DIRECTION

G90 Z((RHEIGHT * -1 + $LTEN * 0.1)/1.4) F1 ; MOVE TO THE BOTTOM OF THE
LAYER
G90 X(-0.1 * $LEIGHT) ; MOVE TO THE CORRECT POSITION IN THE X-DIRECTION

FOR $LNINE = 0 TO (0.1 / DRL) ; LOOP FOR NUMBER OF LAYERS IN EACH
CUBE

G91 Y(RWIDTH * $YFLAG) F$SPEED[0] ; SCAN ONE LINE IN X-DIRECTION
G91 X(DRT * $XFLAG) F1 ; MOVE TO THE NEXT SHELL
$XFLAG = $XFLAG * -1 ; CHANGE DIRECTION
$YFLAG = $YFLAG * -1 ; CHANGE DIRECTION
G91 Y(RWIDTH * $YFLAG) F$SPEED[0] ; SCAN ONE LINE BACKWARD IN X-
DIRECTION
$YFLAG = $YFLAG *-1 ; CHANGE DIRECTION
G91 Z((DRL)/1.4) F1 ; MOVE UP ONE LAYER

NEXT $LNINE

NEXT $LEIGHT

$YFLAG = -1
$XFLAG = -1

NEXT $LTEN

G90 Z2 F10 ; MOVE ABOVE THE FOCAL PLANE
G90 X0 Y0 F10 ; MOVE BACK TO THE ORIGINAL REFERENCE POINT

; ***MAKING RESERVOIR 2 (MAKING SMALL RESERVOIRS)***

FOR $LTHIRTEEN = 0 TO INT((RLENGTH/RLENGTHSMALL)/2) ; LOOP FOR
NUMBER OF WASTE RESERVOIRS

$YFLAG = -1
\$XFLAG = -1

IF \$LTHIRTEEN == 0 ; MAKING 1 OF 3 SMALL RESERVOIRS
G90 Y(LENGTH * -1 - RWIDTH) ; MOVE TO THE RESERVOIR ON THE OTHER END
G92 X0 Y0 ; SET A NEW REFERENCE POINT
ELSE ; MAKING 2 OR 3 OF 3 SMALL RESERVOIRS
G90 X(RLENGTHSMALL * 2 * \$XFLAG) F10 ; MOVE TO THE STARTING POINT OF THE NEXT SMALL RESERVOIR
G92 X0 Y0 ; SET THE NEW REFERENCE POINT
ENDIF

FOR \$LTEN = 0 TO ((RHEIGHT / 0.1)-1) ; LOOP FOR NUMBER OF LAYERS IN THE OPTICAL AXIS DIRECTION
G90 Z2 F10 ; MOVE ABOVE THE FOCAL PLANE
G90 X0 Y0 F10 ; MOVE BACK TO THE ORIGINAL REFERENCE POINT
G90 Z((RHEIGHT * -1 + \$LTEN * 0.1)/1.4) F1 ; MOVE TO THE BOTTOM OF THE LAYER
FOR \$LELEVEN = 0 TO (RLENGTHSMALL/DRT) ; LOOP FOR NUMBER OF LINES AT THE BOTTOM LAYER
G91 Y(RWIDTH * \$YFLAG) F\$SPEED[0] ; SCAN ONE LINE IN THE Y-DIRECTION
G91 X(DRT * \$XFLAG) F1 ; MOVE TO THE NEXT LINE
\$YFLAG = \$YFLAG * -1 ; CHANGE DIRECTION
NEXT \$LELEVEN
G90 LINEAR Z2 F100 ; MOVE ABOVE THE FOCAL PLANE
G90 X0 Y0 F10 ; MOVE BACK TO THE ORIGINAL POSITION
\$YFLAG = -1
\$XFLAG = -1

FOR \$LEIGHT = 0 TO (RWIDTH/0.1) ; LOOP FOR NUMBER OF LAYERS IN THE WIDTHWISE DIRECTION
G90 Z((RHEIGHT * -1 + \$LTEN * 0.1)/1.4) F1 ; MOVE TO THE BOTTOM OF THE LAYER
G90 Y(-0.1 * \$LEIGHT) ; MOVE TO THE CORRECT LAYER POSITION IN THE Y-DIRECTION
FOR \$LNINE = 0 TO (0.1 / DRL) ; LOOP FOR NUMBER OF LAYERS IN EACH CUBE
Appendix A. G-code: Particle Counting Cytometer

G91 X(RLENGTHSMALL * $XFLAG) F$SPEED[0] ; SCAN ONE LINE IN X-DIRECTION
$XFLAG = $XFLAG * -1 ; CHANGE DIRECTION
G91 Y(DRT * $YFLAG) F1 ; MOVE TO THE NEXT SHELL
$YFLAG = $YFLAG * -1 ; CHANGE DIRECTION
G91 X(RLENGTHSMALL * $XFLAG) F$SPEED[0] ; SCAN ONE LINE BACKWARD
IN X-DIRECTION
$XFLAG = $XFLAG * -1 ; CHANGE DIRECTION
G91 Z((DRL)/1.4) F1 ; MOVE UP ONE LAYER

NEXT $LNINE

NEXT $LEIGHT

G90 LINEAR Z2 F100 ; MOVE ABOVE THE FOCAL PLANE
G90 X0 Y0 F10 ; MOVE BACK TO THE ORIGINAL POSITION
$YFLAG = -1
$XFLAG = -1

FOR $LEIGHT = 0 TO (RLENGTHSMALL/0.1) ; LOOP FOR NUMBER OF LAYERS IN THE LENGTHWISE DIRECTION
G90 Z((RHEIGHT * -1 + $LTEN * 0.1)/1.4) F1 ; MOVE TO THE BOTTOM OF THE LAYER
G90 X(-0.1 * $LEIGHT) ; MOVE TO THE CORRECT LAYER POSITION IN THE X-DIRECTION
FOR $LNINE = 0 TO (0.1 / DRL) ; LOOP FOR NUMBER OF LAYERS IN EACH CUBE
G91 Y(RWIDTH * $YFLAG) F$SPEED[0] ; SCAN ONE LINE IN X-DIRECTION
$YFLAG = $YFLAG * -1 ; CHANGE DIRECTION
G91 X(DRT * $XFLAG) F1 ; MOVE TO THE NEXT SHELL
$XFLAG = $XFLAG * -1 ; CHANGE DIRECTION
G91 Y(RWIDTH * $YFLAG) F$SPEED[0] ; SCAN ONE LINE BACKWARD IN X-DIRECTION
$YFLAG = $YFLAG * -1 ; CHANGE DIRECTION
G91 Z((DRL)/1.4) F1 ; MOVE UP ONE LAYER

NEXT $LNINE

NEXT $LEIGHT

$YFLAG = -1
$XFLAG = -1
NEXT $LTEN
G90 Z2 F10 ; MOVE ABOVE THE FOCAL PLANE
G90 X0 Y0 F10 ; MOVE BACK TO THE ORIGINAL REFERENCE POINT FOR THE SECOND RESERVOIR

NEXT $LTHIRTEEN

G90 X(2 * RLENGTHSMALL * INT((RLENGTH/RLENGTHSMALL)/2)) Y0 F10 ; MOVE BACK TO THE ORIGINAL REFERENCE POINT FOR THE SECOND RESERVOIR
G92 X0 Y0 ; SET THE REFERENCE POINT BACK TO THE BEGINNING OF THE SECOND RESERVOIR

***MAKING A CURVED BOTTOM AT THE TAPER***

G90 Z2 F10 ; MOVE ABOVE THE FOCAL PLANE
G90 UU18.771 E1 ; SET THE POWER FOR THE CURVED TAPER CHANNEL BOTTOM
G90 X0 Y0 F10 ; MOVE BACK TO THE REFERENCE POSITION FOR THE LAST RESERVOIR

G90 Z($DEPTH[0] + $LDEPTH[0] * NLAYER *-1) F1 ; MOVE TO THE DESIRED CHANNEL DEPTH FROM THE SURFACE

FOR $LZERO = 0 TO (CURVERADIUS/DELTAY - 1) ; LOOP FOR NUMBER OF SETS IN THE Y DIRECTION

$CIRPOS = (CURVERADIUS * 1000) - SQRT((CURVERADIUS * 1000)**2 - (CURVERADIUS * 1000 - DELTAY * $LZERO * 1000)**2) ; NUMBER OF LINES IN THE Z DIRECTION FOR THE CURVE
$CURVELINES = INT($CIRPOS)
G91 Z($CURVELINES * -1 / 1000) F10 ; MOVE TO THE BOTTOM OF THE TAPER CURVE

FOR $LFIVE = 0 TO ($CURVELINES/($LDEPTH[0] * 1000) - 1) ; LOOP FOR NUMBER OF LINES IN THE Z DIRECTION

G91 Z($LDEPTH[0]) F1 ; MOVE UP ONE LINE IN THE Z DIRECTION
G91 X((RLENGTH - 2 * DRT * $LZERO) * $XFLAG) F$SPEED[0] ; SCANNING ONE LINE IN X DIRECTION
$XFLAG = $XFLAG * -1 ; CHANGE DIRECTION

NEXT $LFIVE

G91 X(DRT * $XFLAG) Y(DELTAY) F10 ; MOVE TO THE NEXT SET
G90 Z($DEPTH[0] + $LDEPTH[0] * NLAYER *-1) F1 ; MOVE BACK TO THE DESIRED CHANNEL DEPTH

NEXT $LZERO

;***MAKING A CURVED TOP AT THE TAPER***

G90 Z2 F10 ; MOVE ABOVE THE FOCAL PLANE
G90 UU18.771 E1 ; SET THE POWER FOR THE CURVED TAPER CHANNEL TOP
G90 X0 Y0 F10 ; MOVE BACK TO THE REFERENCE POSITION FOR THE LAST RESERVOIR

G90 Z($DEPTH[0] + NLAYER * $LDEPTH[0] + $LDEPTH[0] * NLAYER *-1) F1 ; MOVE TO THE DESIRED CHANNEL DEPTH FROM THE SURFACE

FOR $LZERO = 0 TO (CURVERADIUS2/DELTAY - 1) ; LOOP FOR NUMBER OF SETS IN THE Y DIRECTION

$CIRPOS = (CURVERADIUS2 * 1000) - SQRT((CURVERADIUS2 * 1000)**2 - (CURVERADIUS2 * 1000 - DELTAY * $LZERO * 1000)**2) ; NUMBER OF LINES IN THE Z DIRECTION FOR THE CURVE
$CURVELINES = INT($CIRPOS)
G91 Z($CURVELINES / 1000) F10 ; MOVE TO THE TOP OF THE TAPER CURVE

FOR $LFIVE = 0 TO ($CURVELINES/($LDEPTH[0] * 1000) - 1) ; LOOP FOR NUMBER OF LINES IN THE Z DIRECTION

G91 Z($LDEPTH[0] * -1) F1 ; MOVE DOWN ONE LINE IN THE Z DIRECTION
G91 X((RLENGTH - 2 * DRT * $LZERO) * $XFLAG) F$SPEED[0] ; SCANNING ONE LINE IN X DIRECTION
$XFLAG = $XFLAG * -1 ; CHANGE DIRECTION

NEXT $LFIVE

G91 X(DRT * $XFLAG) Y(DELTAY) F10 ; MOVE TO THE NEXT SET
G90 Z($DEPTH[0] + NLAYER * $LDEPTH[0] + $LDEPTH[0] * NLAYER *-1) F1 ; MOVE BACK TO THE DESIRED CHANNEL DEPTH

NEXT $LZERO

;***MAKING RESERVOIR 3 (WASTE RESERVOIR)***

$YFLAG = -1
$XFLAG = -1
G90 Z2 F10 ; MOVE ABOVE THE FOCAL PLANE
G90 X0 F10 ; MOVE BACK TO THE REFERENCE X POSITION
G90 Y(LENGTH2 * -1 - RWIDTH) ; MOVE TO THE RESERVOIR ON THE OTHER END
G92 X0 Y0 ; SET A NEW REFERENCE POINT

FOR $LTEN = 0 TO ((RHEIGHT / 0.1)-1) ; LOOP FOR NUMBER OF LAYERS IN THE OPTICAL AXIS DIRECTION
G90 Z2 F10 ; MOVE ABOVE THE FOCAL PLANE
G90 X0 Y0 F10 ; MOVE BACK TO THE ORIGINAL REFERENCE POINT
G90 Z((RHEIGHT * -1 + $LTEN * 0.1)/1.4) F1 ; MOVE TO THE BOTTOM OF THE LAYER

FOR $LELEVEN = 0 TO (RLENGTH/DRT) ; LOOP FOR NUMBER OF LINES AT THE BOTTOM LAYER
G91 Y(RWIDTH * $YFLAG) F$SPEED[0] ; SCAN ONE LINE IN THE Y-DIRECTION
G91 X(DRT * $XFLAG) F1 ; MOVE TO THE NEXT LINE
$YFLAG = $YFLAG * -1 ; CHANGE DIRECTION

NEXT $LELEVEN

G90 LINEAR Z2 F100 ; MOVE ABOVE THE FOCAL PLANE
G90 X0 Y0 F10 ; MOVE BACK TO THE ORIGINAL POSITION
$YFLAG = -1
$XFLAG = -1

FOR $LEIGHT = 0 TO (RWIDTH/0.1) ; LOOP FOR NUMBER OF LAYERS IN THE WIDTHWISE DIRECTION
G90 Z((RHEIGHT * -1 + $LTEN * 0.1)/1.4) F1 ; MOVE TO THE BOTTOM OF THE LAYER
G90 Y(-0.1 * $LEIGHT) ; MOVE TO THE CORRECT LAYER POSITION IN THE Y-DIRECTION

FOR $LNINE = 0 TO (0.1 / DRL) ; LOOP FOR NUMBER OF LAYERS IN EACH CUBE
G91 X(RLENGTH * $XFLAG) F$SPEED[0] ; SCAN ONE LINE IN X-DIRECTION
$XFLAG = $XFLAG * -1 ; CHANGE DIRECTION
G91 Y(DRT * $YFLAG) F1 ; MOVE TO THE NEXT SHELL
$YFLAG = $YFLAG * -1 ; CHANGE DIRECTION
G91 X(RLENGTH * $XFLAG) F$SPEED[0] ; SCAN ONE LINE BACKWARD IN X-DIRECTION
$XFLAG = $XFLAG * -1 ; CHANGE DIRECTION
G91 Z((DRL)/1.4) F1 ; MOVE UP ONE LAYER
NEXT $LNINE

NEXT $LEIGHT
G90 LINEAR Z2 F100 ; MOVE ABOVE THE FOCAL PLANE
G90 X0 Y0 F10 ; MOVE BACK TO THE ORIGINAL POSITION
$YFLAG = -1
$XFLAG = -1
FOR $LEIGHT = 0 TO (RLENGTH/0.1) ; LOOP FOR NUMBER OF LAYERS IN THE LENGTHWISE DIRECTION
G90 Z((RHEIGHT * -1 + $LTEN * 0.1)/1.4) F1 ; MOVE TO THE BOTTOM OF THE LAYER
G90 X(-0.1 * $LEIGHT) ; MOVE TO THE CORRECT LAYER POSITION IN THE X-DIRECTION
FOR $LNINE = 0 TO (0.1 / DRL) ; LOOP FOR NUMBER OF LAYERS IN EACH CUBE
G91 Y(RWIDTH * $YFLAG) F$SPEED[0] ; SCAN ONE LINE IN X-DIRECTION
$YFLAG = $YFLAG * -1 ; CHANGE DIRECTION
G91 X(DRT * $XFLAG) F1 ; MOVE TO THE NEXT SHELL
$XFLAG = $XFLAG * -1 ; CHANGE DIRECTION
G91 Y(RWIDTH * $YFLAG) F$SPEED[0] ; SCAN ONE LINE BACKWARD IN X-DIRECTION
$YFLAG = $YFLAG * -1 ; CHANGE DIRECTION
G90 Z((DRL)/1.4) F1 ; MOVE UP ONE LAYER
NEXT $LNINE
NEXT $LEIGHT
$YFLAG = -1
$XFLAG = -1
NEXT $LTEN

;***MAKING MICROCHANNEL WITH VERTICAL HOLES***
G90 UU18.771 E1 ;SETTING THE POWER FOR THE OUSIDE WALLS OF THE CHANNELS
$YFLAG = -1
$XFLAG = -1
$XSFT = -0.100

G90 Z2 F10 ; MOVE ABOVE THE FOCAL PLANE
G90 X0 Y(LENGTH + LENGTH2 + RWIDTH + RWIDTH) F10 ; MOVE BACK TO
THE ORIGINAL REFERENCE POINT
G90 X(RLENGTH/2 * -1 + NLINE / 2 * $LINESEP[0]) F10 ; MOVE TO THE MID-
DLE OF THE RESERVOIR
G91 Y(RWIDTH * -1 + CEXTRA) F10 ; MOVE TO THE END OF THE RESERVOIR
AND INTO THE RESERVOIR WITH CEXTRA
G92 X0 Y0 ; CHANGE THE REFERENCE POINT FOR X AND Y AXIS

FOR $LFIVE = 0 TO (CDEPTH -1)
G90 LINEAR Z($DEPTH[$LFIVE]) F1 ; MOVE TO THE DESIRED CHANNEL DEPTH
FROM THE SURFACE
FOR $LZERO = 0 TO (NSPEED -1) ; LOOP LAYER ZERO FOR DIFFERENT SCAN
SPEED
FOR $LSEVEN = 0 TO (HOLESEP -1) ; LOOP LAYER SEVEN FOR DIFFERENT
HOLE SEPARATION DISTANCE
FOR $LONE = 0 TO (NLDEPTH -1) ; LOOP LAYER ONE FOR DIFFERENT LAYER
SEPARATION
FOR $LTWO = 0 TO (NLINESEP -1) ; LOOP LAYER TWO FOR DIFFERENT LINE
SEPARATION IN EACH LAYER
G91 LINEAR Z($LDEPTH[$LONE] * NLAYER *-1) F1 ; MOVE TO THE DESIRED
CHANNEL DEPTH FOR BOTTOM UP SCANNING
FOR $LTHREE = 0 TO (NLAYER) ; LOOP LAYER THREE FOR NUMBER OF
LAYERS IN EACH CHANNEL
FOR $LFOUR = 0 TO (NLINE -1) ; LOOP LAYER FOUR FOR NUMBER OF LINES
IN EACH LAYER
IF(($LTHREE == 0) OR ($LTHREE == NLAYER) OR ($LFOUR == 0))
G90 UU18.771 E1 ; SET THE POWER FOR THE OUTSIDE BOUNDARY OF THE
CHANNEL
ELSE
G90 UU18.771 E1 ; SET THE POWER FOR THE INSIDE OF THE CHANNEL
ENDIF
Appendix A. G-code: Particle Counting Cytometer

G91 LINEAR Y((LENGTH + 2 * CEXTRA) * $YFLAG) F($SPEED[$LZERO]); SINGLE TRACK IN Y-DIRECTION

$YFLAG = $YFLAG * (-1); CHANGE DIRECTION

G91 LINEAR X($LINESEP[$LTWO] * $XFLAG) F1; SHIFT TO THE NEXT LINE NEXT $LFOUR

G90 UU18.771 E1; SET THE POWER FOR THE OUTSIDE BOUNDARY OF THE CHANNEL

G91 LINEAR Y((LENGTH + 2 * CEXTRA)* $YFLAG) F($SPEED[$LZERO]); AN EXTRA SINGLE TRACK FOR 5-BY-5 CHANNEL

$XFLAG = $XFLAG * (-1)

$YFLAG = $YFLAG * (-1)

IF($LTHREE < NLAYER)
G91 LINEAR Z($LDEPTH[$LONE]) F1; SHIFT TO THE NEXT LAYER WITH BOTTOM UP
ENDIF

NEXT $LTHREE

;***FIRST SET OF VERTICAL ACCESSS PORTS***

$YFLAG = -1

G90 LINEAR Z(ABOVE) F1; SHIFT ABOVE THE SURFACE

G90 LINEAR Y0 F2; MOVE BACK TO THE ORIGINAL Y POSITION

G91 LINEAR X($LINESEP[$LTWO] * $XFLAG * 5) F1; SHIFT BACK TO THE SIDE MIDDLE OF THE CHANNEL IN X-DIRECTION

$COUNTER = LENGTH/$HSEP[$LSEVEN]; COUNTER EQUALS THE LENGTH OF THE CHANNEL

G90 UU16.147 E1; SET THE POWER FOR THE VERTICAL ACCESS PORTS

FOR $LSIX = 0 TO ($COUNTER - 1); LOOP FOR NUMBER OF VERTICAL ACCESS PORTS

G91 LINEAR Y($HSEP[$LSEVEN] * $YFLAG) F1; SHIFT TO THE NEXT VERTICAL HOLE

IF ($LSIX != 0) OR ($LSIX != 1) OR ($LSIX != 2) OR ($LSIX != 3) OR ($LSIX != 4) OR ($LSIX != 5) OR ($LSIX != ($COUNTER - 1)) OR ($LSIX != ($COUNTER - 2)) OR ($LSIX != ($COUNTER - 3)) OR ($LSIX != ($COUNTER - 4)) OR ($LSIX != ($COUNTER - 5)) OR ($LSIX != ($COUNTER - 6))

G91 LINEAR Z(ABOVE * -1 + $DEPTH[0] + OVERSHOOT * -1) F1; VERTICAL TRACK DOWNWARD

G91 LINEAR Z(ABOVE + $DEPTH[0] * -1 + OVERSHOOT) F1; VERTICAL
Appendix A. G-code: Particle Counting Cytometer

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TRACK UPWARD (DOUBLE PASSED)

ENDIF

NEXT $LSIX

G90 LINEAR Z($DEPTH[$LFIVE]) F1 ; SHIFT BACK TO THE DESIGNED CHANNEL DEPTH

;***REMAINING SETS OF VERTICAL ACCESS PORTS***

FOR $LTWELVE = 0 TO (NLINE/10 -2) ; LOOP FOR THE REMAINING SETS OF VERTICAL ACCESS PORTS

G90 LINEAR Z(ABOVE$) F1 ; SHIFT ABOVE THE SURFACE
G91 LINEAR X($LINESEP[$LTWO] * $XFLAG * 10) F1 ; SHIFT TO THE OTHER SIDE MIDDLE OF THE CHANNEL IN X-DIRECTION
G90 LINEAR Y0 F2 ; MOVE BACK TO THE OTHER SIDE
$COUNTER = LENGTH/$HSEP[$LSEVEN] ; COUNTER EQUALS THE LENGTH OF THE CHANNEL

FOR $LSIX = 0 TO ($COUNTER - 1)

G91 LINEAR Y($HSEP[$LSEVEN] * $YFLAG) F1 ; SHIFT TO THE NEXT VERTICAL HOLE

IF ($LSIX != 0) OR ($LSIX != 1) OR ($LSIX != 2) OR ($LSIX != 3) OR ($LSIX != 4) OR ($LSIX != 5) OR ($LSIX != ($COUNTER - 1)) OR ($LSIX != ($COUNTER - 2)) OR ($LSIX != ($COUNTER - 3)) OR ($LSIX != ($COUNTER - 4)) OR ($LSIX != ($COUNTER - 5)) OR ($LSIX != ($COUNTER - 6))

G91 LINEAR Z(Above$ * -1 + $DEPTH[0] + OVERSHOOT * -1) F1 ; VERTICAL TRACK DOWNWARD
G91 LINEAR Z(Above$ + $DEPTH[0] * -1 + OVERSHOOT) F1 ; VERTICAL TRACK UPWARD (DOUBLE PASSED)

ENDIF

NEXT $LSIX

G90 LINEAR Z($DEPTH[$LFIVE]) F1 ; SHIFT BACK TO THE DESIGNED CHANNEL DEPTH

NEXT $LTWELVE
NEXT $LTWO

NEXT $LONE

NEXT $LSEVEN

NEXT $LZERO

NEXT $LFIVE

; ***MAKING THE TAPERED SECTION ON THE FIRST CHANNEL***

G90 UU18.771 E1 ; SET THE POWER FOR OUTSIDE BOUNDARY OF THE CHANNEL

$XSFT = -0.100

$XFLAG = 1

$YFLAG = -1

G90 Z2 F10 ; MOVE ABOVE THE FOCAL PLANE

G90 X(NLINE * $LINESEP[0] * -1) F10 ; MOVE TO THE SIDE OF THE CHANNEL

G90 Y((LENGTH - TAPERLENGTH + CEXTRA) * -1) F10 ; MOVE TO THE END OF THE FIRST CHANNEL

G92 X0 Y0 ; CHANGE THE REFERENCE POINT FOR X AND Y AXIS

FOR $LFIVE = 0 TO (CDEPTH -1)

G90 Z2 F10 ; MOVE ABOVE THE FOCAL PLANE

G90 X(NLINE * $LINESEP[0] * -1) F10 ; MOVE TO THE SIDE OF THE CHANNEL

G90 Y((LENGTH - TAPERLENGTH + CEXTRA) * -1) F10 ; MOVE TO THE END OF THE FIRST CHANNEL

G92 X0 Y0 ; CHANGE THE REFERENCE POINT FOR X AND Y AXIS

FOR $LZERO = 0 TO (NSPEED -1) ; LOOP LAYER ZERO FOR DIFFERENT SCAN SPEED

FOR $LSEVEN = 0 TO (HOLESEP -1) ; LOOP LAYER SEVEN FOR DIFFERENT HOLE SEPARATION DISTANCE

FOR $LONE = 0 TO (NLDEPTH -1) ; LOOP LAYER ONE FOR DIFFERENT LAYER SEPARATION

FOR $LTWO = 0 TO (NLINESEP -1) ; LOOP LAYER TWO FOR DIFFERENT LINE SEPARATION IN EACH LAYER

G91 LINEAR Z($LDEPTH[$LONE] * NLAYER *-1) F1 ; MOVE TO THE DESIRED CHANNEL DEPTH FOR BOTTOM UP SCANNING
FOR $LTHREE = 0 TO (NLAYER) ; LOOP LAYER THREE FOR NUMBER OF LAYERS IN EACH CHANNEL

FOR $LFOUR = 0 TO ((TAPERLENGTH / DRT) - 1) ; LOOP LAYER FOUR FOR NUMBER OF LINES IN EACH LAYER

IF(($LTHREE == 0) OR ($LTHREE == NLAYER) OR ($LFOUR == 0))
G90 UU18.771 E1 ; SET POWER FOR THE OUTSIDE BOUNDARY OF THE CHANNEL
ELSE
G90 UU18.771 E1 ; SET POWER FOR THE INSIDE BOUNDARY OF THE CHANNEL
ENDIF

IF($XFLAG == 1)
IF($YFLAG == -1)
G91 LINEAR X(DRT * $XFLAG) Y(DRT * $YFLAG) F($SPEED[$LZERO]) ; MOVE TO THE STARTING POINT OF THE NEXT LINE
G91 LINEAR Y((TAPERLENGTH - DRT * ($LFOUR + 1)) * $YFLAG) F($SPEED[$LZERO]) ; SINGLE TRACK IN NEGATIVE Y-DIRECTION
$YFLAG = $YFLAG * (-1) ; CHANGE DIRECTION
ELSE
G91 LINEAR X(DRT * $XFLAG) F($SPEED[$LZERO]) ; MOVE TO THE STARTING POINT OF THE NEXT LINE
G91 LINEAR Y((DRT * ($LFOUR)) * $YFLAG) F($SPEED[$LZERO]) ; SINGLE TRACK IN POSITIVE Y-DIRECTION
$YFLAG = $YFLAG * (-1) ; CHANGE DIRECTION
ENDIF
ELSE
IF($YFLAG == 1)
G91 LINEAR X(DRT * $XFLAG) Y(DRT * $YFLAG) F($SPEED[$LZERO]) ; MOVE TO THE STARTING POINT OF THE NEXT LINE
$YFLAG = $YFLAG * (-1) ; CHANGE DIRECTION
G91 LINEAR Y((DRT * ($LFOUR)) * $YFLAG) F($SPEED[$LZERO]) ; SINGLE TRACK IN NEGATIVE Y-DIRECTION
ELSE
G91 LINEAR X(DRT * $XFLAG) F($SPEED[$LZERO]) ; MOVE TO THE STARTING POINT OF THE NEXT LINE
$YFLAG = $YFLAG * (-1) ; CHANGE DIRECTION
G91 LINEAR Y((DRT * ($LFOUR)) * $YFLAG) F($SPEED[$LZERO]) ; SINGLE TRACK IN NEGATIVE Y-DIRECTION
TRACK IN THE POSITIVE Y-DIRECTION
ENDIF
ENDIF
NEXT $LFOUR
G90 UU18.771 E1 ; SET POWER FOR THE OUTSIDE BOUNDARY OF THE CHANNEL
$XFLAG = $XFLAG * (-1)
$YFLAG = $YFLAG * (-1)
IF($LTHREE < NLAYER)
G91 LINEAR Z($LDEPTH[$LONE]) F1 ; SHIFT TO THE NEXT LAYER WITH BOTTOM UP
ENDIF
NEXT $LTHREE
NEXT $LTWO
NEXT $LONE
NEXT $LSEVEN
NEXT $LZERO
NEXT $LFIVE
;***MAKING THE SECOND TAPERED SECTION ON THE FIRST CHANNEL***
G90 UU18.771 E1 ; SET POWER FOR THE OUTSIDE BOUNDARY OF THE CHANNELS
$XSFT = -0.100
$XFLAG = -1
$YFLAG = -1
G90 Z2 F10 ; MOVE ABOVE THE FOCAL PLANE
G90 X(NLINE * $LINESEP[0]) Y0 F10 ; MOVE TO THE LEFT SIDE OF THE CHANNEL
G92 X0 Y0 ; CHANGE THE REFERENCE POINT FOR X AND Y AXIS
FOR \$LFIVE = 0 TO (CDEPTH -1) ; LOOP FOR NUMBER OF DIFFERENT CHANNEL DEPTH

G90 LINEAR Z($DEPTH[$LFIVE]) F1 ; MOVE TO THE DESIRED CHANNEL DEPTH FROM THE SURFACE

FOR \$LZERO = 0 TO (NSPEED -1) ; LOOP LAYER ZERO FOR DIFFERENT SCAN SPEED

FOR \$LSEVEN = 0 TO (HOLESEP -1) ; LOOP LAYER SEVEN FOR HOLE SEPARATION DISTANCE

FOR \$LONE = 0 TO (NLDEPTH -1) ; LOOP LAYER ONE FOR DIFFERENT LAYER SEPARATION

FOR \$LTWO = 0 TO (NLINESEP -1) ; LOOP LAYER TWO FOR DIFFERENT LINE SEPARATION IN EACH LAYER

G91 LINEAR Z($LDEPTH[$LONE] * NLAYER *-1) F1 ; MOVE TO THE DESIRED CHANNEL DEPTH FOR BOTTOM UP SCANNING

FOR \$LTHREE = 0 TO (NLAYER) ; LOOP LAYER THREE FOR NUMBER OF LAYERS IN EACH CHANNEL

FOR \$LFOUR = 0 TO ((TAPERLENGTH / DRT) -1) ; LOOP LAYER FOUR FOR NUMBER OF LINES IN EACH LAYER

IF(($LTHREE == 0) OR ($LTHREE == NLAYER) OR ($LFOUR == 0))

G90 UU18.771 E1 ; SET POWER FOR THE OUTSIDE BOUNDARY OF THE CHANNEL

ELSE

G90 UU18.771 E1 ; SET POWER FOR THE INSIDE BOUNDARY OF THE CHANNEL

ENDIF

IF($XFLAG == -1)

IF($YFLAG == -1)

G91 LINEAR X(DRT * $XFLAG) Y(DRT * $YFLAG) F($SPEED[$LZERO]) ; MOVE TO THE STARTING POINT OF THE NEXT LINE

G91 LINEAR Y((TAPERLENGTH - DRT * ($LFOUR + 1)) * $YFLAG ) F($SPEED[$LZERO]) ; SINGLE TRACK IN NEGATIVE Y-DIRECTION

$YFLAG = $YFLAG * (-1) ; CHANGE DIRECTION
ELSE

G91 LINEAR X(DRT * $XFLAG) F($SPEED[$LZERO]) ;MOVE TO THE STARTING
POINT OF THE NEXT LINE
G91 LINEAR Y((TAPERLENGTH - DRT * ($LFOUR + 1)) * $YFLAG)
F($SPEED[$LZERO]) ;SINGLE TRACK IN POSITIVE Y-DIRECTION
$YFLAG = $YFLAG * (-1) ;CHANGE DIRECTION

ENDIF

ELSE

IF($YFLAG == 1)

G91 LINEAR X(DRT * $XFLAG) Y(DRT * $YFLAG) F($SPEED[$LZERO]) ;MOVE
TO THE STARTING POINT OF THE NEXT LINE
$YFLAG = $YFLAG * (-1) ;CHANGE DIRECTION
G91 LINEAR Y((DRT * ($LFOUR)) * $YFLAG) F($SPEED[$LZERO]) ;SINGLE TRACK
IN NEGATIVE Y-DIRECTION

ELSE

G91 LINEAR X(DRT * $XFLAG) F($SPEED[$LZERO]) ;MOVE TO THE STARTING
POINT OF THE NEXT LINE
$YFLAG = $YFLAG * (-1) ; CHANGE DIRECTION
G91 LINEAR Y((DRT * ($LFOUR)) * $YFLAG) F($SPEED[$LZERO]) ;SINGLE TRACK
IN THE POSITIVE Y-DIRECTION

ENDIF

ENDIF

NEXT $LFOUR

G90 UU18.771 E1 ;SET POWER FOR THE OUTSIDE BOUNDARY OF THE CHANNEL
$XFLAG = $XFLAG * (-1)
$YFLAG = $YFLAG * (-1)

IF($LTHREE < NLAYER)

G91 LINEAR Z($LDEPTH[$LONE]) F1 ;SHIFT TO THE NEXT LAYER WITH BOTTOM UP
ENDIF

NEXT $LTHREE

NEXT $LTWO

NEXT $LONE

NEXT $LSEVEN

NEXT $LZERO

NEXT $LFIVE

; ***MAKING STOP VALVE CHANNELS WITH VERTICAL ACCESS PORTS***

G90 UU18.771 E1 ; SET POWER FOR THE OUTSIDE BOUNDARY OF THE CHANNEL

$XSFT = -0.100

G90 Z2 F10 ; MOVE ABOVE THE FOCAL PLANE

G90 X0 Y(-1 * TAPERLENGTH + -1 * RWIDTH + CEXTRA) F10 ; MOVE TO THE NEW REFERENCE POINT IN Y-DIRECTION

G90 X(-1 * NLINE / 2 * $LINESEP[0] - NLINE2 / 2 * $LINESEP[0]) F10 ; MOVE TO THE MIDDLE OF THE RESERVOIR

G92 X0 Y0 ; CHANGE THE REFERENCE LOCATION FOR X AND Y AXIS

G90 X(RLENGTHSMALL * INT((RLENGTH/RLENGTHSMALL)/2)) F10 ; MOVE TO THE FIRST SMALL RESERVOIR ON THE RIGHT

G92 X0 ; CHANGE THE REFERENCE LOCATION OF X

FOR $LTHIRTEEN = 0 TO INT((RLENGTH/RLENGTHSMALL)/2) ; LOOP FOR NUMBER OF SMALL CHANNELS

IF ($LTHIRTEEN == 1)

G90 Z2 F10
G90 X(RLENGTHSMALL * -1 * 2) Y0 F10 ; MOVE TO THE NEXT SMALL CHANNEL POSITION
G92 X0 Y0 ; SET THE REFERENCE LOCATION TO THE NEW SMALL CHANNEL

ELSEIF ($LTHIRTEEN == 2)
Appendix A. G-code: Particle Counting Cytometer

G90 Z2 F10
G90 X(RLENGTHSMALL * -1 * 2) Y0 F10 ; MOVE TO THE NEXT SMALL CHANNEL POSITION
G92 X0 Y0 ; SET THE REFERENCE LOCATION TO THE NEW SMALL CHANNEL
ENDIF
FOR $LFIVE = 0 TO (CDEPTH -1) ; LOOP FOR NUMBER OF DIFFERENT CHANNEL DEPTH
$YFLAG = -1
$XFLAG = 1
G90 LINEAR Z(-1 * CHANNEL2DEPTH) F1 ; MOVE TO THE DESIRED CHANNEL DEPTH FROM THE SURFACE
FOR $LZERO = 0 TO (NSPEED -1) ; LOOP LAYER ZERO FOR DIFFERENT SCAN SPEED
FOR $LSEVEN = 0 TO (HOLESEP -1) ; LOOP LAYER SEVEN FOR DIFFERENT HOLE SEPARATION DISTANCE
FOR $LONE = 0 TO (NLDEPTH -1) ; LOOP LAYER ONE FOR DIFFERENT LAYER SEPARATION
FOR $LTWO = 0 TO (NLINESEP -1) ; LOOP LAYER TWO FOR DIFFERENT LINE SEPARATION IN EACH LAYER
G91 LINEAR Z($LDEPTH[$LONE] * NLAYER2 *-1) F1 ; MOVE TO THE DESIRED CHANNEL DEPTH FOR BOTTOM UP SCANNING
FOR $LTHREE = 0 TO (NLAYER2) ; LOOP LAYER THREE FOR NUMBER OF LAYERS IN EACH CHANNEL
FOR $LFOUR = 0 TO (NLINE2 -1) ; LOOP LAYER FOUR FOR NUMBER OF LINES IN EACH LAYER
IF(($LTHREE == 0) OR ($LTHREE == NLAYER2) OR ($LFOUR == 0))
G90 UU18.771 E1 ; SET POWER FOR THE OUTSIDE BOUNDARY OF THE CHANNEL
ELSE

Appendix A. G-code: Particle Counting Cytometer

G90 UU18.771 E1 ; SET POWER FOR THE INSIDE OF THE CHANNEL

ENDIF

G91 LINEAR Y((LENGTH2 + 2 * CEXTRA) * $YFLAG) F($SPEED[$LZERO]) ; SINGLE TRACK IN Y-DIRECTION
$YFLAG = $YFLAG * (-1) ; CHANGE DIRECTION
G91 LINEAR X($LINESEP[$LTWO] * $XFLAG) F1 ; SHIFT TO THE NEXT LINE

NEXT $LFOUR

G90 UU18.771 E1 ; SET POWER FOR THE OUTSIDE BOUNDARY OF THE CHANNEL
G91 LINEAR Y((LENGTH2 + 2 * CEXTRA) * $YFLAG) F($SPEED[$LZERO]) ; AN EXTRA SINGLE TRACK FOR 5-BY-5 CHANNEL
$XFLAG = $XFLAG * (-1)
$YFLAG = $YFLAG * (-1)

IF($LTHREE < $NLAYER2)

G91 LINEAR Z($LDEPTH[$LONE]) F1 ; SHIFT TO THE NEXT LAYER WITH BOTTOM UP

ENDIF

NEXT $LTHREE

;***SCANNING VERTICAL ACCESS PORTS FOR THE STOP VALVE CHANNEL***

G90 LINEAR Z(ABOVES) F1 ; SHIFT ABOVE THE SURFACE
G91 LINEAR X($LINESEP[$LTWO] * $XFLAG * NLINE2 / 2) F1 ; SHIFT BACK TO THE SIDE MIDDLE OF THE CHANNEL IN X-DIRECTION
$COUNTER = LENGTH2/$HSEP[$LSEVEN] ; COUNTER EQUALS THE LENGTH OF THE CHANNEL

G90 UU16.147 E1 ; SET POWER FOR THE VERTICAL ACCESS PORTS

FOR $LSIX = 0 TO ($COUNTER - 1)

G91 LINEAR Y($HSEP[$LSEVEN] * $YFLAG) F1 ; SHIFT TO THE NEXT VERTICAL HOLE
G91 LINEAR Z(ABOVES * -1 - CHANNEL2DEPTH + OVERSHOOT * -1) F1 ; VERTICAL TRACK DOWNWARD
G91 LINEAR Z(ABOVES + CHANNEL2DEPTH + OVERSHOOT) F1 ; VERTICAL TRACK UPWARD (DOUBLE PASSED)
NEXT $LSIX
NEXT $LTWO
NEXT $LONE
NEXT $LSEVEN
NEXT $LZERO
NEXT $LFIVE
NEXT $LTHIRTEEN
G90 LINEAR Z2 F100 ; MOVE ABOVE THE FOCAL PLANE
G90 UU40 E1 ; TURN THE POWER DOWN
G90 X0 Y0 F10 ; MOVE BACK TO THE ORIGINAL POSITION
M2 ; TERMINATE PROGRAM
Appendix B

G-code: Multi-Level Capillary Electrophoresis Device

The G-code used for controlling the xyz-stages in laser patterning the multi-level capillary electrophoresis device is presented below. There are two similar sets of code. Part 1 patterns two reservoirs with a curved channel that changes depth, whereas Part 2 patterns the remaining three sets of two reservoirs with a curved channel having constant depth. Each of Part 1 and Part 2 is run once to pattern two curved channels side-by-side, and the sample substrate is turned 90° to pattern the rest of the two curved channels by running Part 2.

G-code Part 1:

```
// Title: Multilevel Capillary Electrophoresis Device Part 1
// Author: Stephen Ho
// Date Created: April 27, 2011
// Description: Writes one curved channel that move from a deeper focal plane to a shallower depth with two reservoirs

// Parameter (USER INPUT REQUIRED)

#DEFINE NSPEED 1 ; NUMBER OF SCAN SPEED
#DEFINE NLDEPTH 1 ; NUMBER OF LAYER DEPTH
#DEFINE NLINSEP 1 ; NUMBER OF TRANSVERSE LINE TO LINE SEPARATION
#DEFINE NVERSE 3 ; NUMBER OF SETS OF VERTICAL ACCESS PORT
#DEFINE CDEPTH 2 ; NUMBER OF CHANNEL DEPTH
#DEFINE HOLESEP 1 ; NUMBER OF VERTICAL ACCESS PORT SEPARATION
#DEFINE CHANNEL1 2.5 ; LENGTH OF SHORT STRAIGHT ARM
#DEFINE CHANNEL2 0.2 ; LENGTH OF SHORT SHORT STRAIGHT ARM
#DEFINE CHANNEL3 0.5 ; LENGTH OF THE DIAGONAL ARM
#DEFINE CHANNEL4 4.3 ; LENGTH OF THE REST OF THE STRAIGHT ARM
#DEFINE RADIUS 2.5 ; RADIUS OF CIRCULAR ARC
```
#DEFINE PI 3.14159265358979323846264338327950288419716939937
#DEFINE DEG 2.5 * PI / 180

#DEFINE NLAYER 16 ; NUMBER OF LAYERS
#DEFINE NLINE 30 ; NUMBER OF LINES IN EACH LAYER
#DEFINE ABOVE 0.5 ; THE DISTANCE ABOVE THE SAMPLE SURFACE
#DEFINE OVERSHOOT 0.005 ; THE OVERSHOOT DISTANCE INTO THE CHANNEL
#DEFINE CEXTRA 0.050 ; THE EXTRA CHANNEL LENGTH INTO EACH RESERVOIR
#DEFINE RWIDTH 0.5 ; THE WIDTH OF THE RESERVOIR
#DEFINE RLENGTH 0.5 ; THE LENGTH OF THE RESERVOIR
#DEFINE RHEIGHT 0.2 ; THE HEIGHT OF THE RESERVOIR
#DEFINE NSHELL 2 ; NUMBER OF SHELLS AT EACH BOARDER
#DEFINE DRT 0.002 ; TRANSVERSE SPACING IN RESERVOIR
#DEFINE DRL 0.005 ; LAYER SPACING IN RESERVOIR

DVAR $YSFT $XSFT $LZERO $LONE $LTWO $LTHREE $LFOUR $LFIVE $LSIX $LSEVEN $LEIGHT $LNINE $LTEN $LELEVEN $YFLAG $XFLAG $ZFLAG $COUNTER $LENGTH $RADIUS1 $RADIUS2 $VERSET

METRIC SECONDS

; PRESET PARAMETERS

; HORIZONTAL SEPARATION FOR EACH VERTICAL HOLE
$HSEP[0]=0.1

; SCAN SPEED FOR THROUGH CHANNELS
$SPEED[0]=0.5

; SEPARATION FOR EACH LAYER IN EACH CHANNEL FROM BOTTOM UP
$LDEPTH[0]=0.002

; SEPARATION FOR EACH LINE IN EACH LAYER
$LINESEP[0]=-0.0015
;THE CHANNEL DEPTH (MACHINE) FROM THE SURFACE (MULTIPLY 1.4 TO GET THE CORRECT DEPTH FROM THE SURFACE)

$DEPTH[0]=-0.100
$DEPTH[1]=-0.050

;NUMBER OF LINES TO BE MOVED TO GET TO THE SET OF VERTICAL ACCESS PORTS

$HOLES[0] = 5
$HOLES[1] = 10
$HOLES[2] = 10

;NUMBER OF LINES OR LINE SPACINGS TO SUBTRACT OR ADD TO THE RADIUS

$LINES[0] = 5
$LINES[1] = 15
$LINES[2] = 25

;LENGTH OF THE SAMPLE OF INTEREST

$LENGTH = 2*(CHANNEL1 + RADIUS + RADIUS - NLINE * $LINESEP[0] * -1) + CHANNEL2 + CHANNEL3 + CHANNEL4

$XSFT = 0 ; DEFINE OFFSITE LENGTH ON X-AXIS
$YFLAG = -1
$XFLAG = 1

; ***START OF PROGRAM***
; ***MAKING FIRST RESERVOIR***
G90 LINEAR UU24.642 E1 ; SET POWER FOR THE RESERVOIR
$YFLAG = -1
$XFLAG = -1

FOR $LTEN = 0 TO ((RHEIGHT / 0.1)-1) ; LOOP FOR NUMBER OF LAYERS IN THE OPTICAL AXIS DIRECTION
G90 LINEAR Z2 F10 ; MOVE ABOVE THE FOCAL PLANE
G90 LINEAR X0 Y0 F10 ; MOVE BACK TO THE ORIGINAL REFERENCE POINT
G90 LINEAR Z(RHEIGHT * -1 + $LTEN * 0.1) F1 ; MOVE TO THE BOTTOM LAYER

FOR $LELEVEN = 0 TO (RLENGTH/DRT) ; LOOP FOR NUMBER OF LINES AT THE BOTTOM LAYER
G91 LINEAR Y(RWIDTH * $YFLAG) F$SPEED[0] ; SCAN ONE LINE IN THE Y-DIRECTION
G91 LINEAR X(DRT * $XFLAG) F1 ; MOVE TO THE NEXT LINE
$YFLAG = $YFLAG * -1 ; CHANGE DIRECTION
NEXT $LELEVEN

G90 LINEAR Z2 F100 ; MOVE ABOVE THE FOCAL PLANE
G90 LINEAR X0 Y0 F10 ; MOVE BACK TO THE ORIGINAL POSITION
$YFLAG = -1
$XFLAG = -1

FOR $LEIGHT = 0 TO (RLENGTH/0.1) ; LOOP FOR NUMBER OF LAYERS IN THE WIDTHWISE DIRECTION
G90 LINEAR Z(RHEIGHT * -1 + $LTEN * 0.1) F1 ; MOVE TO THE BOTTOM OF THE LAYER
G90 LINEAR Y(-0.1 * $LEIGHT) ; MOVE TO THE CORRECT LAYER POSITION IN THE Y-DIRECTION
FOR $LNINE = 0 TO (0.1 / DRL) ; LOOP FOR NUMBER OF LAYERS IN EACH CUBE
G91 LINEAR X(RLENGTH * $XFLAG) F$SPEED[0] ; SCAN A X-DIRECTION LINE
$XFLAG = $XFLAG * -1 ; CHANGE DIRECTION
G91 LINEAR Y(DRT * $YFLAG) F1 ; MOVE TO THE NEXT SHELL
$YFLAG = $YFLAG * -1 ; CHANGE DIRECTION
G91 LINEAR X(RLENGTH * $XFLAG) F$SPEED[0] ; SCAN ONE LINE BACKWARD IN X-DIRECTION
$XFLAG = $XFLAG * -1 ; CHANGE DIRECTION
G91 LINEAR Z(DRL) F1 ; MOVE UP ONE LAYER

NEXT $LNINE

NEXT $LEIGHT

G90 LINEAR Z2 F100 ; MOVE ABOVE THE FOCAL PLANE
G90 LINEAR X0 Y0 F10 ; MOVE BACK TO THE ORIGINAL POSITION
$YFLAG = -1
$XFLAG = -1

FOR $LEIGHT = 0 TO (RWIDTH/0.1) ; LOOP FOR NUMBER OF LAYERS IN THE LENGTHWISE DIRECTION
G90 LINEAR Z(RHEIGHT * -1 + $LTEN * 0.1) F1 ; MOVE TO THE BOTTOM OF
THE LAYER
G90 LINEAR X(-0.1 * $LEIGHT) ; MOVE TO THE CORRECT LAYER POSITION
IN THE X-DIRECTION
FOR $LNINE = 0 TO (0.1 / DRL) ; LOOP FOR NUMBER OF LAYERS IN EACH
CUBE
G91 LINEAR Y(RWIDTH * $YFLAG) F$SPEED[0] ; SCAN A X-DIRECTION LINE
G91 LINEAR X(DRT * $XFLAG) F1 ; MOVE TO THE NEXT SHELL
$XFLAG = $XFLAG * -1 ; CHANGE DIRECTION
$YFLAG = $YFLAG * -1 ; CHANGE DIRECTION
G91 LINEAR Y(RWIDTH * $YFLAG) F$SPEED[0] ; SCAN ONE LINE BACKWARD
IN X-DIRECTION
$YFLAG = $YFLAG *-1 ; CHANGE DIRECTION
G91 LINEAR Z(DRL) F1 ; MOVE UP ONE LAYER
NEXT $LNINE
NEXT $LEIGHT
NEXT $LTEN
G90 LINEAR Z2 F10 ; MOVE ABOVE THE FOCAL PLANE
G90 LINEAR X0 Y0 F10 ; MOVE BACK TO THE ORIGINAL REFERENCE POINT
; ***MAKING THE SECOND RESERVOIR***
$YFLAG = -1
$XFLAG = -1
G90 LINEAR Y($LENGTH * -1 - RWIDTH) ; MOVE TO THE RESERVOIR ON THE
OTHER END
G92 X0 Y0 ; SET A NEW REFERENCE POINT
FOR $LTEN = 0 TO ((RHEIGHT / 0.1)-1) ; LOOP FOR NUMBER OF LAYERS IN
THE OPTICAL AXIS DIRECTION
G90 LINEAR Z2 F10 ; MOVE ABOVE THE FOCAL PLANE
G90 LINEAR X0 Y0 F10 ; MOVE BACK TO THE ORIGINAL REFERENCE POINT
G90 LINEAR Z(RHEIGHT * -1 + $LTEN * 0.1) F1 ; MOVE TO THE BOTTOM
LAYER
FOR $LELEVEN = 0 TO (RLENGTH/DRT) ; LOOP FOR NUMBER OF LINES AT
THE BOTTOM LAYER
G91 LINEAR Y(RWIDTH * $YFLAG) F$SPEED[0] ; SCAN ONE LINE IN THE Y-DIRECTION
G91 LINEAR X(DRT * $XFLAG) F1 ; MOVE TO THE NEXT LINE
$YFLAG = $YFLAG * -1 ; CHANGE DIRECTION

NEXT $LELEVEN

G90 LINEAR Z2 F100 ; MOVE ABOVE THE FOCAL PLANE
G90 LINEAR X0 Y0 F10 ; MOVE BACK TO THE ORIGINAL POSITION
$YFLAG = -1
$XFLAG = -1

FOR $LEIGHT = 0 TO (RLENGTH/0.1) ; LOOP FOR NUMBER OF LAYERS IN THE WIDTHWISE DIRECTION
G90 LINEAR Z(RHEIGHT * -1 + $LTEN * 0.1) F1 ; MOVE TO THE BOTTOM OF THE LAYER
G90 LINEAR Y(-0.1 * $LEIGHT) ; MOVE TO THE CORRECT LAYER POSITION IN THE Y-DIRECTION
FOR $LNINE = 0 TO (0.1 / DRL) ; LOOP FOR NUMBER OF LAYERS IN EACH CUBE
G91 LINEAR X(RLENGTH * $XFLAG) F$SPEED[0] ; SCAN A X-DIRECTION LINE
$XFLAG = $XFLAG * -1 ; CHANGE DIRECTION
G91 LINEAR Y(DRT * $YFLAG) F1 ; MOVE TO THE NEXT SHELL
$YFLAG = $YFLAG * -1 ; CHANGE DIRECTION
G91 LINEAR X(RLENGTH * $XFLAG) F$SPEED[0] ; SCAN ONE LINE BACKWARD IN X-DIRECTION
$XFLAG = $XFLAG *-1 ; CHANGE DIRECTION
G91 LINEAR Z(DRL) F1 ; MOVE UP ONE LAYER

NEXT $LNINE

NEXT $LEIGHT

G90 LINEAR Z2 F100 ; MOVE ABOVE THE FOCAL PLANE
G90 LINEAR X0 Y0 F10 ; MOVE BACK TO THE ORIGINAL POSITION
$YFLAG = -1
$XFLAG = -1

FOR $LEIGHT = 0 TO (RWIDTH/0.1) ; LOOP FOR NUMBER OF LAYERS IN THE LENGTHWISE DIRECTION
G90 LINEAR Z(RHEIGHT * -1 + $LTEN * 0.1) F1 ; MOVE TO THE BOTTOM OF
THE LAYER

G90 LINEAR X(-0.1 * $LEIGHT) ; MOVE TO THE CORRECT LAYER POSITION IN THE X-DIRECTION

FOR $LNINE = 0 TO (0.1 / DRL) ; LOOP FOR NUMBER OF LAYERS IN EACH CUBE

G91 LINEAR Y(RWIDTH * $YFLAG) F$SPEED[0] ; SCAN A X-DIRECTION LINE
$YFLAG = $YFLAG * -1 ; CHANGE DIRECTION
G91 LINEAR X(DRT * $XFLAG) F1 ; MOVE TO THE NEXT SHELL
$XFLAG = $XFLAG * -1 ; CHANGE DIRECTION
G91 LINEAR Y(RWIDTH * $YFLAG) F$SPEED[0] ; SCAN ONE LINE BACKWARD IN X-DIRECTION
$YFLAG = $YFLAG * -1 ; CHANGE DIRECTION
G91 LINEAR Z(DRL) F1 ; MOVE UP ONE LAYER

NEXT $LNINE

NEXT $LEIGHT

NEXT $LTEN

;***MAKING MICROCHANNEL WITH VERTICAL HOLES*** $XFLAG = -1
$YFLAG = -1
$ZFLAG = -1

G90 LINEAR UU28.842 E1 ; SET POWER FOR OUTSIDE BOUNDARY OF THE CHANNEL

$XSFT = -0.100

G90 LINEAR Z2 F10 ; MOVE ABOVE THE FOCAL PLANE
G90 LINEAR X0 Y($LENGTH + RWIDTH) F10 ; MOVE BACK TO THE ORIGINAL REFERENCE POINT
G90 LINEAR X(RLENGTH/2 * -1) F10 ; MOVE TO THE MIDDLE OF THE RESERVOIR
G91 LINEAR Y(RWIDTH * -1) F10 ; MOVE TO THE END OF THE RESERVOIR AND INTO THE RESERVOIR WITH CEXTRA
G92 X0 Y0 ; CHANGE THE REFERENCE POINT FOR X AND Y AXIS
G90 LINEAR Z($DEPTH[$LFIVE]) F1 ; MOVE TO THE DESIRED CHANNEL DEPTH FROM THE SURFACE

FOR $LZERO = 0 TO (NSPEED -1) ; LOOP LAYER ZERO FOR DIFFERENT SPEED

FOR $LSEVEN = 0 TO (HOLESEP -1) ; LOOP LAYER SEVEN FOR DIFFERENT
HOLE SEPARATION DISTANCE

FOR $LONE = 0 TO (NLDEPTH -1) ; LOOP LAYER ONE FOR DIFFERENT LAYER SEPARATION

FOR $LTWO = 0 TO (NLINESEP -1) ; LOOP LAYER TWO FOR DIFFERENT LINE SEPARATION IN EACH LAYER

G91 LINEAR Z($LDEPTH[$LONE] * NLAYER *-1) F1 ; MOVE TO THE DESIRED CHANNEL DEPTH FOR BOTTOM UP SCANNING

FOR $LTHREE = 0 TO (NLAYER) ; LOOP LAYER THREE FOR NUMBER OF LAYERS IN EACH CHANNEL

$RADIUS1 = RADIUS ; DEFINE THE OUTER RADIUS
$RADIUS2 = RADIUS - (NLINE * $LINESEP[0] * -1) ; DEFINE THE INNER RADIUS

FOR $LFOUR = 0 TO (NLINE -1) ; LOOP LAYER FOUR FOR NUMBER OF LINES IN EACH LAYER

IF(($LTHREE == 0) OR ($LTHREE == NLAYER) OR ($LFOUR == 0))
G90 LINEAR UU28.842 E1 ; SET POWER FOR THE OUTSIDE BOUNDARY OF THE CHANNEL
ELSE
G90 LINEAR UU28.842 E1 ; SET POWER FOR THE INSIDE OF THE CHANNEL
ENDIF

VELOCITY ON
M146

G91 LINEAR Y(CHANNEL1 * $YFLAG) F($SPEED[$LZERO]) ; FIRST SECTION SHORT CHANNEL ARM

IF ($XFLAG == -1) ; MOVING TOWARD RIGHT
IF ($YFLAG == -1) ; MOVING DOWNWARD
CCW P180 Q270 R($RADIUS1) F($SPEED[$LZERO]) ; SECOND SECTION CIRCULAR ARC
CW P90 Q0 R($RADIUS2) F($SPEED[$LZERO]) ; THIRD SECTION CIRCULAR ARC
ELSE
CW P180 Q90 R($RADIUS1) F($SPEED[$LZERO])
CCW P270 Q0 R($RADIUS2) F($SPEED[$LZERO])
ENDIF
ELSE ; MOVING TOWARD LEFT

IF ($YFLAG == 1) ; MOVING UPWARD
CW P180 Q90 R($RADIUS2) F($SPEED[$LZERO])
CCW P270 Q0 R($RADIUS1) F($SPEED[$LZERO])
ELSE
CCW P180 Q270 R($RADIUS2) F($SPEED[$LZERO])
CW P90 Q0 R($RADIUS1) F($SPEED[$LZERO])
ENDIF
ENDIF

G91 LINEAR Y(CHANNEL2 * $YFLAG) F($SPEED[$LZERO]) ; FOURTH SECTION
LONG CHANNEL ARM
G91 LINEAR Y(CHANNEL3 * $YFLAG) Z(($DEPTH[0] - $DEPTH[1]) * $ZFLAG)
F($SPEED[$LZERO]) ; FIFTH CHANNEL GOING UP
G91 LINEAR Y(CHANNEL4 * $YFLAG) F($SPEED[$LZERO]) ; SIXTH CHANNEL
STRAIGHT

IF ($XFLAG == -1)

IF ($YFLAG == -1)
CW P0 Q270 R($RADIUS2) F($SPEED[$LZERO]) ; SEVENTH SECTION CIRCULAR
ARC
CCW P90 Q180 R($RADIUS1) F($SPEED[$LZERO]) ; EIGHTH SECTION CIRCULAR
ARC
ELSE
CCW P0 Q90 R($RADIUS2) F($SPEED[$LZERO])
CW P270 Q180 R($RADIUS1) F($SPEED[$LZERO])
ENDIF
ELSE

IF ($YFLAG == 1)
CCW P0 Q90 R($RADIUS1) F($SPEED[$LZERO])
CW P270 Q180 R($RADIUS2) F($SPEED[$LZERO])
ELSE
CW P0 Q270 R($RADIUS1) F($SPEED[$LZERO])
CCW P90 Q180 R($RADIUS2) F($SPEED[$LZERO])
ENDIF
ENDIF

G91 LINEAR Y(CHANNEL1 * $YFLAG) F($SPEED[$LZERO]) ; SEVENTH SECTION
SHORT CHANNEL ARM
$
YFLAG = YFLAG * (-1) ; CHANGE DIRECTION
$
ZFLAG = ZFLAG * (-1)

$RADIUS1 = RADIUS1 - (LINESEP[LTWO] * -1) ; DECREASE THE OUTER RADIUS
$RADIUS2 = RADIUS2 + (LINESEP[LTWO] * -1) ; INCREASE THE INNER RADIUS

G91 LINEAR X(LINESEP[LTWO] * $XFLAG) F1 ; SHIFT TO THE NEXT LINE
VELOCITY OFF

NEXT $LFOUR

G90 LINEAR UU28.842 E1 ; SET POWER FOR THE OUTSIDE BOUNDARY OF THE CHANNEL

VELOCITY ON
M146

G91 LINEAR Y(CHANNEL1 * $YFLAG) F($SPEED[LZERO]) ; FIRST SECTION SHORT CHANNEL ARM

IF ($XFLAG == -1) ; MOVING TOWARD RIGHT

IF ($YFLAG == -1) ; MOVING DOWNWARD
CCW P180 Q270 R($RADIUS1) F($SPEED[LZERO]) ; SECOND SECTION CIRCULAR ARC
CW P90 Q0 R($RADIUS2) F($SPEED[LZERO]) ; THIRD SECTION CIRCULAR ARC
ELSE
CW P180 Q90 R($RADIUS1) F($SPEED[LZERO])
CCW P270 Q0 R($RADIUS2) F($SPEED[LZERO])
ENDIF

ELSE ; MOVING TOWARD LEFT

IF ($YFLAG == 1) ; MOVING UPWARD
CW P180 Q90 R($RADIUS2) F($SPEED[LZERO])
CCW P270 Q0 R($RADIUS1) F($SPEED[LZERO])
ELSE
CCW P180 Q270 R($RADIUS2) F($SPEED[LZERO])
CW P90 Q0 R($RADIUS1) F($SPEED[LZERO])
ENDIF
ENDIF

G91 LINEAR Y(CHANNEL2 * $YFLAG) F($SPEED[$LZERO]) ; FOURTH SECTION LONG CHANNEL ARM
G91 LINEAR Y(CHANNEL3 * $YFLAG) Z(($DEPTH[0] - $DEPTH[1]) * $ZFLAG) F($SPEED[$LZERO]) ; FIFTH CHANNEL GOING UP
G91 LINEAR Y(CHANNEL4 * $YFLAG) F($SPEED[$LZERO]) ; SIXTH CHANNEL STRAIGHT

IF ($XFLAG == -1)

IF ($YFLAG == -1)
CW P0 Q270 R($RADIUS2) F($SPEED[$LZERO]) ; SEVENTH SECTION CIRCULAR ARC
CCW P90 Q180 R($RADIUS1) F($SPEED[$LZERO]) ; EIGHTH SECTION CIRCULAR ARC
ELSE
CW P0 Q90 R($RADIUS2) F($SPEED[$LZERO])
CW P270 Q180 R($RADIUS1) F($SPEED[$LZERO])
ENDIF

ELSE

IF ($YFLAG == 1)
CCW P0 Q90 R($RADIUS1) F($SPEED[$LZERO])
CW P270 Q180 R($RADIUS2) F($SPEED[$LZERO])
ELSE
CW P0 Q270 R($RADIUS1) F($SPEED[$LZERO])
CCW P90 Q180 R($RADIUS2) F($SPEED[$LZERO])
ENDIF

ENDIF

G91 LINEAR Y(CHANNEL1 * $YFLAG) F($SPEED[$LZERO]) ; SEVENTH SECTION SHORT CHANNEL ARM

$XFLAG = $XFLAG * (-1)
$YFLAG = $YFLAG * (-1)
$ZFLAG = $ZFLAG * (-1)

VELOCITY OFF

IF($LTHREE < NLAYER)
G91 LINEAR Z($LDEPTH[$LONE]) F1 ; SHIFT TO THE NEXT LAYER WITH BOT-
TOM UP
ENDIF
NEXT $LTHREE

;***SETS OF VERTICAL HOLES***
G90 X0 F1

FOR $VERSET = 0 TO (NVERSET - 1) ; LOOP THE NUMBER OF SETS OF VERTICAL ACCESS PORTS

$XFLAG = -1
$YFLAG = -1
$RADIUS1 = RADIUS - ($LINES[$VERSET] * $LINESEP[0] * -1) ; DEFINE THE OUTER RADIUS
$RADIUS2 = RADIUS - (NLINE - $LINES[$VERSET]) * $LINESEP[0] * -1 ; DEFINE THE INNER RADIUS

G90 LINEAR Z(ABOVES) F1 ; SHIFT ABOVE THE SURFACE
G91 LINEAR X($LINESEP[$LTWO] * $XFLAG * $HOLES[$VERSET]) F1 ; SHIFT BACK TO THE SIDE MIDDLE OF THE CHANNEL IN X-DIRECTION
G91 LINEAR Y($LENGTH) F2 ; SHIFT BACK TO THE FIRST RESERVOIR SIDE

$COUNTER = CHANNEL1/$HSEP[$LSEVEN] ; COUNTER EQUALS THE LENGTH OF THE FIRST SECTION SHORT CHANNEL

G90 LINEAR UU24.642 E1 ; SET POWER FOR VERTICAL ACCESS PORT

FOR $LSIX = 0 TO ($COUNTER - 1)

G91 LINEAR Z(ABOVES * -1 + $DEPTH[0] + OVERSHOOT * -1) F1 ; VERTICAL TRACK DOWNWARD
G91 LINEAR Z(ABOVES + $DEPTH[0] * -1 + OVERSHOOT) F1 ; VERTICAL TRACK UPWARD (DOUBLE PASSED)
G91 LINEAR Y($HSEP[$LSEVEN] * $YFLAG) F1 ; SHIFT TO THE NEXT VERTICAL ACCESS PORT

NEXT $LSIX

G92 X0 Y0 ; SET A NEW REFERENCE POINT

$COUNTER = 90/2.5 ; COUNTER EQUALS THE NUMBER OF HOLES FOR EVERY 2.5 DEGREES IN SECOND SECTION
FOR $LSIX = 0 TO ($COUNTER - 1)

G91 LINEAR Z(ABOVES * -1 + $DEPTH[0] + OVERSHOOT * -1) F1 ; VERTICAL TRACK DOWNWARD
G91 LINEAR Z(ABOVES + $DEPTH[0] * -1 + OVERSHOOT) F1 ; VERTICAL TRACK UPWARD (DOUBLE PASSED)
G90 LINEAR X($RADIUS1 - $RADIUS1 * COS(DEG*($LSIX + 1))) Y($RADIUS1 * SIN(DEG*($LSIX + 1)) * $YFLAG) F1 ; SHIFT TO THE NEXT VERTICAL ACCESS PORT

NEXT $LSIX

G92 X0 Y0 ; SET A NEW REFERENCE POINT

$COUNTER = 90/2.5 ; COUNTER EQUALS THE NUMBER OF HOLES FOR EVERY 2.5 DEGREES IN THIRD SECTION

FOR $LSIX = 0 TO ($COUNTER - 1)

G91 LINEAR Z(ABOVES * -1 + $DEPTH[0] + OVERSHOOT * -1) F1 ; VERTICAL TRACK DOWNWARD
G91 LINEAR Z(ABOVES + $DEPTH[0] * -1 + OVERSHOOT) F1 ; VERTICAL TRACK UPWARD (DOUBLE PASSED)
G90 LINEAR X($RADIUS2 * COS(PI/2 - DEG*($LSIX + 1))) Y(($RADIUS2 * $YFLAG) F1 ; SHIFT TO THE NEXT VERTICAL ACCESS PORT

NEXT $LSIX

G92 X0 Y0

$COUNTER = CHANNEL2/$HSEP[$LSEVEN] ; COUNTER EQUALS THE LENGTH OF THE FOURTH SECTION LONG CHANNEL

FOR $LSIX = 0 TO ($COUNTER - 1)

G91 LINEAR Z(ABOVES * -1 + $DEPTH[0] + OVERSHOOT * -1) F1 ; VERTICAL TRACK DOWNWARD
G91 LINEAR Z(ABOVES + $DEPTH[0] * -1 + OVERSHOOT) F1 ; VERTICAL TRACK UPWARD (DOUBLE PASSED)
G91 LINEAR Y($HSEP[$LSEVEN] * $YFLAG) F1 ; SHIFT TO THE NEXT VERTICAL HOLE

NEXT $LSIX
G92 X0 Y0

$COUNTER = CHANNEL3/$HSEP[$LSEVEN] ; COUNTER EQUALS THE LENGTH OF THE FIFTH CHANNEL

FOR $LSIX = 0 TO ($COUNTER - 1)

G91 LINEAR Z(ABOVES * -1 + $DEPTH[0] + (($DEPTH[1]-$DEPTH[0])/CHANNEL3 * $HSEP[$LSEVEN] * ($LSIX)) + OVERSHOOT * -1) F1 ; VERTICAL TRACK DOWNWARD
G91 LINEAR Z(ABOVES + $DEPTH[0] * -1 - (($DEPTH[1]-$DEPTH[0])/CHANNEL3 * $HSEP[$LSEVEN] * ($LSIX)) + OVERSHOOT) F1 ; VERTICAL TRACK UPWARD (DOUBLE PASSED)
G91 LINEAR Y($HSEP[$LSEVEN] * $YFLAG) F1 ; SHIFT TO THE NEXT VERTICAL ACCESS PORT

NEXT $LSIX

G92 X0 Y0

$COUNTER = CHANNEL4/$HSEP[$LSEVEN] ; COUNTER EQUALS THE LENGTH OF THE FOURTH CHANNEL

FOR $LSIX = 0 TO ($COUNTER)

G91 LINEAR Z(ABOVES * -1 + $DEPTH[1] + OVERSHOOT * -1) F1 ; VERTICAL TRACK DOWNWARD
G91 LINEAR Z(ABOVES + $DEPTH[1] * -1 + OVERSHOOT) F1 ; VERTICAL TRACK UPWARD (DOUBLE PASSED)
G91 LINEAR Y($HSEP[$LSEVEN] * $YFLAG) F1 ; SHIFT TO THE NEXT VERTICAL ACCESS PORT

NEXT $LSIX

G92 X0 Y0 ; SET A NEW REFERENCE POINT

$COUNTER = 90/2.5 ; COUNTER EQUALS THE NUMBER OF HOLES FOR EVERY 2.5 DEGREES IN FIFTH SECTION

FOR $LSIX = 0 TO ($COUNTER - 1)

G91 LINEAR Z(ABOVES * -1 + $DEPTH[1] + OVERSHOOT * -1) F1 ; VERTICAL TRACK DOWNWARD
G91 LINEAR Z(ABOVES + $DEPTH[1] * -1 + OVERSHOOT) F1 ; VERTICAL TRACK UPWARD (DOUBLE PASSED)
G90 LINEAR X((RADIUS2 - RADIUS2 * COS(DEG*(LSIX + 1))) * XFLAG) Y(RADIUS2 * SIN(DEG*(LSIX + 1)) * YFLAG) F1 ; SHIFT TO THE NEXT VERTICAL HOLE

NEXT $LSIX

G92 X0 Y0 ; SET A NEW REFERENCE POINT

$COUNTER = 90/2.5 ; COUNTER EQUALS THE NUMBER OF HOLES FOR EVERY 2.5 DEGREES IN SIXTH SECTION

FOR $LSIX = 0 TO ($COUNTER - 1)

G91 LINEAR Z(ABOVES * -1 + DEPTH[1] + OVERSHOOT * -1) F1 ; VERTICAL TRACK DOWNWARD
G91 LINEAR Z(ABOVES + DEPTH[1] * -1 + OVERSHOOT) F1 ; VERTICAL TRACK UPWARD (DOUBLE PASSED)
G90 LINEAR X((RADIUS1 * COS(PI/2 - DEG*(LSIX + 1))) * XFLAG) Y((RADIUS1 - RADIUS1 * SIN(PI/2 - DEG*(LSIX + 1))) * YFLAG) F1 ; SHIFT TO THE NEXT VERTICAL ACCESS PORT

NEXT $LSIX

$COUNTER = CHANNEL1/HSEP[$LSEVEN] ; COUNTER EQUALS THE LENGTH OF THE FIRST SECTION SHORT CHANNEL

FOR $LSIX = 0 TO ($COUNTER - 1)

G91 LINEAR Z(ABOVES * -1 + DEPTH[1] + OVERSHOOT * -1) F1 ; VERTICAL TRACK DOWNWARD
G91 LINEAR Z(ABOVES + DEPTH[1] * -1 + OVERSHOOT) F1 ; VERTICAL TRACK UPWARD (DOUBLE PASSED)
G91 LINEAR Y(HSEP[$LSEVEN] * YFLAG) F1 ; SHIFT TO THE NEXT VERTICAL ACCESS PORT

NEXT $LSIX

G90 LINEAR Z(ABOVES) F1 ; SHIFT ABOVE THE SURFACE

NEXT $VERSE

NEXT $LTWO

NEXT $LONE
1593  NEXT $LSEVEN
1594  NEXT $LZERO
1595  G90 LINEAR Z2 F100 ; MOVE ABOVE THE FOCAL PLANE
1596  G90 LINEAR UU40 E2 ; LOWER THE POWER
1597  M2 ; PROGRAM TERMINATION
1598
1599  G-code Part 2:

1 // Title: Multilevel Capillary Electrophoresis Device Part 2
2 // Author: Stephen Ho
3 // Date Created: April 28, 2011
4 // Description: Writes one curved channel that stay at one focal plane with two reservoirs
5 // ////////////////////////////////////////////////////////////////////////
6 // Parameters (USER INPUT REQUIRED)
7 // DEFINE NSPEED 1 ; NUMBER OF SCAN SPEED
8 #DEFINE NLDEPTH 1 ; NUMBER OF LAYER DEPTH
9 #DEFINE NLINESEP 1 ; NUMBER OF TRANSVERSE LINE TO LINE SEPARATION
10 #DEFINE NVERSET 3 ; NUMBER OF SETS OF VERTICAL ACCESS PORT
11 #DEFINE CDEPTH 1 ; NUMBER OF CHANNEL DEPTH
12 #DEFINE HOLESEP 1 ; NUMBER OF VERTICAL ACCESS PORT SEPARATION
13 #DEFINE CHANNEL1 2.5 ; LENGTH OF SHORT STRAIGHT ARM
14 #DEFINE CHANNEL2 5 ; LENGTH OF LONG STRAIGHT ARM
15 #DEFINE RADIUS 2.5 ; RADIUS OF CIRCULAR ARC
16 #DEFINE PI 3.14159265358979323846264338327950288419716939937
17 #DEFINE DEG 2.5 * PI / 180
18
19 DVAR $SPEED[NSPEED] $LDEPTH[NLDEPTH] $LINESEP[NLINESEP]
21
22 #DEFINE NLAYER 16 ; NUMBER OF LAYERS
23 #DEFINE NLINE 30 ; NUMBER OF LINES IN EACH LAYER
24 #DEFINE ABOVE 0.5 ; THE DISTANCE ABOVE THE SAMPLE SURFACE
25 #DEFINE OVERSHOOT 0.005 ; THE OVERSHOOT DISTANCE INTO THE CHANNEL
26 #DEFINE CEXTRA 0.050 ; THE EXTRA CHANNEL LENGTH INTO EACH RESERVOIR
27 #DEFINE RWIDTH 0.5 ; THE WIDTH OF THE RESERVOIR
28 #DEFINE RLENGTH 0.5 ; THE LENGTH OF THE RESERVOIR
29 #DEFINE RHEIGHT 0.2 ; THE HEIGHT OF THE RESERVOIR
#DEFINE NSHELL 2 ; NUMBER OF SHELLS AT EACH BOARDER
#DEFINE DRT 0.002 ; TRANSVERSE SPACING IN RESERVOIR
#DEFINE DRL 0.005 ; LAYER SPACING IN RESERVOIR

DVAR $YSFT $XSFT $LZERO $LONE $LTWO $LTHREE $LFOUR $LFIVE $LSIX $LSEVEN $LEIGHT $LNINE $LTEN $LELEVEN $YFLAG $XFLAG $COUNTER $LENGTH $RADIUS1 $RADIUS2 $VERSET

METRIC
SECONDS

; PRESET PARAMETERS

; HORIZONTAL SEPARATION FOR EACH VERTICAL HOLE
$HSEP[0]=0.1

; SCAN SPEED FOR THROUGH CHANNELS
$SPEED[0]=0.5

; DEPTH FOR EACH LAYER IN EACH CHANNEL FROM BOTTOM UP
$LDEPTH[0]=0.002

; SEPARATION FOR EACH LINE IN EACH LAYER
$LINESEP[0]=-0.0015

; THE CHANNEL DEPTH (MACHINE) FROM THE SURFACE (MULTIPLY 1.4 TO GET THE CORRECT DEPTH FROM THE SURFACE)
$DEPTH[0]=-0.100

; NUMBER OF LINES TO BE MOVED TO GET TO THE SET OF VERTICAL ACCESS PORTS
$HOLES[0] = 5
$HOLES[1] = 10
$HOLES[2] = 10

; NUMBER OF LINES OR LINE SPACINGS TO SUBTRACT OR ADD TO THE RADIUS
$LINES[0] = 5
$LINES[1] = 15
$LINES[2] = 25

;LENGTH OF THE SAMPLE OF INTEREST
$LENGTH = 2*(CHANNEL1 + RADIUS + RADIUS - NLINE * $LINESEP[0] * -1) + CHANNEL2

$XSFT = 0 ; DEFINE OFFSITE LENGTH ON X-AXIS
$YFLAG = -1
$XFLAG = 1

; ***START OF PROGRAM***
; ***MAKING RESERVOIR***
G90 LINEAR UU24.642 E1 ; SET POWER FOR THE RESERVOIR
$YFLAG = -1
$XFLAG = -1

FOR $LTEN = 0 TO ((RHEIGHT / 0.1)-1) ; LOOP FOR NUMBER OF LAYERS IN THE OPTICAL AXIS DIRECTION
G90 LINEAR Z2 F10 ; MOVE ABOVE THE FOCAL PLANE
G90 LINEAR X0 Y0 F10 ; MOVE BACK TO THE ORIGINAL REFERENCE POINT
G90 LINEAR Z(RHEIGHT * -1 + $LTEN * 0.1) F1 ; MOVE TO THE BOTTOM LAYER

FOR $LELEVEN = 0 TO (RLENGTH/DRT) ; LOOP FOR NUMBER OF LINES AT THE BOTTOM LAYER
G91 LINEAR Y(RWIDTH * $YFLAG) F$SPEED[0] ; SCAN ONE LINE IN THE Y-DIRECTION
G91 LINEAR X(DRT * $XFLAG) F1 ; MOVE TO THE NEXT LINE
$YFLAG = $YFLAG * -1 ; CHANGE DIRECTION

NEXT $LELEVEN

G90 LINEAR Z2 F100 ; MOVE ABOVE THE FOCAL PLANE
G90 LINEAR X0 Y0 F10 ; MOVE BACK TO THE ORIGINAL POSITION
$YFLAG = -1
$XFLAG = -1

FOR $LEIGHT = 0 TO (RLENGTH/0.1) ; LOOP FOR NUMBER OF LAYERS IN THE WIDTHWISE DIRECTION
Appendix B. G-code: Multi-Level Capillary Electrophoresis Device

G90 LINEAR Z(RHEIGHT * -1 + $LTEN * 0.1) F1 ; MOVE TO THE BOTTOM LAYER
G90 LINEAR Y(-0.1 * $LEIGHT) ; MOVE TO THE CORRECT LAYER POSITION IN THE Y-DIRECTION

FOR $LNINE = 0 TO (0.1 / DRL) ; LOOP FOR NUMBER OF LAYERS IN EACH CUBE

G91 LINEAR X(RLENGTH * $XFLAG) F$SPEED[0] ; SCAN A X-DIRECTION LINE
$XFLAG = $XFLAG * -1 ; CHANGE DIRECTION
G91 LINEAR Y(DRT * $YFLAG) F1 ; MOVE TO THE NEXT SHELL
$YFLAG = $YFLAG * -1 ; CHANGE DIRECTION
G91 LINEAR X(RLENGTH * $XFLAG) F$SPEED[0] ; SCAN ONE LINE BACKWARD IN X-DIRECTION
$XFLAG = $XFLAG * -1 ; CHANGE DIRECTION
G91 LINEAR Z(DRL) F1 ; MOVE UP ONE LAYER

NEXT $LNINE

NEXT $LEIGHT

G90 LINEAR Z2 F100 ; MOVE ABOVE THE FOCAL PLANE
G90 LINEAR X0 Y0 F10 ; MOVE BACK TO THE ORIGINAL POSITION
$YFLAG = -1
$XFLAG = -1

FOR $LEIGHT = 0 TO (RWIDTH/0.1) ; LOOP FOR NUMBER OF LAYERS IN THE LENGTHWISE DIRECTION

G90 LINEAR Z(RHEIGHT * -1 + $LTEN * 0.1) F1 ; MOVE TO THE BOTTOM OF THE LAYER
G90 LINEAR X(-0.1 * $LEIGHT) ; MOVE TO THE CORRECT LAYER POSITION IN THE X-DIRECTION

FOR $LNINE = 0 TO (0.1 / DRL) ; LOOP FOR NUMBER OF LAYERS IN EACH CUBE

G91 LINEAR Y(RWIDTH * $YFLAG) F$SPEED[0] ; SCAN A X-DIRECTION LINE
G91 LINEAR X(DRT * $XFLAG) F1 ; MOVE TO THE NEXT SHELL
$YFLAG = $YFLAG * -1 ; CHANGE DIRECTION
$XFLAG = $YFLAG * -1 ; CHANGE DIRECTION
$YFLAG = $YFLAG * -1 ; CHANGE DIRECTION
G91 LINEAR Y(RWIDTH * $YFLAG) F$SPEED[0] ; SCAN ONE LINE BACKWARD IN X-DIRECTION
$YFLAG = $YFLAG * -1 ; CHANGE DIRECTION
G91 LINEAR Z(DRL) F1 ; MOVE UP ONE LAYER
G90 LINEAR Z2 F10 ; MOVE ABOVE THE FOCAL PLANE
G90 LINEAR X0 Y0 F10 ; MOVE BACK TO THE ORIGINAL REFERENCE POINT

; ***MAKING SECOND RESERVOIR***
$YFLAG = -1
$XFLAG = -1

G90 LINEAR Y($LENGTH * -1 - RWIDTH) ; MOVE TO THE RESERVOIR ON THE OTHER END
G92 X0 Y0 ; SET A NEW REFERENCE POINT

FOR $LTEN = 0 TO ((RHEIGHT / 0.1)-1) ; LOOP FOR NUMBER OF LAYERS IN THE OPTICAL AXIS DIRECTION
G90 LINEAR Z2 F10 ; MOVE ABOVE THE FOCAL PLANE
G90 LINEAR X0 Y0 F10 ; MOVE BACK TO THE ORIGINAL REFERENCE POINT
G90 LINEAR Z(RHEIGHT * -1 + $LTEN * 0.1) F1 ; MOVE TO THE BOTTOM LAYER

FOR $LELEVEN = 0 TO (RLENGTH/DRT) ; LOOP FOR NUMBER OF LINES AT THE BOTTOM LAYER
G91 LINEAR Y(RWIDTH * $YFLAG) F$SPEED[0] ; SCAN ONE LINE IN THE Y-DIRECTION
G91 LINEAR X(DRT * $XFLAG) F1 ; MOVE TO THE NEXT LINE
$YFLAG = $YFLAG * -1 ; CHANGE DIRECTION

NEXT $LELEVEN

G90 LINEAR Z2 F100 ; MOVE ABOVE THE FOCAL PLANE
G90 LINEAR X0 Y0 F10 ; MOVE BACK TO THE ORIGINAL POSITION
$YFLAG = -1
$XFLAG = -1

FOR $LEIGHT = 0 TO (RLENGTH/0.1) ; LOOP FOR NUMBER OF LAYERS IN THE WIDTHWISE DIRECTION
G90 LINEAR Z(RHEIGHT * -1 + $LTEN * 0.1) F1 ; MOVE TO THE BOTTOM OF THE LAYER
G90 LINEAR Y(-0.1 * $LEIGHT) ; MOVE TO THE CORRECT LAYER POSITION IN THE Y-DIRECTION

FOR $LNINE = 0 TO (0.1 / DRL) ; LOOP FOR NUMBER OF LAYERS IN EACH CUBE

G91 LINEAR X(RLENGTH * $XFLAG) F$SPEED[0] ; SCAN A X-DIRECTION LINE
$XFLAG = $XFLAG * -1 ; CHANGE DIRECTION
G91 LINEAR Y(DRT * $YFLAG) F1 ; MOVE TO THE NEXT SHELL
$YFLAG = $YFLAG * -1 ; CHANGE DIRECTION
G91 LINEAR X(RLENGTH * $XFLAG) F$SPEED[0] ; SCAN ONE LINE BACKWARD IN X-DIRECTION
$XFLAG = $XFLAG *-1 ; CHANGE DIRECTION
G91 LINEAR Z(DRL) F1 ; MOVE UP ONE LAYER

NEXT $LNINE

NEXT $LEIGHT

G90 LINEAR Z2 F100 ; MOVE ABOVE THE FOCAL PLANE
G90 LINEAR X0 Y0 F10 ; MOVE BACK TO THE ORIGINAL POSITION
$YFLAG = -1
$XFLAG = -1

FOR $LEIGHT = 0 TO (RWIDTH/0.1) ; LOOP FOR NUMBER OF LAYERS IN THE LENGTHWISE DIRECTION

G90 LINEAR Z(RHEIGHT * -1 + $LTEN * 0.1) F1 ; MOVE TO THE BOTTOM OF THE LAYER
G90 LINEAR X(-0.1 * $LEIGHT) ; MOVE TO THE CORRECT LAYER POSITION IN THE X-DIRECTION

FOR $LNINE = 0 TO (0.1 / DRL) ; LOOP FOR NUMBER OF LAYERS IN EACH CUBE

G91 LINEAR Y(RWIDTH * $YFLAG) F$SPEED[0] ; SCAN A X-DIRECTION LINE
$YFLAG = $YFLAG * -1 ; CHANGE DIRECTION
G91 LINEAR X(DRT * $XFLAG) F1 ; MOVE TO THE NEXT SHELL
$XFLAG = $XFLAG * -1 ; CHANGE DIRECTION
G91 LINEAR Y(RWIDTH * $YFLAG) F$SPEED[0] ; SCAN ONE LINE BACKWARD IN X-DIRECTION
$YFLAG = $YFLAG *-1 ; CHANGE DIRECTION
G91 LINEAR Z(DRL) F1 ; MOVE UP ONE LAYER
;***MAKING MICROCHANNEL WITH VERTICAL ACCESS PORTS*** $XFLAG = -1
$YFLAG = -1

G90 LINEAR UU28.842 E1 ;SET POWER FOR OUTSIDE BOUNDARY OF THE CHANNELS

$XSFT = -0.100

G90 LINEAR Z2 F10 ; MOVE ABOVE THE FOCAL PLANE
G90 LINEAR X0 Y($LENGTH + RWIDTH)F10 ; MOVE BACK TO THE ORIGINAL REFERENCE POINT
G90 LINEAR X(RLENGTH/2 * -1) F10 ; MOVE TO THE MIDDLE OF THE RESERVOIR
G91 LINEAR Y(RWIDTH * -1) F10 ; MOVE TO THE END OF THE RESERVOIR AND INTO THE RESERVOIR WITH CEXTRA
G92 X0 Y0 ; CHANGE THE REFERENCE POINT FOR X AND Y AXIS

FOR $LFIVE = 0 TO (CDEPTH -1)

G90 LINEAR Z($DEPTH[$LFIVE] * NLAYER *-1) F1 ; MOVE TO THE DESIRED CHANNEL DEPTH FOR BOTTOM UP SCANNING

FOR $LZERO = 0 TO (NSPEED -1) ; LOOP LAYER ZERO FOR DIFFERENT SCAN SPEED

FOR $LSEVEN = 0 TO (HOLESEP -1) ; LOOP LAYER SEVEN FOR DIFFERENT HOLE SEPARATION DISTANCE

FOR $LONE = 0 TO (NLDEPTH -1) ; LOOP LAYER ONE FOR DIFFERENT LAYER SEPARATION

FOR $LTWO = 0 TO (NLINESEP -1) ; LOOP LAYER TWO FOR DIFFERENT LINE SEPARATION IN EACH LAYER

G91 LINEAR Z($LDEPTH[$LONE] * NLAYER *-1) F1 ; MOVE TO THE DESIRED CHANNEL DEPTH FOR BOTTOM UP SCANNING
FOR $LTHREE = 0 TO (NLAYER) ; LOOP LAYER THREE FOR NUMBER OF
LAYERS IN EACH CHANNEL

$RADIUS1 = RADIUS ; DEFINE THE OUTER RADIUS
$RADIUS2 = RADIUS - (NLINE * $LINESEP[0] * -1) ; DEFINE THE INNER RADIUS
FOR $LFOUR = 0 TO (NLINE -1) ; LOOP LAYER FOUR FOR NUMBER OF LINES
IN EACH LAYER

IF(($LTHREE == 0) OR ($LTHREE == NLAYER) OR ($LFOUR == 0))
G90 LINEAR UU28.842 E1 ; SET POWER FOR THE OUTSIDE OF THE CHANNEL
ELSE
G90 LINEAR UU28.842 E1 ; SET POWER FOR THE INSIDE OF THE CHANNEL
ENDIF

VELOCITY ON
M146

G91 LINEAR Y(CHANNEL1 * $YFLAG) F($SPEED[$LZERO]) ; FIRST SECTION
SHORT CHANNEL ARM

IF ($XFLAG == -1) ; MOVING TOWARD RIGHT
IF ($YFLAG == -1) ; MOVING DOWNWARD
CCW P180 Q270 R($RADIUS1) F($SPEED[$LZERO]) ; SECOND SECTION CIRCULAR
ARC
CW P90 Q0 R($RADIUS2) F($SPEED[$LZERO]) ; THIRD SECTION CIRCULAR
ARC
ELSE
CW P180 Q90 R($RADIUS1) F($SPEED[$LZERO])
CCW P270 Q0 R($RADIUS2) F($SPEED[$LZERO])
ENDIF

ELSE ; MOVING TOWARD LEFT
IF ($YFLAG == 1) ; MOVING UPWARD
CW P180 Q90 R($RADIUS2) F($SPEED[$LZERO])
CCW P270 Q0 R($RADIUS1) F($SPEED[$LZERO])
ELSE
CCW P180 Q270 R($RADIUS2) F($SPEED[$LZERO])
CW P90 Q0 R($RADIUS1) F($SPEED[$LZERO])
ENDIF

ENDIF
G91 LINEAR Y(CHANNEL2 * $YFLAG) F($SPEED[$LZERO]) ; FOURTH SECTION
LONG CHANNEL ARM
IF ($XFLAG == -1)
  IF ($YFLAG == -1)
    CW P0 Q270 R($RADIUS2) F($SPEED[$LZERO]) ; FIFTH SECTION CIRCULAR ARC
    CCW P90 Q180 R($RADIUS1) F($SPEED[$LZERO]) ; SIXTH SECTION CIRCULAR ARC
  ELSE
    CCW P0 Q90 R($RADIUS2) F($SPEED[$LZERO])
    CW P270 Q180 R($RADIUS1) F($SPEED[$LZERO])
  ENDIF
ELSE
  IF ($YFLAG == 1)
    CCW P0 Q90 R($RADIUS1) F($SPEED[$LZERO])
    CW P270 Q180 R($RADIUS2) F($SPEED[$LZERO])
  ELSE
    CW P0 Q270 R($RADIUS1) F($SPEED[$LZERO])
    CCW P90 Q180 R($RADIUS2) F($SPEED[$LZERO])
  ENDIF
ENDIF
$YFLAG = $YFLAG * (-1) ; CHANGE DIRECTION
$RADIUS1 = $RADIUS1 - ($LINESEP[$LTWO] * -1) ; DECREASE THE OUTER RADIUS
$RADIUS2 = $RADIUS2 + ($LINESEP[$LTWO] * -1) ; INCREASE THE INNER RADIUS
G91 LINEAR Y(CHANNEL1 * $YFLAG) F($SPEED[$LZERO]) ; SEVENTH SECTION SHORT CHANNEL ARM
$YFLAG = $YFLAG * (-1) ; CHANGE DIRECTION
G90 LINEAR UU28.842 E1 ; SET POWER FOR THE OUTSIDE BOUNDARY OF THE CHANNEL
VELOCITY ON
M146

G91 LINEAR Y(CHANNEL1 * $YFLAG) F($SPEED[$LZERO]) ; FIRST SECTION SHORT CHANNEL ARM

IF ($XFLAG == -1) ; MOVING TOWARD RIGHT
  IF ($YFLAG == -1) ; MOVING DOWNWARD
    CCW P180 Q270 R($RADIUS1) F($SPEED[$LZERO]) ; SECOND SECTION CIRCULAR ARC
    CW P90 Q0 R($RADIUS2) F($SPEED[$LZERO]) ; THIRD SECTION CIRCULAR ARC
  ELSE
    CW P180 Q90 R($RADIUS1) F($SPEED[$LZERO])
    CCW P270 Q0 R($RADIUS2) F($SPEED[$LZERO])
  ENDIF
ELSE ; MOVING TOWARD LEFT
  IF ($YFLAG == 1) ; MOVING UPWARD
    CW P180 Q90 R($RADIUS2) F($SPEED[$LZERO])
    CCW P270 Q0 R($RADIUS1) F($SPEED[$LZERO])
  ELSE
    CCW P180 Q270 R($RADIUS2) F($SPEED[$LZERO])
    CW P90 Q180 R($RADIUS1) F($SPEED[$LZERO])
  ENDIF
ENDIF

G91 LINEAR Y(CHANNEL2 * $YFLAG) F($SPEED[$LZERO]) ; FOURTH SECTION LONG CHANNEL ARM

IF ($XFLAG == -1)
  IF ($YFLAG == -1)
    CW P0 Q270 R($RADIUS2) F($SPEED[$LZERO]) ; FIFTH SECTION CIRCULAR ARC
    CCW P90 Q180 R($RADIUS1) F($SPEED[$LZERO]) ; SIXTH SECTION CIRCULAR ARC
  ELSE
    CCW P0 Q90 R($RADIUS2) F($SPEED[$LZERO])
    CW P270 Q180 R($RADIUS1) F($SPEED[$LZERO])
  ENDIF
ENDIF
ELSE

IF ($YFLAG == 1)
CCW P0 Q90 R($RADIUS1) F($SPEED[$LZERO])
CW P270 Q180 R($RADIUS2) F($SPEED[$LZERO])
ELSE
CW P0 Q270 R($RADIUS1) F($SPEED[$LZERO])
CCW P90 Q180 R($RADIUS2) F($SPEED[$LZERO])
ENDIF

ENDIF

G91 LINEAR Y(CHANNEL1 * $YFLAG) F($SPEED[$LZERO]) ; SEVENTH SECTION SHORT CHANNEL ARM

$XFLAG = $XFLAG * (-1)
$YFLAG = $YFLAG * (-1)

VELOCITY OFF

IF($LTHREE < NLAYER)
G91 LINEAR Z($LDEPTH[$LONE]) F1 ; SHIFT TO THE NEXT LAYER WITH BOTTOM UP
ENDIF

NEXT $LTHREE

;***SETS OF VERTICAL ACCESS PORTS***
G90 X0 F1

FOR $VERSET = 0 TO (NVERSET - 1) ; LOOP THE NUMBER OF SETS OF VERTICAL HOLES

$XFLAG = -1
$YFLAG = -1
$RADIUS1 = RADIUS - ($LINES[$VERSET] * $LINESEP[0] * -1) ; DEFINE THE OUTER RADIUS
$RADIUS2 = RADIUS - (NLINE - $LINES[$VERSET]) * $LINESEP[0] * -1 ; DEFINE THE INNER RADIUS

G90 LINEAR Z(ABOVES) F1 ; SHIFT ABOVE THE SURFACE
G91 LINEAR X($LINESEP[$LTWO] * $XFLAG * $HOLES[$VERSET]) F1 ; SHIFT BACK TO THE SIDE MIDDLE OF THE CHANNEL IN X-DIRECTION
G91 LINEAR Y($LENGTH) F2 ; SHIFT BACK TO THE FIRST RESERVOIR SIDE
$\text{COUNTER} = \text{CHANNEL1}/\text{HSEP}[\text{LSEVEN}]$ ; \text{COUNTER EQUALS THE LENGTH OF THE FIRST SECTION SHORT CHANNEL}

G90 LINEAR UU24.642 E1 ; \text{SET POWER FOR THE VERTICAL ACCESS PORT}

\text{FOR $LSIX = 0 \text{ TO ($COUNTER - 1)}$

G91 LINEAR Z(ABOVES * -1 + $\text{DEPTH}[0] + \text{OVERSHOOT} * -1) F1 ; \text{VERTICAL TRACK DOWNWARD}

G91 LINEAR Z(ABOVES + $\text{DEPTH}[0] * -1 + \text{OVERSHOOT}) F1 ; \text{VERTICAL TRACK UPWARD (DOUBLE PASSED)}

G91 LINEAR Y($\text{HSEP}[\text{LSEVEN}] * \text{YFLAG}) F1 ; \text{SHIFT TO THE NEXT VERTICAL ACCESS PORT}

\text{NEXT $LSIX$

G92 X0 Y0 ; \text{SET A NEW REFERENCE POINT}

$\text{COUNTER} = 90/2.5$ ; \text{COUNTER EQUALS THE NUMBER OF HOLES FOR EVERY 2.5 DEGREES IN SECOND SECTION}

\text{FOR $LSIX = 0 \text{ TO ($COUNTER - 1)}$}

G91 LINEAR Z(ABOVES * -1 + $\text{DEPTH}[0] + \text{OVERSHOOT} * -1) F1 ; \text{VERTICAL TRACK DOWNWARD}

G91 LINEAR Z(ABOVES + $\text{DEPTH}[0] * -1 + \text{OVERSHOOT}) F1 ; \text{VERTICAL TRACK UPWARD (DOUBLE PASSED)}

G90 LINEAR X($\text{RADIUS1} - \text{RADIUS1} \times \cos(\text{DEG}*(\text{LSIX + 1}))) \text{Y}($\text{RADIUS1} \times \sin(\text{DEG}*(\text{LSIX + 1}))) * \text{YFLAG}) F1 ; \text{SHIFT TO THE NEXT VERTICAL ACCESS PORT}

\text{NEXT $LSIX$

G92 X0 Y0 ; \text{SET A NEW REFERENCE POINT}

$\text{COUNTER} = 90/2.5$ ; \text{COUNTER EQUALS THE NUMBER OF HOLES FOR EVERY 2.5 DEGREES IN THIRD SECTION}

\text{FOR $LSIX = 0 \text{ TO ($COUNTER - 1)}$}

G91 LINEAR Z(ABOVES * -1 + $\text{DEPTH}[0] + \text{OVERSHOOT} * -1) F1 ; \text{VERTICAL TRACK DOWNWARD}

G91 LINEAR Z(ABOVES + $\text{DEPTH}[0] * -1 + \text{OVERSHOOT}) F1 ; \text{VERTICAL TRACK UPWARD (DOUBLE PASSED)}
G90 LINEAR X($RADIUS2 * COS(PI/2 - DEG*(LSIX + 1))) Y(($RADIUS2 - $RADIUS2 * SIN(PI/2 - DEG*(LSIX + 1))) * YFLAG) F1 ; SHIFT TO THE NEXT VERTICAL ACCESS PORT

NEXT LSIX

$COUNTER = CHANNEL2/$HSEP[LSIX] ; COUNTER EQUALS THE LENGTH OF THE FOURTH SECTION LONG CHANNEL

FOR LSIX = 0 TO ($COUNTER - 1)

G91 LINEAR Z(ABOVES * -1 + $DEPTH[0] + OVERSHOOT * -1) F1 ; VERTICAL TRACK DOWNWARD
G91 LINEAR Z(ABOVES + $DEPTH[0] * -1 + OVERSHOOT) F1 ; VERTICAL TRACK UPWARD (DOUBLE PASSED)
G91 LINEAR Y($HSEP[LSIX] * YFLAG) F1 ; SHIFT TO THE NEXT VERTICAL ACCESS PORT

NEXT LSIX

G92 X0 Y0 ; SET A NEW REFERENCE POINT

$COUNTER = 90/2.5 ; COUNTER EQUALS THE NUMBER OF HOLES FOR EVERY 2.5 DEGREES IN FIFTH SECTION

FOR LSIX = 0 TO ($COUNTER - 1)

G91 LINEAR Z(ABOVES * -1 + $DEPTH[0] + OVERSHOOT * -1) F1 ; VERTICAL TRACK DOWNWARD
G91 LINEAR Z(ABOVES + $DEPTH[0] * -1 + OVERSHOOT) F1 ; VERTICAL TRACK UPWARD (DOUBLE PASSED)
G90 LINEAR X(($RADIUS2 - $RADIUS2 * COS(DEG*(LSIX + 1))) * XFLAG) Y($RADIUS2 * SIN(DEG*(LSIX + 1))) * YFLAG) F1 ; SHIFT TO THE NEXT VERTICAL ACCESS PORT

NEXT LSIX

G92 X0 Y0 ; SET A NEW REFERENCE POINT

$COUNTER = 90/2.5 ; COUNTER EQUALS THE NUMBER OF HOLES FOR EVERY 2.5 DEGREES IN SIXTH SECTION

FOR LSIX = 0 TO ($COUNTER - 1)
G91 LINEAR Z(ABOVES * -1 + $DEPTH[0] + OVERSHOOT * -1) F1 ; VERTICAL TRACK DOWNWARD
G91 LINEAR Z(ABOVES + $DEPTH[0] * -1 + OVERSHOOT) F1 ; VERTICAL TRACK UPWARD (DOUBLE PASSED)
G90 LINEAR X(($RADIUS1 * COS(PI/2 - DEG*($LSIX + 1))) * $XFLAG) Y(($RADIUS1 - $RADIUS1 * SIN(PI/2 - DEG*($LSIX + 1))) * $YFLAG) F1 ; SHIFT TO THE NEXT VERTICAL ACCESS PORT

NEXT $LSIX

$COUNTER = CHANNEL1/$HSEP[$LSEVEN] ; COUNTER EQUALS THE LENGTH OF THE FIRST SECTION SHORT CHANNEL
FOR $LSIX = 0 TO ($COUNTER - 1)
G91 LINEAR Z(ABOVES * -1 + $DEPTH[0] + OVERSHOOT * -1) F1 ; VERTICAL TRACK DOWNWARD
G91 LINEAR Z(ABOVES + $DEPTH[0] * -1 + OVERSHOOT) F1 ; VERTICAL TRACK UPWARD (DOUBLE PASSED)
G91 LINEAR Y($HSEP[$LSEVEN] * $YFLAG) F1 ; SHIFT TO THE NEXT VERTICAL ACCESS PORT

NEXT $LSIX

G90 LINEAR Z(ABOVES) F1 ; SHIFT ABOVE THE SURFACE

NEXT $VERSET
NEXT $LTWO
NEXT $LONE
NEXT $LSEVEN
NEXT $LZERO
NEXT $LFIVE

G90 LINEAR Z2 F100 ; MOVE ABOVE THE FOCAL PLANE
G90 LINEAR UU42 E2 ; LOWER THE POWER

M2 ; PROGRAM TERMINATION
Appendix C

G-code:
IWP/Chromatographic/Waveguide Probing Device

The G-code used for controlling the xyz-stages in laser patterning the uniform inverted-woodpile structure or microphotonic device or waveguide probing device is presented below. The respective structures can be fabricated by changing the parameters in the G-code.

// Title: Waveguide Probing Device or Microphotonic Chromatographic Device: Channel + PC + WG + Reservoir - v3.0
// Author: Moez Haque, Stephen Ho
// Date Created: August 24, 2010
// Last Modified: November 15, 2010
// // Update: September 09, 2010
// - "$Channel1VerticalPortLengthY" and "$Channel2VerticalPortLengthY" equations corrected.
// Update: September 08, 2010
// - One of the "PSOCONTROL X OFF" lines was corrected to "PSOCONTROL X ON" within the PC section.
// - Value of "$Channel1VerticalPortLengthY" and "$Channel2VerticalPortLengthY" corrected to include // overlapping length of channel with the reservoirs.
// - AOM "Arming" is now done above the glass / air interface so that isolated focal volume modifications // no longer occur during arming.
// Update: November 15, 2010
// - Created power array to fabricate uniform PC
// // Description:
// - A single program that write a biochip with Reservoirs, Channels, WGs and PCs.
// - Channel written along y-axis with adjustable inside volume and outside boundary.
// - PC (Woodpile) written with AOM. Gamma-Z along z-axis.
// - WGs written along x-axis in 3 sets: one through PC, one through channel, and one only // through sample.
Appendix C. G-code: IWP/Chromatographic/Waveguide Probing Device

// - Reservoir written with a "low power" boundary and a mesh within the reservoir //
volume with high power.

/////////////////////////////////////////////////////////////////////////
// Parameters (USER INPUT REQUIRED)

DVAR $WG-LengthX $WG-Power $WG-ScanSpeed $WG-TotalLengthY $WG-Separation $WG-TotalLengthY $WG-Separation from_Reservoir $WG-Multiplier
DVAR $WG-AOM-Distance from_Channel_side $WG-AOM-Distance from_PC_side
DVAR $FLAG_WriteWGs_ALMOST through_Channel
DVAR $ReservoirLengthX $ReservoirLengthY $ReservoirLengthZ
DVAR $PC-LengthX $PC-LengthY $PC-LengthZ $PC-WriteSpeed $PC-PeriodicityA $PC-PeriodicityC $PC-VerticalPortSpacingX $PC-VerticalPortSpacingY $PC-NumVerticalPortsX $PC-VerticalPortsLengthZ $PC-Power
DVAR $PC-VerticalPortLengthX $PC-VerticalPortLengthY $PC-VerticalPortPower $PC-VerticalPortScanSpeed $PC-NumVerticalPortsY
DVAR $ChannelLengthX $ChannelLengthY1 $ChannelLengthY2 $ChannelLengthZ $ChannelSpacingX $ChannelSpacingZ $Channel-PowerBoundary $Channel-PowerInner $Channel-Space
DVAR $Channel-Number-Boundary-Lines $Channel-Vertical-Port-LengthX
$Channel1-Vertical-Port-LengthY $Channel1-NumVertical-PortsY
DVAR $Total-Chip-LengthY $Reservoir-PowerInner $Reservoir-Power-Boundary
$Reservoir-Scan-Speed $Reservoir-SpaceX $Reservoir-SpaceY $Reservoir-SpaceZ
DVAR $Non-Write-Speed $UU-Change-Speed $Global-Z-Offset
DVAR $FLAG_Write_WGs_before_Reservoir $FLAG_Write_WGs_through_Channel
$FLAG_Write_WGs_through_PC $FLAG_Write_WGs_x_minus_dir
$FLAG_Write_WGs_x_plus_dir $FLAG_Write_PC $FLAG_Write_Channel_1
$FLAG_Write_Channel_2 $FLAG_Write_Reservoir_1 $FLAG_Write_Reservoir_2
DVAR $FLAG_Write_WGs_ALMOST through_PC $Reservoir-Num-Boundary-Layers
DVAR $WG_For-loop_counter $OrigX $OrigY $OrigZ $Wait-Time AOM $Acceleration-Buffer
$NumPCLinesXAxisLines $NumPCLinesYAxisLines $NumPCLinesZAxisLines $ForLoopCounterXAxisLines $ForLoopCounterYAxisLines $ForLoopCounterZAxisLines
DVAR $VerticalPortXCounter $VerticalPortYCounter $NumChannelLinesXAxis $NumChannelLinesZAxis
DVAR $Channel-Vertical-Port-Power $Channel-Vertical-Port-Scan-Speed $Channel-Num-Vertical-PortsX $Channel-Vertical-Port-SpaceX $Channel-Vertical-Port-SpaceY $Channel-Vertical-Port-LengthZ
DVAR $Channel2-Vertical-Port-LengthY $Channel2-Num-Vertical-PortsY $Channel-Extension-Fraction-Into-Reservoir $Reservoir-Num-LinesXFace $Reservoir-Num-LinesYFace $Reservoir-Num-LinesZFace
DVAR $Reservoir-Num-SectionsX $Reservoir-Num-SectionsY $Reservoir-Num-SectionsZ $Reservoir-Section-LengthX $Reservoir-Section-LengthY $Reservoir-Section-LengthZ
DVAR $Clock-Total-Start $PATHNAME $Low-Power
DVAR $Layer-Power-Count
DVAR $PowerArray[12] // require user input to change the number of different powers for PC

// WG Parameters
$WG_LengthX = 26.2 // Length of WGs across glass sample (should be longer than glass).
$WG_Power = 29.669 // Angle for power for writing WGs.
$WG_ScanSpeed = 0.5 // Scan speed for writing WGs.
$WG_Separation_DistanceY = 0.1 // Distance along y-axis separating adjacent WGs written along x-axis.
$WG_Multiplier = 4 // Number of identical WGs to be written.
$WG_Separation_from_Reservoir = 3.000 // Distance along y-axis separating the WG set (not through anything) and the reservoir.
$WGAOM_Distance_from_Channel_side = 0.005 // Distance away from the channel sidewall for writing WGs (uses AOM).
$WGAOM_Distance_from_PC_side = 0.005 // Distance away from the channel sidewall for writing WGs (uses AOM).

// PC Parameters (Set the Powers at Different Layer)
$PowerArray[0] = 25.712
$PowerArray[1] = 25.907
$PowerArray[6] = 27.115
$PowerArray[7] = 27.348
$PowerArray[8] = 27.576
$PowerArray[9] = 27.843
$PowerArray[10] = 28.085

$PC_PериодностьA = 0.005 // ”a” periodicity of PC (controls periodicity of woodpile PC along x and y axes).
$PC_PериодностьC = 0.014/1.5 // ”c” periodicity of PC (controls periodicity of woodpile PC along z axis). This value is the MOTION OF THE STAGE ... you must account for the refractive index.
$PC_WriteSpeed = 0.5 // Write Speed for PC fabrication.
$PC_LengthX = 0.045 // Length of PC along x-axis. The channels written ”through” the PC will be centered (along x-axis) through the PC.
$PC_LengthY = 10.000 // Length of PC along y-axis. The WG set written ”through” the PC will be centered through the PC.
$PC_LengthZ = 0.0421/1.5 // Length of PC along z-axis. The channels written ”through” the PC will be centered (along z-axis) through the PC.
// Channel Parameters
$ChannelNumberBoundaryLines = 2 // Number of lines within the channel boundary to use the lower "ChannelPowerBoundary".
$ChannelPowerBoundary = 23.556 // Power for writing channel outside boundary.
$ChannelPowerInner = 23.556 // Power for writing channel interior.
$ChannelScanSpeed = 0.5 // Scan Speed for writing Channels.
$ChannelSpacingX = 0.0015 // Distance between two adjacent scans during channel writing along x-axis.
$ChannelSpacingZ = 0.002 // Distance between two adjacent scans during channel writing along z-axis.
$ChannelLengthX = 0.045 // Length of channel along x-axis.
$ChannelLengthY1 = 16 // Length of channel along y-axis on positive x-side of PC. The WG set written through the channel will be centered through this channel section.
$ChannelLengthY2 = 8 // Length of channel along y-axis on negative x-side of PC. WGs will not be written through this channel. If the PC is not written, this channel will NOT be written.
$ChannelLengthZ = 0.04 // Length of channel along z-axis.
$ChannelExtensionFractionIntoReservoir = 10 // Fraction of the Reservoir length to overlap the channels with. 1 = Full reservoir length, 2 = half of reservoir length, etc.

// Reservoir Parameters
$ReservoirPowerInner = 20.716 // Power for writing Reservoir Interior.
$ReservoirPowerBoundary = 20.716 // Power for writing Reservoirs Boundary.
$ReservoirScanSpeed = 0.5 // Scan speed for writing Reservoirs.
$ReservoirSpacingX = 0.0015 // Spacing between adjacent scan lines in x-axis for writing Reservoirs.
$ReservoirSpacingY = 0.0015 // Spacing between adjacent scan lines in x-axis for writing Reservoirs.
$ReservoirSpacingZ = 0.004 // Spacing between adjacent scan lines in x-axis for writing Reservoirs.
$ReservoirNumBoundaryLayers = 2 // Number of Boundary layers for writing the sides of the reservoirs.
$ReservoirNumSectionsX = 5 // Number of Sections within the reservoir along the x axis.
$ReservoirNumSectionsY = 5 // Number of Sections within the reservoir along the y axis.
$ReservoirNumSectionsZ = 1 // Number of Sections within the reservoir along the z axis.
$ReservoirLengthX = 0.500 // Length of Reservoir along x-axis.
$ReservoirLengthY = 0.500 // Length of Reservoir along y-axis.
$ReservoirLengthZ = 0.200 // Length of Reservoir along z-axis. Bottom of Reservoirs is aligned with bottom of Channels.

// Vertical Access Ports PC Parameters
$PCVerticalPortPower = 20.716 // Power for writing vertical throughports above the
PC.
$PCVerticalPortScanSpeed = 1 // Scan speed for writing vertical throughports above PC.
$PCNumVerticalPortsX = 3 // Number of vertical throughports to have adjacent to each
other along the x-axis above the PC.
$PCVerticalPortSpacingX = 0.015 // Spacing between two adjacent sets of vertical
throughports above the PC along the x-axis.
$PCVerticalPortSpacingY = 0.1 // Spacing between two adjacent sets of vertical through-
ports above the PC along the y-axis.
$PCVerticalPortsLengthZ = 0.2 // Length of vertical throughports written above the PC.

// Vertical Throughports Channel Parameters
$ChannelVerticalPortPower = 20.716 // Power for writing vertical throughports above
Channel 1.
$ChannelVerticalPortScanSpeed = 1 // Scan speed for writing vertical throughports
above Channel 1.
$ChannelNumVerticalPortsX = 3 // Number of vertical throughports to have adjacent
to each other along the x-axis above Channel 1.
$ChannelVerticalPortSpacingX = 0.015 // Spacing between two adjacent sets of vertical
throughports above Channel 1 along the x-axis.
$ChannelVerticalPortSpacingY = 0.1 // Spacing between two adjacent sets of vertical
throughports above Channel 1 along the y-axis.
$ChannelVerticalPortsLengthZ = 0.2 // Length of vertical throughports written above
Channel 1.

// Other Parameters
$NonWriteSpeed = 5 // Scan speed for not writing anything.
$UU_Change_Speed = 5 // Speed for changing UU angles.
$GlobalZOffset = -0.02 // Shift the z position away from seeing focus on the CCD cam-
era for all components of the chip writing. Positive values shift UP!
$WaitTimeAOM = 0.1 // Wait time for arming PSO.
$AccelerationBuffer = 0.15 // Distance for ramping up / down speeds to get constant
velocity.
$LowPower = 40 // Low power for non-writing.

// FLAGS
$FLAG_Write_WGs_before_Reservoir = 1 // 0 = Don’t write WGs before Reservoir. 1
= Write WGs before Reservoir.
$FLAG_Write_WGs_through_Channel = 1 // 0 = Don’t write WGs through channel. 1
= Write WGs through channel. Must write Channel in order to write this set of WGs.
$FLAG_WriteWGs_ALMOST_through_Channel = 1 // 0 = Don’t use AOM to write
WGs through channel; 1 = Use AOM to stop WGs just before and after channel edge.
$FLAG_Write_WGs_through_PC = 1 // 0 = Don’t write WGs through PC. 1 = Write
WGs through PC. Must write PC in order to write this set of WGs.
$FLAG_WriteWGs_ALMOST_through_PC = 1 // 0 = Don’t use AOM to write WGs
through PC; 1 = Use AOM to stop WGs just before and after PC edge.

$FLAG_Write_WGs_x_minus_dir = 1 // 0 = Don’t write WGs in x- direction. 1 = Write WGs in x- direction.

$FLAG_Write_WGs_x_plus_dir = 1 // 0 = Don’t write WGs in x+ direction. 1 = Write WGs in x+ direction.

$FLAG_Write_PC = 1 // 0 = Don’t write PC. 1 = Write PC. ”Channel 2” is not written unless the PC is written.

$FLAG_Write_Channel_1 = 1 // 0 = Don’t write Channel 1. 1 = Write Channel 1.

$FLAG_Write_Channel_2 = 1 // 0 = Don’t write Channel 2. 1 = Write Channel 2. ”Channel 2” is not written unless the PC is written.

$WGTotalLengthY = ($FLAG_Write_WGs_x_minus_dir + $FLAG_Write_WGs_x_plus_dir)*$WGMultiplier*$WGSeparation_DistanceY

$TotalChipLengthY = $FLAG_Write_WGs_before_Reservoir*($WGTotalLengthY + $WG_Separation_from_Reservoir) + $ReservoirLengthY + $ChannelLengthY1 + $FLAG_Write_PC*($PCLengthY + $ChannelLengthY2) + $ReservoirLengthY
$\text{NumPCLinesXAxisLines} = \frac{\text{PCLengthY}}{\text{PCPeriodicityA}}$

$\text{NumPCLinesYAxisLines} = \frac{\text{PCLengthX}}{\text{PCPeriodicityA}}$

$\text{NumPCLinesZAxisLines} = \frac{\text{PCLengthZ}}{\text{PCPeriodicityC}}$

$\text{NumChannelLinesXAxis} = \frac{\text{ChannelLengthX}}{\text{ChannelSpacingX}}$

$\text{NumChannelLinesZAxis} = \frac{\text{ChannelLengthZ}}{\text{ChannelSpacingZ}}$

\[
\text{PCVerticalPortLengthX} = (\text{PCNumVerticalPortsX} - 1) \times \text{PCVerticalPortSpacingX}
\]

\[
\text{PCVerticalPortLengthY} = \text{PCLengthY} - (\text{PCLengthY} \mod \text{PCVerticalPortSpacingY})
\]

\[
\text{PCNumVerticalPortsY} = \frac{\text{PCVerticalPortLengthY}}{\text{PCVerticalPortSpacingY}}
\]

\[
\text{ChannelVerticalPortLengthX} = (\text{ChannelNumVerticalPortsX} - 1) \times \text{ChannelVerticalPortSpacingX}
\]

\[
\text{ChannelVerticalPortLengthY} = \text{ChannelLengthY1} - (\text{ChannelLengthY1} \mod \text{ChannelVerticalPortSpacingY})
\]

\[
\text{Channel1VerticalPortLengthY} = \text{ChannelLengthY1} + (\text{ReservoirLengthY} / \text{ChannelExtensionFractionIntoReservoir}) - ( (\text{ChannelLengthY1} + (\text{ReservoirLengthY} / \text{ChannelExtensionFractionIntoReservoir})) \mod \text{ChannelVerticalPortSpacingY})
\]

\[
\text{Channel1NumVerticalPortsY} = \frac{\text{Channel1VerticalPortLengthY}}{\text{ChannelVerticalPortSpacingY}}
\]

\[
\text{Channel2VerticalPortLengthY} = \text{ChannelLengthY2} - (\text{ChannelLengthY2} \mod \text{ChannelVerticalPortSpacingY})
\]

\[
\text{Channel2VerticalPortLengthY} = \text{ChannelLengthY2} + (\text{ReservoirLengthY} / \text{ChannelExtensionFractionIntoReservoir}) - ( (\text{ChannelLengthY2} + (\text{ReservoirLengthY} / \text{ChannelExtensionFractionIntoReservoir})) \mod \text{ChannelVerticalPortSpacingY})
\]

\[
\text{Channel2NumVerticalPortsY} = \frac{\text{Channel2VerticalPortLengthY}}{\text{ChannelVerticalPortSpacingY}}
\]

// Adjusts the length so that it includes overlaps of the channels through BOTH reservoirs instead of just 1.

\[
\text{IF ( $\text{FLAG}_\text{Write}_\text{PC} < 1) }
\]

\[
\text{Channel1VerticalPortLengthY} = (\text{ChannelLengthY1} + 2 \times (\text{ReservoirLengthY} / \text{ChannelExtensionFractionIntoReservoir})) - ( (\text{ChannelLengthY1} + 2 \times (\text{ReservoirLengthY} / \text{ChannelExtensionFractionIntoReservoir})) \mod \text{ChannelVerticalPortSpacingY})
\]

ENDIF

\[
\text{ReservoirNumLinesXFace} = \frac{\text{ReservoirLengthX}}{\text{ReservoirSpacingX} / 2}
\]

\[
\text{ReservoirNumLinesYFace} = \frac{\text{ReservoirLengthY}}{\text{ReservoirSpacingY} / 2}
\]

\[
\text{ReservoirNumLinesZFace} = \frac{\text{ReservoirLengthZ}}{\text{ReservoirSpacingZ} / 2}
\]

\[
\text{ReservoirSectionLengthX} = \frac{\text{ReservoirLengthX}}{\text{ReservoirNumSectionsX}}
\]

\[
\text{ReservoirSectionLengthY} = \frac{\text{ReservoirLengthY}}{\text{ReservoirNumSectionsY}}
\]

\[
\text{ReservoirSectionLengthZ} = \frac{\text{ReservoirLengthZ}}{\text{ReservoirNumSectionsZ}}
\]
// TURN ME ON!
G4 F2.0 // Wait 2.0 seconds

//***Writing WG set beside reservoir***
PSOCONTROL X ON
IF ( $FLAG_Write_WGs_before_Reservoir > 0 )

MSGLAMP3 WHITE "Wr. Res. WGs"
PSOCONTROL X ON

G91 Y(+TotalChipLengthY/2 - $WGTotalLengthY) F($NonWriteSpeed) // Move to WG writing position
G90 UU($WG_Power) E($UU_Change_Speed) // Set power to write WGs

FOR $WG_For_loop_counter = 1 TO $WGMultiplier // Loop for number of WGs
IF ( $FLAG_Write_WGs_x_plus_dir > 0 )
    PSOCONTROL X OFF
    G91 X(+$WG_LengthX) F($WG_ScanSpeed) // Write one WG in positive direction
    PSOCONTROL X ON
    G91 Y(+$WGSeparation_DistanceY) F($NonWriteSpeed) // Move to the next WG position
ELSE
    PSOCONTROL X ON
    G91 X(+$WG_LengthX) F($NonWriteSpeed) // Write one WG in positive direction
ENDIF

MSGLAMP1 YELLOW "%6.3f" "Total Time (Hrs): " (Clock.X - $ClockTotalStart) / 1000 / 60 / 60 // Calculates elapsed time for running program in hours.

IF ( $FLAG_Write_WGs_x_minus_dir > 0 )
    PSOCONTROL X OFF G91 X(-$WG_LengthX) F($WG_ScanSpeed) // Write on WG in negative direction
    PSOCONTROL X ON
    G91 Y(+$WGSeparation_DistanceY) F($NonWriteSpeed) // Move to the next WG position
ELSE
    PSOCONTROL X ON
    G91 X(-$WG_LengthX) F($NonWriteSpeed) // Write one WG in negative direction
ENDIF

NEXT $WG_For_loop_counter
PSOCONTROL X ON
G90 X0 Y0 Z0 F($NonWriteSpeed) // Move back to the original position
FILEWRITE $PATHNAME ";#F3 #F" "Finished writing WG Set (not through channel
or PC): (#DT #TS)"
ELSE
FILEWRITE $PATHNAME ";#F3 #F" "Did NOT write WG Set (not through channel
or PC): (#DT #TS)"
ENDIF

// ***Writing WG set through channel***
PSOCONTROL X ON
IF ( $FLAG_Write_WGs_through_Channel > 0 )
MSGLAMP3 WHITE "Wr. Channel WGs"
PSOCONTROL X ON
G91 Y(+$TotalChipLengthY/2 - $WGTotalLengthY - $WG_Separation_from_Reservoir -
$ReservoirLengthY - $ChannelLengthY1 / 2 - $WGTotalLengthY / 2) F($NonWriteSpeed)
G90 UU($WG_Power) E($UU_Change_Speed)
IF ( $FLAG_WriteWGs_ALMOST_through_Channel > 0 )
PSOCONTROL X ARM
G4 F($WaitTimeAOM) // Dwell time for PSO and AOM
ELSE
PSOCONTROL X OFF
G4 F($WaitTimeAOM) // Dwell time for PSO and AOM
ENDIF
FOR $WG_For_loop_counter = 1 TO $WGMultiplier // Loop for number of WGs
IF ( $FLAG_Write_WGs_x_plus_dir > 0 )
IF ( $FLAG_WriteWGs_ALMOST_through_Channel > 0 )
PSOCONTROL X ARM
G4 F($WaitTimeAOM) // Dwell time for PSO and AOM
ELSE
PSOCONTROL X OFF
G4 F($WaitTimeAOM) // Dwell time for PSO and AOM
ENDIF

Appendix C. G-code: IWP/Chromatographic/Waveguide Probing Device

G91 X(+$WG_LengthX) F($WG_ScanSpeed) // Write one WG in positive direction
PSOCONTROL X ON
G4 F($WaitTimeAOM) // Dwell time for PSO and AOM
G91 Y(+$WG_Separation_DistanceY) F($NonWriteSpeed) // Move to the next WG position
ELSE
PSOCONTROL X ON
G4 F($WaitTimeAOM) // Dwell time for PSO and AOM
G91 X(+$WG_LengthX) F($NonWriteSpeed) // Write one WG in positive direction
ENDIF

MSG LAMP1 YELLOW “%6.3f” ”Total Time (Hrs): “ (Clock.X - $Clock_TotalStart) / 1000 / 60 / 60 // Calculates elapsed time for running program in hours.

IF ( $FLAG_Write_WGs_x_minus_dir >0 )
IF ( $FLAG_WriteWGs_ALMOST_through_Channel >0 )
PSOCONTROL X ARM
G4 F($WaitTimeAOM) // Dwell time for PSO and AOM
ELSE
PSOCONTROL X OFF
G4 F($WaitTimeAOM) // Dwell time for PSO and AOM
ENDIF

G91 X(-$WG_LengthX) F($WG_ScanSpeed) // Write on WG in negative direction
PSOCONTROL X ON
G4 F($WaitTimeAOM) // Dwell time for PSO and AOM
G91 Y(+$WG_Separation_DistanceY) F($NonWriteSpeed) // Move to the next WG position
ELSE
PSOCONTROL X ON
G4 F($WaitTimeAOM) // Dwell time for PSO and AOM
G91 X(-$WG_LengthX) F($NonWriteSpeed) // Write on WG in negative direction
ENDIF

NEXT $WG_For_loop_counter

PSOCONTROL X ON
G90 X0 Y0 Z0 F($NonWriteSpeed) // Move back to the original position

FILEWRITE $PATHNAME ”#F3 #F” ”Finished writing WG Set through channel: (#DT #TS)”
ELSE
FILEWRITE $PATHNAME ”#F3 #F” ”Did NOT write WG Set through channel: (#DT #TS)”
ENDIF
// ***Writing WG set through PC***
PSOCONTROL X ON
IF ( ($FLAG_Write_WGs_through_PC > 0) AND ($FLAG_Write_PC > 0) )
MSG LAMP3 WHITE "Wr. PC WGs"
PSOCONTROL X ON
G91 Y(+$TotalChipLengthY/2 - $WGTotalLengthY - $WG_Separation_from_Reservoir
- $ReservoirLengthY - $ChannelLengthY1 - $PCLengthY / 2 - $WGTotalLengthY / 2)
F($NonWriteSpeed)
G90 UU($WG_Power) E($UU_Change_Speed)

IF ( $FLAG_WriteWGs_ALMOST_through_PC > 0 )
PSOCONTROL X RESET
PSOWINDOW X 1 INPUT 6
PSOOUTPUT X WINDOW
PSOWINDOW X 1 RANGE ($WG_Len gthX/2 - $PCLengthX/2 - $WGAOM_Distance_from_PC_side) ($WG_Len gthX/2 + $PCLengthX/2 + $WGAOM_Distance_from_PC_side) UNITS
PSOWINDOW X 1 ON
PSOCONTROL X ARM
ELSE
PSOCONTROL X OFF
ENDIF

FOR $WG_For_loop_counter = 1 TO $WGMultiplier
IF ( $FLAG_Write_WGs_x_plus_dir > 0 )
IF ( $FLAG_WriteWGs_ALMOST_through_Channel > 0 )
PSOCONTROL X ARM
G4 F($WaitTimeAOM) // Dwell time for PSO and AOM
ELSE
PSOCONTROL X OFF
G4 F($WaitTimeAOM) // Dwell time for PSO and AOM
ENDIF

G91 X(+$WG_LengthX) F($WG_ScanSpeed) // Write one WG in the positive direction
PSOCONTROL X ON
G4 F($WaitTimeAOM) // Dwell time for PSO and AOM
G91 Y(+$WG_Separation_DistanceY) F($NonWriteSpeed) // Move to the next WG position
ELSE
PSOCONTROL X ON
G4 F($WaitTimeAOM) // Dwell time for PSO and AOM
G91 X(+$WG_LengthX) F($NonWriteSpeed) // Write on WG in the positive direction
MSGLAMP1 YELLOW "%.3f" "Total Time (Hrs): " (Clock.X - $ClockTotalStart) / 1000 / 60 / 60 // Calculates elapsed time for running program in hours.

IF ( $FLAG_Write_WGs_x_minus_dir > 0 )
IF ( $FLAG_WriteWGs_ALMOST_through_Channel > 0 )
PSOCONTROL X ARM
G4 F($WaitTimeAOM) // Dwell time for PSO and AOM
ELSE
PSOCONTROL X OFF
G4 F($WaitTimeAOM) // Dwell time for PSO and AOM
ENDIF

G91 X(-$WG_LengthX) F($WG_ScanSpeed) // Write one WG in the negative direction
PSOCONTROL X ON
G4 F($WaitTimeAOM) // Dwell time for PSO and AOM
G91 Y(+$WG_Separation_DistanceY) F($NonWriteSpeed) // Move to the next WG position
ELSE
PSOCONTROL X ON
G4 F($WaitTimeAOM) // Dwell time for PSO and AOM
G91 X(-$WG_LengthX) F($NonWriteSpeed) // Write one WG in the negative direction
ENDIF

NEXT $WG_For_loop_counter

PSOCONTROL X ON
G90 X0 Y0 Z0 F($NonWriteSpeed) // Move back to the original position

FILEWRITE $PATHNAME "#F3 #F" "Finished writing WG Set through PC: (#DT #TS)"
ELSE
FILEWRITE $PATHNAME "#F3 #F" "Did NOT write WG Set through PC: (#DT #TS)"
ENDIF

M0 // Pause the program to check for laser alignment

FILEWRITE $PATHNAME "#F3 #F" "Writing PC"

$LayerPowerCount = -1
MSGLAMP3 WHITE "Wr. PC"
PSOCONTROL X ON
IF ( $FLAG_Write_PC >0 )
G91 Z(-$PCLengthZ/2 - $PCPeriodicityC/4) F($NonWriteSpeed)
FOR $ForLoopCounterZAxisLines = 1 TO $NumPCLinesZAxisLines
MSG Lamp1 YELLOW "%6.3f" "Total Time (Hrs): " (Clock.X - $ClockTotalStart ) / 1000 / 60 / 60 // Calculates elapsed time for running program in hours.
PSOCtrl X ON
G91 Z(+$PCPeriodicityC/4) F($NonWriteSpeed)
G90 X(+$WG_LengthX/2 - $PCLengthX/2) F($NonWriteSpeed)
G90 Y(+$TotalChipLengthY/2 - $WG_TotalLengthY - $WG_Sep_from_Reservoir
- $ReservoirLengthY - $ChannelLengthY1 - $PCLengthY - $AccelerationBuffer) F($NonWriteSpeed)
// Setting up PSO Control
PSOCtrl Y RESET
PSOWINDOW Y 1 INPUT 7
PSOOUTPUT Y WINDOW
PSOWINDOW Y 1 RANGE ($AccelerationBuffer) ($AccelerationBuffer + $PCLengthY) UNITS
PSOWINDOW Y 1 ON INVERT
$LayerPowerCount = $LayerPowerCount + 1 // Set counter to the correct layer
G90 UU($PowerArray[$LayerPowerCount]) // Set power for the respective layer
FOR $ForLoopCounterYAxisLines = 1 TO $NumPCLinesYAxisLines // Loop for number of lines in the Y direction
G91 Z(+$PCVerticalPortsLengthZ) F($NonWriteSpeed)
PSOCtrl Y ARM
G4 F($WaitTimeAOM) // Wait 0.1 seconds to arm the PSO
G91 Z(-$PCVerticalPortsLengthZ) F($NonWriteSpeed)
G91 Y(+$AccelerationBuffer + $PCLengthY + $AccelerationBuffer) F($PCWriteSpeed)
// Write one line in the Y direction
PSOCtrl X ON
G91 Y(-$AccelerationBuffer - $PCLengthY - $AccelerationBuffer) F($NonWriteSpeed)
// Move back in the Y direction
G91 X(+$PCPeriodicityA) F($NonWriteSpeed) // Move to the next line
NEXT $ForLoopCounterYAxisLines
PSOCtrl X ON
G90 X(+$WG_LengthX/2 - $PCLengthX/2 - $AccelerationBuffer) F($NonWriteSpeed)
G90 Y(+$TotalChipLengthY/2 - $WG_TotalLengthY - $WG_Sep_from_Reservoir
- $ReservoirLengthY - $ChannelLengthY1 - $PCLengthY) F($NonWriteSpeed)
G91 Z(+$PCPeriodicityC/4) F($NonWriteSpeed) // Move up by C/4
// Setting up PSO Control
PSOCONTROL X RESET
PSOWINDOW X 1 INPUT 6
PSOOUTPUT X WINDOW
PSOWINDOW X 1 RANGE ($AccelerationBuffer) ($AccelerationBuffer + $PCLengthX)
UNITS
PSOWINDOW X 1 ON INVERT

$LayerPowerCount = $LayerPowerCount + 1 // Complete one layer; add 1 to the counter
G90 UU($PowerArray[$LayerPowerCount]) // Set power for the layer

FOR $ForLoopCounterXAxisLines = 1 TO ($NumPCLinesXAxisLines - 1) // Loop for number of lines in the X direction
G91 Z(+$PCVerticalPortsLengthZ) F($NonWriteSpeed)
PSOCONTROL X ARM
G4 F($WaitTimeAOM) // Wait 0.1 seconds to arm the PSO
G91 Z(-$PCVerticalPortsLengthZ) F($NonWriteSpeed)

G91 X(+$AccelerationBuffer + $PCLengthX + $AccelerationBuffer) F($PCWriteSpeed)
// Write one line in the X direction
PSOCONTROL X ON
G91 X(-$AccelerationBuffer - $PCLengthX - $AccelerationBuffer) F($NonWriteSpeed)
// Move back in the X direction
G91 Y(+$PCPeriodicityA) F($NonWriteSpeed) // Move to the next line
NEXT $ForLoopCounterYAxisLines

PSOCONTROL X ON
G90 X(+$WGLengthX/2 - $PCLengthX/2 + $PCPeriodicityA/2) F($NonWriteSpeed)
G90 Y(+$TotalChipLengthY/2 - $WGTotalLengthY - $WG_Separation_from_Reservoir - $ReservoirLengthY - $ChannelLengthY1 - $PCLengthY - $AccelerationBuffer) F($NonWriteSpeed)
G91 Z(+$PCPeriodicityC/4) F($NonWriteSpeed) // Move up by C/4

// Setting up PSO Control
PSOCONTROL Y RESET
PSOWINDOW Y 1 INPUT 7
PSOOUTPUT Y WINDOW
PSOWINDOW Y 1 RANGE ($AccelerationBuffer) ($AccelerationBuffer + $PCLengthY)
UNITS
PSOWINDOW Y 1 ON INVERT

$LayerPowerCount = $LayerPowerCount + 1 // Complete one layer; add 1 to the counter
G90 UU($PowerArray[$LayerPowerCount]) // Set power for the layer
FOR $ForLoopCounterYAxisLines = 1 TO $NumPCLinesYAxisLines
G91 Z(+PCVerticalPortsLengthZ) F($NonWriteSpeed)
PSOCONTROL Y ARM
G4 F($WaitTimeAOM) // Wait 0.1 seconds to arm the PSO
G91 Z(-PCVerticalPortsLengthZ) F($NonWriteSpeed)
G91 Y(+AccelerationBuffer + PCLengthY + AccelerationBuffer) F(PCWriteSpeed)
// Write one line in the Y direction
PSOCONTROL X ON
G91 Y(-AccelerationBuffer - PCLengthY - AccelerationBuffer) F($NonWriteSpeed)
// Move back in the Y direction
G91 X(+PCPeriodicityA) F($NonWriteSpeed) // Move to the next line
NEXT $ForLoopCounterYAxisLines
PSOCONTROL X ON
G90 X(+WGLengthX/2 - PCLengthX/2 - AccelerationBuffer) F($NonWriteSpeed)
G90 Y(+TotalChipLengthY/2 - WGTotalLengthY - WG_Separation_from_Reservoir
- ReservoirLengthY - ChannelLengthY1 - PCLengthY + PCPeriodicityA/2)
F($NonWriteSpeed)
G91 Z(+PCPeriodicityC/4) F($NonWriteSpeed) // Move up by C/4
// Setting up PSO Control
PSOCONTROL X RESET
PSOWINDOW X 1 INPUT 6
PSOOUTPUT X WINDOW
PSOWINDOW X 1 RANGE (AccelerationBuffer) (AccelerationBuffer + PCLengthX)
UNITS
PSOWINDOW X 1 ON INVERT
$LayerPowerCount = $LayerPowerCount + 1 // Complete one layer; add 1 to the counter
G90 UU($PowerArray[$LayerPowerCount]) // set power for the layer
FOR $ForLoopCounterXAxisLines = 1 TO ($NumPCLinesXAxisLines - 1)
G91 Z(+PCVerticalPortsLengthZ) F($NonWriteSpeed)
PSOCONTROL X ARM
G4 F($WaitTimeAOM) // Wait 0.1 seconds to arm the PSO
G91 Z(-PCVerticalPortsLengthZ) F($NonWriteSpeed)
G91 X(+AccelerationBuffer + PCLengthX + AccelerationBuffer) F(PCWriteSpeed)
// Write one line in the X direction
PSOCONTROL X ON
G91 X(-AccelerationBuffer - PCLengthX - AccelerationBuffer) F($NonWriteSpeed)
// Move back in the X direction
G91 Y(+PCPeriodicityA) F($NonWriteSpeed) // Move to the next line
NEXT $ForLoopCounterYAxisLines
NEXT $ForLoopCounterZAxisLines

PSOCONTROL X ON
G90 X0 Y0 Z0 F($NonWriteSpeed) // Move back to the original position

// ***Writing Vertical Throughports above PC***
PSOCONTROL X ON
G90 UU($PCVerticalPortPower) E($UU_Change_Speed) // Set power for writing vertical access ports
G90 X(+$WG_LengthX/2 - $PCVerticalPortLengthX/2) F($NonWriteSpeed)
G90 Y(+$TotalChipLengthY/2 - $WGTotalLengthY - $WG_Separation_from_Reservoir - $ReservoirLengthY - $ChannelLengthY1 - $PCLengthY/2 - $PCVerticalPortLengthY/2) F($NonWriteSpeed)
G90 Z(-$PCLengthZ/2) F($NonWriteSpeed)

PSOCONTROL Z RESET
PSOWINDOW Z 1 INPUT 8
PSOOUTPUT Z WINDOW
PSOWINDOW Z 1 RANGE ($PCLengthZ) ($PCLengthZ + $PCVerticalPortsLengthZ) UNITS
PSOWINDOW Z 1 ON INVERT
PSOCONTROL Z ARM
G4 F($WaitTimeAOM)

FOR $VerticalPortYCounter = 1 TO $PCNumVerticalPortsY // Loop for number of vertical access ports in Y direction
FOR $VerticalPortXCounter = 1 TO $PCNumVerticalPortsX // Loop for number of vertical access ports in X direction
G91 Z(+$PCLengthZ + $PCVerticalPortsLengthZ + $AccelerationBuffer) F($PCVerticalPortScanSpeed) // Scan one track up
G91 Z(-$PCLengthZ - $PCVerticalPortsLengthZ - $AccelerationBuffer) F($PCVerticalPortScanSpeed) // Scan one track down (double pass)
G91 X(+$PCVerticalPortSpacingX) F($NonWriteSpeed) // Move to the next vertical access port in X direction
NEXT $VerticalPortXCounter

G90 X(+$WG_LengthX/2 - $PCVerticalPortLengthX/2) F($NonWriteSpeed)
G91 Y(+$PCVerticalPortSpacingY) F($NonWriteSpeed) // Move to the next vertical access port in Y direction
NEXT $VerticalPortYCounter

PSOCONTROL X ON
G90 X0 Y0 Z0 F($NonWriteSpeed) // Move back to the original position
```plaintext
FILEWRITE $PATHNAME "#F3 #F" "Finished writing PC: (#DT #TS)"
ELSE
FILEWRITE $PATHNAME "#F3 #F" "Did NOT write PC or vertical throughports through PC: (#DT #TS)"
ENDIF
M0 // Pause the program to check for laser alignment

/*/*/*/*/*/*/*/*/*/*/*/*/*/*/*/*/*/*/*/*/*/*/*/*/ // WRITING CHANNELS
PSOCONTROL X ON
IF ( $FLAG_Write_Channel_1 > 0 )
MSGLAMP3 WHITE "Wr. Ch. 1"
PSOCONTROL X ON
G90 UU($ChannelPowerBoundary) E($UU_Change_Speed) // Set power for the outside boundary of channel
G90 X(+$WG_LengthX/2 - $ChannelLengthX/2) F($NonWriteSpeed)
IF ( $FLAG_Write_PC > 0 )
G90 Y(+$TotalChipLengthY/2 - $WGTotalLengthY - $WG_Separation_from_Reservoir - $ReservoirLengthY - $ChannelLengthY1 - $AccelerationBuffer) F($NonWriteSpeed)
ELSE
G90 Y(+$TotalChipLengthY/2 - $WGTotalLengthY - $WG_Separation_from_Reservoir - $ReservoirLengthY - $ChannelLengthY1 - $ReservoirLengthY/$ChannelExtensionFractionIntoReservoir - $ChannelLengthY1 - $AccelerationBuffer) F($NonWriteSpeed)
ENDIF
G91 Z(-$ChannelLengthZ/2) F($NonWriteSpeed)
PSOCONTROL Y RESET
PSOWINDOW Y 1 INPUT 7
PSOOUTPUT Y WINDOW
IF ( $FLAG_Write_PC > 0 )
PSOWINDOW Y 1 RANGE ($AccelerationBuffer) ($AccelerationBuffer + $ChannelLengthY1 + $ReservoirLengthY/$ChannelExtensionFractionIntoReservoir) UNITS
ELSE
PSOWINDOW Y 1 RANGE ($AccelerationBuffer) ($AccelerationBuffer + $ReservoirLengthY/$ChannelExtensionFractionIntoReservoir + $ChannelLengthY1 + $ReservoirLengthY/4) UNITS
ENDIF
PSOWINDOW Y 1 ON INVERT
G4 F($WaitTimeAOM)
FOR $ForLoopCounterZAxisLines = 1 TO $NumChannelLinesZAxis // Loop for number of layers in the z direction
```

FOR $ForLoopCounterXAxisLines = 1 TO $NumChannelLinesXAxis // Loop for number of lines in the x direction
MSG LAMP1 YELLOW "%6.3f" "Total Time (Hrs): " (Clock.X - $ClockTotalStart) / 1000 / 60 / 60 // Calculates elapsed time for running program in hours.

PSOCONTROL Y ON
IF ( $ForLoopCounterZAxisLines <= $ChannelNumberBoundaryLines )
G90 UU($ChannelPowerBoundary) E($UU_Change_Speed) // Set power to write channel outside boundary
ELSE IF ( $ForLoopCounterZAxisLines >= $NumChannelLinesZAxis - $ChannelNumberBoundaryLines + 1 )
G90 UU($ChannelPowerBoundary) E($UU_Change_Speed) // Set power to write channel outside boundary
ELSE IF ( $ForLoopCounterXAxisLines <= $ChannelNumberBoundaryLines )
G90 UU($ChannelPowerBoundary) E($UU_Change_Speed) // Set power to write channel outside boundary
ELSE IF ( $ForLoopCounterXAxisLines >= $NumChannelLinesXAxis - $ChannelNumberBoundaryLines + 1 )
G90 UU($ChannelPowerBoundary) E($UU_Change_Speed) // Set power to write channel outside boundary
ELSE
G90 UU($ChannelPowerInner) E($UU_Change_Speed) // Set power to write channel inside
ENDIF
G91 Z(+$PCVerticalPortsLengthZ) F($NonWriteSpeed)
PSOCONTROL Y ARM
G4 F($WaitTimeAOM) // Dwell time for PSO and AOM
G91 Z(-$PCVerticalPortsLengthZ) F($NonWriteSpeed)
IF ( $FLAG_Write_PC > 0 )
G91 Y(+$AccelerationBuffer + $ChannelLengthY1 + $ReservoirLengthY / $ChannelExtensionFractionIntoReservoir + $AccelerationBuffer) F($ChannelScanSpeed)
ELSE
G91 Y(+$AccelerationBuffer + $ReservoirLengthY / $ChannelExtensionFractionIntoReservoir + $ChannelLengthY1 + $ReservoirLengthY / $ChannelExtensionFractionIntoReservoir + $AccelerationBuffer) F($ChannelScanSpeed)
ENDIF
PSOCONTROL Y ON
IF ( $FLAG_Write_PC > 0 )
G91 Y(-$AccelerationBuffer - $ChannelLengthY1 - $ReservoirLengthY / $ChannelExtensionFractionIntoReservoir - $AccelerationBuffer) F($NonWriteSpeed)
ELSE
G91 Y(-$AccelerationBuffer - $ReservoirLengthY/$ChannelExtensionFractionIntoReservoir
- $ChannelLengthY1 - $ReservoirLengthY/$ChannelExtension FractionIntoReservoir - $Acc-
celerationBuffer) F($NonWriteSpeed)
ENDIF

G91 X(+$ChannelSpacingX) F($NonWriteSpeed) // Move to the next line in X direction
NEXT $ForLoopCounterXAxisLines

G90 X(+$WG LengthX/2 - $ChannelLengthX/2) F($NonWriteSpeed)
G91 Z(+$ChannelSpacingZ) F($NonWriteSpeed)
NEXT $ForLoopCounterXAxisLines

PSOCONTROL X ON
G90 X0 Y0 Z0 F($NonWriteSpeed) // Move back to the original position

FILEWRITE $PATHNAME "#F3 #F" "Finished writing Channel 1: (#DT #TS)"

// ***Writing Vertical Throughports above Channel 1***
PSOCONTROL X ON
G90 UU($ChannelVerticalPortPower) E($UU Change Speed) // Set power to write ver-
tical access ports
G90 X(+$WG LengthX/2 - $ChannelVerticalPortLengthX/2) F($NonWriteSpeed)
G90 Y(+$TotalChipLengthY/2 - $WGTotalLengthY - $WG Separation from Reservoir
- $ReservoirLengthY - $ChannelLengthY1/2 - $Channel1VerticalPortLengthY/2)
F($NonWriteSpeed)
G90 Z(-$ChannelLengthZ/2) F($NonWriteSpeed)

PSOCONTROL Z RESET
PSOWINDOW Z 1 INPUT 8
PSOOUTPUT Z WINDOW
PSOWINDOW Z 1 RANGE ($ChannelLengthZ) ($ChannelLengthZ + $ChannelVerti-
calPortLengthZ) UNITS
PSOWINDOW Z 1 ON INVERT
PSOCONTROL Z ARM
G4 F($WaitTimeAOM) // Dwell time for PSO and AOM

FOR $VerticalPortYCounter = 1 TO $Channel1NumVerticalPortsY // Loop for number
of vertical access ports in Y direction
FOR $VerticalPortXCounter = 1 TO $ChannelNumVerticalPortsX // Loop for number
of vertical access ports in X direction
MSGLAMP1 YELLOW "%.6f" "Total Time (Hrs): " (Clock.X - $ClockTotalStart) / 1000 / 60 / 60 // Calculates elapsed time for running program in hours.

G91 Z(+$ChannelLengthZ + $ChannelVerticalPortLengthZ + $AccelerationBuffer)
F($ChannelVerticalPortScanSpeed) // Write one line up
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G91 Z(-$ChannelLengthZ - $ChannelVerticalPortLengthZ - $AccelerationBuffer) F($ChannelVerticalPortScanSpeed) // Write one line down (double pass)
G91 X(+$ChannelVerticalPortSpacingX) F($NonWriteSpeed) // Move to the next vertical port in X direction
NEXT $VerticalPortXCounter

G90 X(+$WGLengthX/2 - $ChannelVerticalPortLengthX/2) F($NonWriteSpeed)
G91 Y(+$ChannelVerticalPortSpacingY) F($NonWriteSpeed) // Move to the next vertical port in Y direction
NEXT $VerticalPortYCounter

PSOCONTROL X ON
G90 X0 Y0 Z0 F($NonWriteSpeed) // Move back to the original position

FILEWRITE $PATHNAME ”#F3 #F” ”Finished writing vertical throughports through channel 1: (#DT #TS)”
ELSE
FILEWRITE $PATHNAME ”#F3 #F” ”Did not write Channel 1 or vertical throughports through Channel 1: (#DT #TS)”
ENDIF

PSOCONTROL X ON
IF ( ($FLAG_Write_PC >0) AND ($FLAG_Write_Channel_2 >0) )
MSGLAMP3 WHITE ”Wr. Ch. 2”

PSOCONTROL X ON
G90 UU($ChannelPowerBoundary) E($UU_Change_Speed) // Set power for channel outside boundary
G90 X(+$WG_LengthX/2 - $ChannelLengthX/2) F($NonWriteSpeed)
G90 Y(+$TotalChipLengthY/2 - $WG_TotalLengthY - $WG_Separation_from_Reservoir - $ReservoirLengthY - $ChannelLengthY1 - $PCLengthY - $ChannelLengthY2 - $ReservoirLengthY/$ChannelExtensionFractionIntoReservoir - $AccelerationBuffer) F($NonWriteSpeed)
G91 Z(-$ChannelLengthZ/2) F($NonWriteSpeed)

PSOCONTROL Y RESET
PSOWINDOW Y 1 INPUT 7
PSOOUTPUT Y WINDOW
PSOWINDOW Y 1 RANGE ($AccelerationBuffer) ($AccelerationBuffer + $ReservoirLengthY/$ChannelExtensionFractionIntoReservoir + $ChannelLengthY2) UNITS
PSOWINDOW Y 1 ON INVERT
G4 F($WaitTimeAOM)

FOR $ForLoopCounterZAxisLines = 1 TO $NumChannelLinesZAxis // Loop for number of layers
FOR $ForLoopCounterXAxisLines = 1 TO $NumChannelLinesXAxis // Loop for number of lines in X direction
MSGLAMP1 YELLOW "%.3f" "Total Time (Hrs): " (Clock.X - $ClockTotalStart) / 1000 / 60 / 60 // Calculates elapsed time for running program in hours.

PSOCONTROL Y ON
IF ( $ForLoopCounterZAxisLines <= $ChannelNumberBoundaryLines )
G90 UU($ChannelPowerBoundary) E($UUChangeSpeed) // Set power for channel outside boundary
ELSE IF ( $ForLoopCounterZAxisLines >= $NumChannelLinesZAxis - $ChannelNumberBoundaryLines + 1 )
G90 UU($ChannelPowerBoundary) E($UUChangeSpeed) // Set power for channel outside boundary
ELSE IF ( $ForLoopCounterXAxisLines <= $ChannelNumberBoundaryLines )
G90 UU($ChannelPowerBoundary) E($UUChangeSpeed) // Set power for channel outside boundary
ELSE IF ( $ForLoopCounterXAxisLines >= $NumChannelLinesXAxis - $ChannelNumberBoundaryLines + 1 )
G90 UU($ChannelPowerBoundary) E($UUChangeSpeed) // Set power for channel outside boundary
ELSE
G90 UU($ChannelPowerInner) E($UUChangeSpeed) // Set power for channel inside
ENDIF

G91 Z(+$PCVerticalPortsLengthZ) F($NonWriteSpeed)
PSOCONTROL Y ARM
G4 F($WaitTimeAOM)
G91 Z(-$PCVerticalPortsLengthZ) F($NonWriteSpeed)

G91 Y(+$AccelerationBuffer + $ReservoirLengthY / $ChannelExtensionFractionIntoReservoir + $ChannelLengthY2 + $AccelerationBuffer) F($ChannelScanSpeed)
PSOCONTROL Y ON
G91 Y(-$AccelerationBuffer - $ReservoirLengthY / $ChannelExtensionFractionIntoReservoir - $ChannelLengthY2 - $AccelerationBuffer) F($NonWriteSpeed)
G91 X(+$ChannelSpacingX) F($NonWriteSpeed) // Move to the next line in X direction
NEXT $ForLoopCounterXAxisLines

G90 X(+$WGLengthX/2 - $ChannelLengthX/2) F($NonWriteSpeed)
G91 Z(+$ChannelSpacingZ) F($NonWriteSpeed) // Move to the next layer up
NEXT $ForLoopCounterXAxisLines

PSOCONTROL X ON
G90 X0 Y0 Z0 F($NonWriteSpeed) // Move back to the original position
FILEWRITE $PATHNAME "#F3 #F" "Finished writing channel 2: (#DT #TS)"

// ***Writing Vertical Throughports above Channel 2***
PSOCONTROL X ON
G90 UU($ChannelVerticalPortPower) E($UU_Change_Speed) // Set power for vertical access ports
G90 X(+$WG_LengthX/2 - $ChannelVerticalPortLengthX/2) F($NonWriteSpeed)
G90 Y(+$TotalChipLengthY/2 - $WG_TotalLengthY - $WG_Separation_from_Reservoir - $ReservoirLengthY - $ChannelLengthY1 - $PCLengthY - $ChannelLengthY2/2 - $Channel2VerticalPortLengthY/2) F($NonWriteSpeed)
G90 Z(-$ChannelLengthZ/2) F($NonWriteSpeed)

PSOCONTROL Z RESET
PSOWINDOW Z 1 INPUT 8
PSOOUTPUT Z WINDOW
PSOWINDOW Z 1 RANGE ($ChannelLengthZ) ($ChannelLengthZ + $ChannelVerticalPortLengthZ) UNITS
PSOWINDOW Z 1 ON INVERT
PSOCONTROL Z ARM
G4 F($WaitTimeAOM)

FOR $VerticalPortYCounter = 1 TO $Channel2NumVerticalPortsY // Loop for number of vertical access ports in Y direction
FOR $VerticalPortXCounter = 1 TO $ChannelNumVerticalPortsX // Loop for number of vertical access ports in X direction
MSGLAMP1 YELLOW "%.3f" "Total Time (Hrs): " (Clock.X - $ClockTotalStart) / 1000 / 60 / 60 // Calculates elapsed time for running program in hours.

G91 Z(+$ChannelLengthZ + $ChannelVerticalPortLengthZ + $AccelerationBuffer) F($ChannelVerticalPortScanSpeed) // Write one line up
G91 Z(-$ChannelLengthZ - $ChannelVerticalPortLengthZ - $AccelerationBuffer) F($ChannelVerticalPortScanSpeed) // Write one line down (double pass)
G91 X(+$ChannelVerticalPortSpacingX) F($NonWriteSpeed) // Move to the next line in X direction
NEXT $VerticalPortXCounter

G90 X(+$WG_LengthX/2 - $ChannelVerticalPortLengthX/2) F($NonWriteSpeed)
G91 Y(+$ChannelVerticalPortSpacingY) F($NonWriteSpeed) // Move to the next line in Y direction
NEXT $VerticalPortYCounter

PSOCONTROL X ON
G90 X0 Y0 Z0 F($NonWriteSpeed) // Move back to the original position

FILEWRITE $PATHNAME "#F3 #F" "Finished writing vertical throughports through
Appendix C. G-code: IWP/Chromatographic/Waveguide Probing Device

Channel 2: (#DT #TS)
ELSE
FILEWRITE $PATHNAME "#F3 #F" "Did NOT write Channel 2 or vertical through-ports through Channel 2: (#DT #TS)"
ENDIF

// ***Reservoir Boundary Face -Z***
MSGLAMP1 YELLOW "%6.3f" "Total Time (Hrs): " (Clock.X - $ClockTotalStart) / 1000 / 60 / 60 // Calculates elapsed time for running program in hours.
PSOCONTROL X ON
G90 UU($ReservoirPowerBoundary) E($UU_Change_Speed) // Set power for reservoir outside boundary
G90 X(+$WG_LengthX/2 - $ReservoirLengthX/2) F($NonWriteSpeed)
G90 Y(+$TotalChipLengthY/2 - $WGTotalLengthY - $WG_Separation_from_Reservoir - $ReservoirLengthY - $AccelerationBuffer) F($NonWriteSpeed)
G90 Z(-$ChannelLengthZ/2) F($NonWriteSpeed)

PSOCONTROL Y RESET
PSOWINDOW Y 1 INPUT 7
PSOOUTPUT Y WINDOW
PSOWINDOW Y 1 RANGE ($AccelerationBuffer) ($AccelerationBuffer + $Reservoir-LengthY) UNITS
PSOWINDOW Y 1 ON INVERT
PSOCONTROL Y ARM
G4 F($WaitTimeAOM)

FOR $ForLoopCounterZAxisLines = 1 TO $ReservoirNumBoundaryLayers // Loop for number of lines in Z direction
FOR $ForLoopCounterXAxisLines = 1 TO $ReservoirNumLinesXFace // Loop for number of lines in X direction
G91 Y(+$AccelerationBuffer + $ReservoirLengthY + $AccelerationBuffer) F($ReservoirScanSpeed)
G91 X(+$ReservoirSpacingX) F($NonWriteSpeed)
G91 Y(-$AccelerationBuffer - $ReservoirLengthY - $AccelerationBuffer) F($ReservoirScanSpeed)
G91 X(+$ReservoirSpacingX) F($NonWriteSpeed)
NEXT $ForLoopCounterXAxisLines

G90 X(+$WG_LengthX/2 - $ReservoirLengthX/2) F($NonWriteSpeed)
G91 Z(+$ReservoirSpacingZ) F($NonWriteSpeed)
NEXT $ForLoopCounterZAxisLines

PSOCONTROL X ON
G90 X0 Y0 Z0 F($NonWriteSpeed) // Move back to the original position

// ***Reservoir Boundary Face -X***
MSGLAMP1 YELLOW "%6.3f" "Total Time (Hrs): " (Clock.X - $ClockTotalStart) / 1000 / 60 / 60 // Calculates elapsed time for running program in hours.
PSOCONTROL X ON
G90 UU($ReservoirPowerBoundary) E($UU_Change_Speed) // Set power for reservoir outside boundary
G90 X(+$WG_LengthX/2 - $ReservoirLengthX/2) F($NonWriteSpeed)
G90 Y(+$TotalChipLengthY/2 - $WGTotalLengthY - $WG_Separation_from_Reservoir - $ReservoirLengthY - $AccelerationBuffer) F($NonWriteSpeed)
G90 Z(-$ChannelLengthZ/2) F($NonWriteSpeed)

PSOCONTROL Y RESET
PSOWINDOW Y 1 INPUT 7
PSOOUTPUT Y WINDOW
PSOWINDOW Y 1 RANGE ($AccelerationBuffer) ($AccelerationBuffer + $Reservoir-LengthY) UNITS
PSOWINDOW Y 1 ON INVERT
PSOCONTROL Y ARM
G4 F($WaitTimeAOM)

FOR $ForLoopCounterXAxisLines = 1 TO $ReservoirNumBoundaryLayers // Loop for number of lines in X direction
FOR $ForLoopCounterZAxisLines = 1 TO $ReservoirNumLinesZFace // Loop for number of lines in Z direction
G91 Y(+$AccelerationBuffer + $ReservoirLengthY + $AccelerationBuffer) F($ReservoirScanSpeed)
G91 Z(+$ReservoirSpacingZ) F($NonWriteSpeed)
G91 Y(-$AccelerationBuffer - $ReservoirLengthY - $AccelerationBuffer) F($ReservoirScanSpeed)
G91 Z(+$ReservoirSpacingZ) F($NonWriteSpeed)
NEXT $ForLoopCounterZAxisLines

G90 Z(-$ChannelLengthZ/2) F($NonWriteSpeed)
G91 X(+$ReservoirSpacingX) F($NonWriteSpeed)
NEXT $ForLoopCounterXAxisLines

PSOCONTROL X ON
G90 X0 Y0 Z0 F($NonWriteSpeed) // Move back to the original position
// ***Reservoir Boundary Face +X***
MSGLAMP1 YELLOW "%6.3f" "Total Time (Hrs): " (Clock.X - $ClockTotalStart) / 1000 / 60 / 60 // Calculates elapsed time for running program in hours.
PSOCONTROL X ON
G90 UU($ReservoirPowerBoundary) E($UU_Change_Speed) // Set power for channel outside boundary
G90 X($NonWriteSpeed) // Set power for channel outside boundary
G90 X(+$WG_LengthX/2 + $ReservoirLengthX/2) F($NonWriteSpeed)
G90 Y(+$TotalChipLengthY/2 - $WGTotalLengthY - $WG_Separation_from_Reservoir - $ReservoirLengthY - $AccelerationBuffer) F($NonWriteSpeed)
G90 Z(-$ChannelLengthZ/2) F($NonWriteSpeed)
PSOCONTROL Y RESET
PSOWINDOW Y 1 INPUT 7
PSOOUTPUT Y WINDOW
PSOWINDOW Y 1 RANGE ($AccelerationBuffer) ($AccelerationBuffer + $ReservoirLengthY) UNITS
PSOWINDOW Y 1 ON INVERT
PSOCONTROL Y ARM
G4 F($WaitTimeAOM)
FOR $ForLoopCounterXAxisLines = 1 TO $ReservoirNumBoundaryLayers // Loop for number of lines in X direction
FOR $ForLoopCounterZAxisLines = 1 TO $ReservoirNumLinesZFace // Loop for number of lines in Z direction
G91 Y(+$AccelerationBuffer + $ReservoirLengthY + $AccelerationBuffer) F($ReservoirScanSpeed)
G91 Z(+$ReservoirSpacingZ) F($NonWriteSpeed)
G91 Y(-$AccelerationBuffer - $ReservoirLengthY - $AccelerationBuffer) F($ReservoirScanSpeed)
G91 Z(+$ReservoirSpacingZ) F($NonWriteSpeed)
NEXT $ForLoopCounterZAxisLines
G90 X(-$ReservoirSpacingX) F($NonWriteSpeed)
NEXT $ForLoopCounterXAxisLines
PSOCONTROL X ON
G90 X0 Y0 Z0 F($NonWriteSpeed) // Move back to the original position

// ***Reservoir Boundary Face -Y***
MSGLAMP1 YELLOW "%6.3f" "Total Time (Hrs): " (Clock.X - $ClockTotalStart) / 1000 / 60 / 60 // Calculates elapsed time for running program in hours.
PSOCONTROL X ON
G90 UU($ReservoirPowerBoundary) E($UU_Change_Speed) // Set power for channel outside boundary
G90 X(+$WG\_LengthX/2 - $ReservoirLengthX/2 - $AccelerationBuffer) F($NonWriteSpeed)
G90 Y(+$TotalChipLengthY/2 - $WTotX - $ReservoirLengthY - $ReservoirLengthY) F($NonWriteSpeed)
G90 Z(-$ChannelLengthZ/2) F($NonWriteSpeed)

PSOCONTROL X RESET
PSOWINDOW X 1 INPUT 6
PSOOUTPUT X WINDOW
PSOWINDOW X 1 RANGE ($AccelerationBuffer) ($AccelerationBuffer + $ReservoirLengthX) UNITS
PSOWINDOW X 1 ON INVERT
PSOCONTROL X ARM
G4 F($WaitTimeAOM)

FOR $ForLoopCounterYAxisLines = 1 TO $ReservoirNumBoundaryLayers // Loop for number of lines in Y direction
FOR $ForLoopCounterZAxisLines = 1 TO $ReservoirNumLinesZFace // Loop for number of lines in Z direction
G91 X(+$AccelerationBuffer + $ReservoirLengthX + $AccelerationBuffer) F($ReservoirScanSpeed)
G91 Z(+$ReservoirSpacingZ) F($NonWriteSpeed)
G91 X(-$AccelerationBuffer - $ReservoirLengthX - $AccelerationBuffer) F($ReservoirScanSpeed)
G91 Z(+$ReservoirSpacingZ) F($NonWriteSpeed)
NXT $ForLoopCounterZAxisLines
G90 Z(-$ChannelLengthZ/2) F($NonWriteSpeed)
G91 Y(+$ReservoirSpacingX) F($NonWriteSpeed)
NXT $ForLoopCounterYAxisLines

PSOCONTROL X ON
G90 X0 Y0 Z0 F($NonWriteSpeed) // Move back to the original position

// ***Reservoir Boundary Face +Y***
MSGGLAMP1 YELLOW "%6.3f" "Total Time (Hrs): " (Clock.X - $ClockTotalStart) / 1000 / 60 / 60 // Calculates elapsed time for running program in hours.
PSOCONTROL X ON
G90 UU($ReservoirPowerBoundary) E($UU\_Change\_Speed) // Set power for channel outside boundary
G90 X(+$WG\_LengthX/2 - $ReservoirLengthX/2 - $AccelerationBuffer) F($NonWriteSpeed)
G90 Y(+$TotalChipLengthY/2 - $WTotX - $ReservoirLengthY - $ReservoirLengthY) F($NonWriteSpeed)
G90 Z(-$ChannelLengthZ/2) F($NonWriteSpeed)
Appendix C. G-code: IWP/Chromatographic/Waveguide Probing Device

PSOCONTROL X RESET
PSOWINDOW X 1 INPUT 6
PSOUTPUT X WINDOW
PSWINDOW X 1 RANGE ($AccelerationBuffer) ($AccelerationBuffer + $Reservoir-LengthX) UNITS
PSWINDOW X 1 ON INVERT
PSOCONTROL X ARM
G4 F($WaitTimeAOM)

FOR $ForLoopCounterYAxisLines = 1 TO $ReservoirNumBoundaryLayers // Loop for number of lines in Y direction
FOR $ForLoopCounterZAxisLines = 1 TO $ReservoirNumLinesZFace // Loop for number of lines in Z direction
G91 X(+$AccelerationBuffer + $ReservoirLengthX + $AccelerationBuffer)
F($ReservoirScanSpeed)
G91 Z(+$ReservoirSpacingZ) F($NonWriteSpeed)
G91 X(-$AccelerationBuffer - $ReservoirLengthX - $AccelerationBuffer)
F($ReservoirScanSpeed)
G91 Z(+$ReservoirSpacingZ) F($NonWriteSpeed)
NEXT $ForLoopCounterZAxisLines

G90 Z(-$ChannelLengthZ/2) F($NonWriteSpeed)
G91 Y(-$ReservoirSpacingX) F($NonWriteSpeed)
NEXT $ForLoopCounterYAxisLines

PSOCONTROL X ON
G90 X0 Y0 Z0 F($NonWriteSpeed) // Move back to the original position
FILEWRITE $PATHNAME ”#F3 #F” ”Finished writing Boundary of Reservoir 1: (#DT #TS)”

// ***Inner Reservoir Z-planes***
MSGLAMP1 YELLOW ”%6.3f” ”Total Time (Hrs): ” (Clock.X - $ClockTotalStart ) / 1000 / 60 / 60 // Calculates elapsed time for running program in hours.
MSGLAMP3 WHITE ”Wr. Inner Res 1”

PSOCONTROL X ON
G90 UU($ReservoirPowerInner) E($UU_Change_Speed) // Set power for channel outside boundary
G90 X(+$WG_LengthX/2 - $ReservoirLengthX/2) F($NonWriteSpeed)
G90 Y(+$TotalChipLengthY/2 - $WTTotalLengthY - $WG_Separation_from_Reservoir - $ReservoirLengthY - $AccelerationBuffer) F($NonWriteSpeed)
G90 Z(-$ChannelLengthZ/2) F($NonWriteSpeed)

PSOCONTROL Y RESET
PSOWINDOW Y 1 INPUT 7
PSOOUTPUT Y WINDOW
PSOWINDOW Y 1 RANGE ($AccelerationBuffer) ($AccelerationBuffer + $ReservoirLengthY) UNITS
PSOWINDOW Y 1 ON INVERT
PSOCOREY ARM
G4 F($WaitTimeAOM)

FOR $ForLoopCounterZAxisLines = 1 TO ($ReservoirNumSectionsZ - 1) // Loop for number of lines in Z direction
G91 Z(+$ReservoirSectionLengthZ) F($NonWriteSpeed)

FOR $ForLoopCounterXAxisLines = 1 TO $ReservoirNumLinesXFace // Loop for number of lines in X direction
G91 Y(+$AccelerationBuffer + $ReservoirLengthY + $AccelerationBuffer) F($ReservoirScanSpeed)
G91 X(+$ReservoirSpacingX) F($NonWriteSpeed)
G91 Y(-$AccelerationBuffer - $ReservoirLengthY - $AccelerationBuffer) F($ReservoirScanSpeed)
G91 X(+$ReservoirSpacingX) F($NonWriteSpeed)
NEXT $ForLoopCounterXAxisLines

G90 X(+$WGLengthX/2 - $ReservoirLengthX/2) F($NonWriteSpeed)
NEXT $ForLoopCounterZAxisLines

PSOCOREXY ON
G90 X0 Y0 Z0 F($NonWriteSpeed) // Move back to the original position

// ***Inner Reservoir X-planes***
MSGLAMP1 YELLOW "%.3f" "Total Time (Hrs): " (Clock.X - $ClockTotalStart) / 1000 / 60 / 60 // Calculates elapsed time for running program in hours.
PSOCOREX ON
G90 UU($ReservoirPowerInner) E($UU Change Speed) // Set power for channel outside boundary
G90 X(+$WGLengthX/2 - $ReservoirLengthX/2) F($NonWriteSpeed)
G90 Y(+$TotalChipLengthY/2 - $WTLengthY - $WSeparation_from_Reservoir - $ReservoirLengthY - $AccelerationBuffer) F($NonWriteSpeed)
G90 Z(-$ChannelLengthZ/2) F($NonWriteSpeed)

PSOCOREY RESET
PSOWINDOW Y 1 INPUT 7
PSOOUTPUT Y WINDOW
PSOWINDOW Y 1 RANGE ($AccelerationBuffer) ($AccelerationBuffer + $ReservoirLengthY) UNITS
PSOWINDOW Y 1 ON INVERT
PSOCONTROL Y ARM
G4 F($WaitTimeAOM)
FOR $ForLoopCounterXAxisLines = 1 TO ($ReservoirNumSectionsX - 1) // Loop for
color of lines in X direction
G91 X(+$ReservoirSectionLengthX) F($NonWriteSpeed)
FOR $ForLoopCounterZAxisLines = 1 TO $ReservoirNumLinesZFace // Loop for num-
color of lines in Z direction
G91 Y(+$AccelerationBuffer + $ReservoirLengthY + $AccelerationBuffer)
F($ReservoirScanSpeed)
G91 Z(+$ReservoirSpacingZ) F($NonWriteSpeed)
G91 Y(-$AccelerationBuffer - $ReservoirLengthY - $AccelerationBuffer)
F($ReservoirScanSpeed)
G91 Z(+$ReservoirSpacingZ) F($NonWriteSpeed)
NEXT $ForLoopCounterZAxisLines
G90 Z(-$ChannelLengthZ/2) F($NonWriteSpeed)
NEXT $ForLoopCounterXAxisLines
PSOCONTROL X ON
G90 X0 Y0 Z0 F($NonWriteSpeed) // Move back to the original position
// ***Inner Reservoir Y-planes***
MSGLAMP1 YELLOW "%6.3f" "Total Time (Hrs): " (Clock.X - $ClockTotalStart) / 1000 / 60 / 60 // Calculates elapsed time for running program in hours.
PSOCONTROL X ON
G90 UU($ReservoirPowerInner) E($UU.Change.Speed) // Set power for channel outside
boundary
G90 X(+$WG.LengthX/2 - $ReservoirLengthX/2 - $AccelerationBuffer)
F($NonWriteSpeed)
G90 Y(+$TotalChipLengthY/2 - $WTotalLengthY - $WG.Separation_from_Reservoir
-$ReservoirLengthY) F($NonWriteSpeed)
G90 Z(-$ChannelLengthZ/2) F($NonWriteSpeed)
PSOCONTROL X RESET
PSWINDOW X 1 INPUT 6
PSOUTPUT X WINDOW
PSWINDOW X 1 RANGE ($AccelerationBuffer) ($AccelerationBuffer + $Reservoir-
LengthX) UNITS
PSWINDOW X 1 ON INVERT
PSOCONTROL X ARM
G4 F($WaitTimeAOM)
FOR $ForLoopCounterYAxisLines = 1 TO ($ReservoirNumSectionsY - 1) // Loop for
number of lines in Y direction
G91 Y(+$ReservoirSectionLengthY) F($NonWriteSpeed)
FOR $ForLoopCounterZAxisLines = 1 TO $ReservoirNumLinesZFace  // Loop for number of lines in Z direction
G91 X(+$AccelerationBuffer + $ReservoirLengthX + $AccelerationBuffer) F($ReservoirScanSpeed)
G91 Z(+$ReservoirSpacingZ) F($NonWriteSpeed)
G91 X(-$AccelerationBuffer - $ReservoirLengthX - $AccelerationBuffer) F($ReservoirScanSpeed)
G91 Z(+$ReservoirSpacingZ) F($NonWriteSpeed)
NEXT $ForLoopCounterZAxisLines
G90 Z(-$ChannelLengthZ/2) F($NonWriteSpeed)
NEXT $ForLoopCounterYAxisLines
PSOCONTROL X ON
G90 X0 Y0 Z0 F($NonWriteSpeed)  // Move back to the original position
FILEWRITE $PATHNAME ”#F3 #F” ”Finished writing interior of Reservoir 1: (#DT #TS)”
ELSE
FILEWRITE $PATHNAME ”#F3 #F” ”Did NOT write Reservoir 1: (#DT #TS)”
ENDIF
PSOCONTROL X ON
IF ( $FLAG_Write_Reservoir_2 > 0 )
MSGLAMP3 WHITE ”Wr. Res Bdy 2”
// ***Reservoir Boundary Face -Z***
MSGLAMP1 YELLOW ”%6.3f” ”Total Time (Hrs): ” (Clock.X - $ClockTotalStart) / 1000 / 60 / 60 // Calculates elapsed time for running program in hours.
PSOCONTROL X ON
G90 UU($ReservoirPowerBoundary) E($UU_Change_Speed)  // Set power for channel outside boundary
G90 X(+$WG_LengthX/2 - $ReservoirLengthX/2) F($NonWriteSpeed)
G90 Y(+$TotalChipLengthY/2 - $WG_TotalLengthY - $WG_Sep_from_Reservoir - $ReservoirLengthY - $ChannelLengthY1 - $FLAG_Write_PC*(SPCLengthY + $ChannelLengthY2) - $ReservoirLengthY - $AccelerationBuffer) F($NonWriteSpeed)
G90 Z(-$ChannelLengthZ/2) F($NonWriteSpeed)
PSOCONTROL Y RESET
PSOWINDOW Y 1 INPUT 7
PSOUTPUT Y WINDOW
PSWINDOW Y 1 RANGE ($AccelerationBuffer) ($AccelerationBuffer + $Reservoir-
LengthY) UNITS
PSOWINDOW Y 1 ON INVERT
PSOCONTROL Y ARM
G4 F($WaitTimeAOM)

FOR $ForLoopCounterZAxisLines = 1 TO $ReservoirNumBoundaryLayers // Loop for
number of lines in Z direction
FOR $ForLoopCounterXAxisLines = 1 TO $ReservoirNumLinesXFace // Loop for num-
ber of lines in X direction
G91 Y(+$AccelerationBuffer + $ReservoirLengthY + $AccelerationBuffer)
F($ReservoirScanSpeed)
G91 X(+$ReservoirSpacingX) F($NonWriteSpeed)
G91 Y(-$AccelerationBuffer - $ReservoirLengthY - $AccelerationBuffer)
F($ReservoirScanSpeed)
G91 X(+$ReservoirSpacingX) F($NonWriteSpeed)
NEXT $ForLoopCounterXAxisLines

G90 X(+$WGLengthX/2 - $ReservoirLengthX/2) F($NonWriteSpeed)
G91 Z(+$ReservoirSpacingZ) F($NonWriteSpeed)
NEXT $ForLoopCounterZAxisLines

PSOCONTROL X ON
G90 X0 Y0 Z0 F($NonWriteSpeed) // Move back to the original position

// ***Reservoir Boundary Face -X***
MSGLAMP1 YELLOW "%6.3f" "Total Time (Hrs): " (Clock.X - $ClockTotalStart ) / 1000 / 60 / 60 // Calculates elapsed time for running program in hours.
PSOCONTROL X ON
G90 UU($ReservoirPowerBoundary) E($UUChangeSpeed) // Set power for channel
outside boundary
G90 X(+$WGLengthX/2 - $ReservoirLengthX/2) F($NonWriteSpeed)
G90 Y(+$TotalChipLengthY/2 - $WGTotallengthY - $WG_Separation_from_Reservoir
- $ReservoirLengthY - $ChannelLengthY1 - $FLAG_Write_PC*($PCLengthY + $Chan-
nelLengthY2) - $ReservoirLengthY - $AccelerationBuffer) F($NonWriteSpeed)
G90 Z(-$ChannelLengthZ/2) F($NonWriteSpeed)

PSOCONTROL Y RESET
PSOWINDOW Y 1 INPUT 7
PSOOUTPUT Y WINDOW
PSOWINDOW Y 1 RANGE ($AccelerationBuffer) ($AccelerationBuffer + $Reservoir-
LengthY) UNITS
PSOWINDOW Y 1 ON INVERT
PSOCONTROL Y ARM
G4 F($WaitTimeAOM)
FOR $ForLoopCounterXAxisLines = 1 TO $ReservoirNumBoundaryLayers // Loop for
total number of lines in X direction
FOR $ForLoopCounterZAxisLines = 1 TO $ReservoirNumLinesZFace // Loop for num-
ter number of lines in Z direction
G91 Y(+$AccelerationBuffer + $ReservoirLengthY + $AccelerationBuffer)
F($ReservoirScanSpeed)
G91 Z(+$ReservoirSpacingZ) F($NonWriteSpeed)
G91 Y(-$AccelerationBuffer - $ReservoirLengthY - $AccelerationBuffer)
F($ReservoirScanSpeed)
G91 Z(+$ReservoirSpacingZ) F($NonWriteSpeed)
NEXT $ForLoopCounterZAxisLines
G90 Z(-$ChannelLengthZ/2) F($NonWriteSpeed)
G91 X(+$ReservoirSpacingX) F($NonWriteSpeed)
NEXT $ForLoopCounterXAxisLines
PSOCONTROL X ON
G90 X0 Y0 Z0 F($NonWriteSpeed) // Move back to the original position
// ***Reservoir Boundary Face +X***
MSGLAMP1 YELLOW "%.3f" "Total Time (Hrs): " (Clock.X - $ClockTotalStart ) / 
1000 / 60 / 60 // Calculates elapsed time for running program in hours.
PSOCONTROL X ON
G90 UU($ReservoirPowerBoundary) E($UU_Change_Speed) // Set power for channel
outside boundary
G90 X(+$WG_LengthX/2 + $ReservoirLengthX/2) F($NonWriteSpeed)
G90 Y(+$TotalChipLengthY/2 - $WGTotalLengthY - $WG_Separation_from_Reservoir
- $ReservoirLengthY - $ChannelLengthY1 - $FLAG_Write_PC*($PCLengthY + $Channel
LengthY2) - $ReservoirLengthY - $AccelerationBuffer) F($NonWriteSpeed)
G90 Z(-$ChannelLengthZ/2) F($NonWriteSpeed)
PSOCONTROL Y RESET
PSOWINDOW Y 1 INPUT 7
PSOOUTPUT Y WINDOW
PSOWINDOW Y 1 RANGE ($AccelerationBuffer) ($AccelerationBuffer + $Reservoir-
LengthY) UNITS
PSOWINDOW Y 1 ON INVERT
PSOCONTROL Y ARM
G4 F($WaitTimeAOM)
FOR $ForLoopCounterXAxisLines = 1 TO $ReservoirNumBoundaryLayers // Loop for
total number of lines in X direction
FOR $ForLoopCounterZAxisLines = 1 TO $ReservoirNumLinesZFace // Loop for num-
ter number of lines in Z direction
G91 Y(+$AccelerationBuffer + $ReservoirLengthY + $AccelerationBuffer)
F(\text{$\text{ReservoirScanSpeed}$})
G91 Z(+\text{$\text{ReservoirSpacingZ}$}) F(\text{$\text{NonWriteSpeed}$})
G91 Y(-\text{$\text{AccelerationBuffer}$} - \text{$\text{ReservoirLengthY}$} - \text{$\text{AccelerationBuffer}$})
F(\text{$\text{ReservoirScanSpeed}$})
G91 Z(+\text{$\text{ReservoirSpacingZ}$}) F(\text{$\text{NonWriteSpeed}$})
NEXT $\text{ForLoopCounterZAxisLines}$

G90 Z(-\text{$\text{ChannelLengthZ/2}$}) F(\text{$\text{NonWriteSpeed}$})
G91 X(-\text{$\text{ReservoirSpacingX}$}) F(\text{$\text{NonWriteSpeed}$})
NEXT $\text{ForLoopCounterXAxisLines}$

PSOCONTROL X ON
G90 X0 Y0 Z0 F(\text{$\text{NonWriteSpeed}$}) // Move back to the original position

// ***Reservoir Boundary Face -Y***
MSGLAMP1 YELLOW "\text{\text{"%6.3f" Total Time (Hrs): " (Clock.X - $\text{ClockTotalStart}$) / 1000 / 60 / 60 }" Calculates elapsed time for running program in hours.
PSOCONTROL X ON
G90 UU(\text{$\text{ReservoirPowerBoundary}$}) E(\text{$\text{$\text{UU}\_\text{Change\_Speed}$}$}) // Set power for channel outside boundary
G90 X(+\text{\text{SWG\_LengthX/2} - $\text{ReservoirLengthX/2} - \text{\text{AccelerationBuffer}$})
F(\text{$\text{NonWriteSpeed}$})
G90 Y(+\text{$\text{TotalChipLengthY/2} - $\text{WG\_TotalLengthY} - $\text{WG\_Separation\_from\_Reservoir}$ - $\text{ReservoirLengthY} - $\text{ChannelLengthY1} - $\text{\text{FLAG\_Write\_PC}\_($\text{PC\_LengthY} + $\text{ChannelLengthY2})} - $\text{ReservoirLengthY}$}) F(\text{$\text{NonWriteSpeed}$})
G90 Z(-\text{$\text{ChannelLengthZ/2}$}) F(\text{$\text{NonWriteSpeed}$})

PSOCONTROL X RESET
PSOWINDOW X 1 INPUT 6
PSOOUTPUT X WINDOW
PSOWINDOW X 1 RANGE ($\text{AccelerationBuffer}$) ($\text{AccelerationBuffer} + $\text{ReservoirLengthX}$) UNITS
PSOWINDOW X 1 ON INVERT
PSOCONTROL X ARM
G4 F(\text{$\text{Wait\_TimeAOM}$})

FOR $\text{ForLoopCounterYAxisLines} = 1$ TO $\text{ReservoirNumBoundaryLayers}$ // Loop for number of lines in Y direction
FOR $\text{ForLoopCounterZAxisLines} = 1$ TO $\text{ReservoirNumLinesZFace}$ // Loop for number of lines in Z direction
G91 X(+\text{$\text{AccelerationBuffer} + $\text{ReservoirLengthX} + $\text{AccelerationBuffer}$})
F($\text{ReservoirScanSpeed}$)
G91 Z(+\text{$\text{ReservoirSpacingZ}$}) F($\text{NonWriteSpeed}$)
G91 X(-$\text{$\text{AccelerationBuffer} - $\text{ReservoirLengthX} - \text{\text{AccelerationBuffer}$})
F($\text{$\text{ReservoirScanSpeed}$}$)
Appendix C. G-code: IWP/Chromatographic/Waveguide Probing Device

G91 Z(+$ReservoirSpacingZ) F($NonWriteSpeed)
NEXT $ForLoopCounterZAxisLines

G90 Z(-$ChannelLengthZ/2) F($NonWriteSpeed)
G91 Y(+$ReservoirSpacingX) F($NonWriteSpeed)
NEXT $ForLoopCounterYAxisLines

PSOCONTROL X ON
G90 X0 Y0 Z0 F($NonWriteSpeed) // Move back to the original position

// ***Reservoir Boundary Face +Y***
MSGLAMP1 YELLOW "%6.3f" "Total Time (Hrs): " (Clock.X - $ClockTotalStart) / 1000 / 60 / 60 // Calculates elapsed time for running program in hours.
PSOCONTROL X ON
G90 UU($ReservoirPowerBoundary) E($UU Change Speed) // Set power for channel outside boundary
G90 X(+$WGLengthX/2 - $ReservoirLengthX/2 - $AccelerationBuffer) F($NonWriteSpeed)
G90 Y(+$TotalChipLengthY/2 - $WGTotalLengthY - $WG Separation from Reservoir - $ReservoirLengthY - $ChannelLengthY1 - $FLAG Write PC*(PCLengthY + $ChannelLengthY2)) F($NonWriteSpeed)
G90 Z(-$ChannelLengthZ/2) F($NonWriteSpeed)

PSOCONTROL X RESET
PSOWINDOW X 1 INPUT 6
PSOOUTPUT X WINDOW
PSOWINDOW X 1 RANGE ($AccelerationBuffer) ($AccelerationBuffer + $ReservoirLengthX) UNITS
PSOWINDOW X 1 ON INVERT
PSOCONTROL X ARM
G4 F($WaitTimeAOM)

FOR $ForLoopCounterYAxisLines = 1 TO $ReservoirNumBoundaryLayers // Loop for number of lines in Y direction
FOR $ForLoopCounterZAxisLines = 1 TO $ReservoirNumLinesZFace // Loop for number of lines in Z direction
G91 X(+$AccelerationBuffer + $ReservoirLengthX + $AccelerationBuffer) F($ReservoirScanSpeed)
G91 Z(+$ReservoirSpacingZ) F($NonWriteSpeed)
G91 X(-$AccelerationBuffer - $ReservoirLengthX - $AccelerationBuffer) F($ReservoirScanSpeed)
G91 Z(+$ReservoirSpacingZ) F($NonWriteSpeed)
NEXT $ForLoopCounterZAxisLines

G90 Z(-$ChannelLengthZ/2) F($NonWriteSpeed)
G91 Y(-$ReservoirSpacingX) F($NonWriteSpeed)
NEXT $ForLoopCounterYAxisLines

PSOCONTROL X ON
G90 X0 Y0 Z0 F($NonWriteSpeed) // Move back to the original position
FILEWRITE $PATHNAME " #F3 #F" "Finished writing boundary of Reservoir 2: (#DT #TS)"

// ***Inner Reservoir Z-planes***
MSGLAMP1 YELLOW "%.3f" "Total Time (Hrs): " (Clock.X - $ClockTotalStart) / 1000 / 60 / 60 // Calculates elapsed time for running program in hours.
MSGLAMP3 WHITE "Wr. Inner Res 1"

PSOCONTROL X ON
G90 UU($ReservoirPowerInner) E($UUChangeSpeed) // Set power for channel outside boundary
G90 X(+$WGLengthX/2 - $ReservoirLengthX/2) F($NonWriteSpeed)
G90 Y(+$TotalChipLengthY/2 - $WGTotalLengthY - $WG_Separation_from_Reservoir - $ReservoirLengthY - $ChannelLengthY1 - $FLAGWritePC*(PCLengthY + $ChannelLengthY2) - $ReservoirLengthY - $AccelerationBuffer) F($NonWriteSpeed)
G90 Z(-$ChannelLengthZ/2) F($NonWriteSpeed)

PSOCONTROL Y RESET
PSOWINDOW Y 1 INPUT 7
PSOOUTPUT Y WINDOW
PSOWINDOW Y 1 RANGE ($AccelerationBuffer) ($AccelerationBuffer + $ReservoirLengthY) UNITS
PSOWINDOW Y 1 ON INVERT
PSOCONTROL Y ARM
G4 F($WaitTimeAOM)

FOR $ForLoopCounterZAxisLines = 1 TO ($ReservoirNumSectionsZ - 1) // Loop for number of lines in Z direction
G91 Z(+$ReservoirSectionLengthZ) F($NonWriteSpeed)

FOR $ForLoopCounterXAxisLines = 1 TO $ReservoirNumLinesXFace // Loop for number of lines in X direction
G91 Y(+$AccelerationBuffer + $ReservoirLengthY + $AccelerationBuffer) F($ReservoirScanSpeed)
G91 X(+$ReservoirSpacingX) F($NonWriteSpeed)
G91 Y(-$AccelerationBuffer - $ReservoirLengthY - $AccelerationBuffer) F($ReservoirScanSpeed)
G91 X(+$ReservoirSpacingX) F($NonWriteSpeed)
NEXT $ForLoopCounterXAxisLines
G90 X(+$WG.LengthX/2 - $ReservoirLengthX/2) F($NonWriteSpeed) NEXT $ForLoopCounterZAxisLines

PSOCONTROL X ON
G90 X0 Y0 Z0 F($NonWriteSpeed) // Move back to the original position

// ***Inner Reservoir X-planes***
MSG Lamp1 YELLOW "%.3f" "Total Time (Hrs): " (Clock.X - $ClockTotalStart) / 1000 / 60 / 60 // Calculates elapsed time for running program in hours.
PSOCONTROL X ON
G90 UU($ReservoirPowerInner) E(UU_Change_Speed) // Set power for channel outside boundary
G90 X(+$WG.LengthX/2 - $ReservoirLengthX/2) F($NonWriteSpeed)
G90 Y(+$TotalChipLengthY/2 - $WT TotalLengthY - $WG.Separation_from_Reservoir - $ReservoirLengthY - $ChannelLengthY1 - $FLAG_Write_PC*(SPChannelLengthY + $ChannelLengthY2) - $ReservoirLengthY - $AccelerationBuffer) F($NonWriteSpeed)
G90 Z(-$ChannelLengthZ/2) F($NonWriteSpeed)

PSOCONTROL Y RESET
PSOWINDOW Y 1 INPUT 7
PSOOUTPUT Y WINDOW
PSOWINDOW Y 1 RANGE ($AccelerationBuffer) ($AccelerationBuffer + $Reservoir-LengthY) UNITS
PSOWINDOW Y 1 ON INVERT
PSOCONTROL Y ARM
G4 F($WaitTimeAOM)

FOR $ForLoopCounterXAxisLines = 1 TO ($ReservoirNumSectionsX - 1) // Loop for number of lines in X direction
G91 X(+$ReservoirSectionLengthX) F($NonWriteSpeed)

FOR $ForLoopCounterZAxisLines = 1 TO $ReservoirNumLinesZFace // Loop for number of lines in Z direction
G91 Y(+$AccelerationBuffer + $ReservoirLengthY + $AccelerationBuffer) F($ReservoirScanSpeed)
G91 Z(+$ReservoirSpacingZ) F($NonWriteSpeed)
G91 Y(-$AccelerationBuffer - $ReservoirLengthY - $AccelerationBuffer) F($ReservoirScanSpeed)
G91 Z(+$ReservoirSpacingZ) F($NonWriteSpeed)
NEXT $ForLoopCounterZAxisLines
G90 Z(-$ChannelLengthZ/2) F($NonWriteSpeed)
NEXT $ForLoopCounterXAxisLines
PSOCONTROL X ON
G90 X0 Y0 Z0 F($NonWriteSpeed) // Move back to the original position

// ***Inner Reservoir Y-planes***
MSGLAMP1 YELLOW "%6.3f" "Total Time (Hrs): " (Clock.X - $ClockTotalStart) / 1000 / 60 / 60 // Calculates elapsed time for running program in hours.
PSOCONTROL X ON
G90 UU($ReservoirPowerInner) E($UU Change Speed) // Set power for channel outside boundary
G90 X(+$WG LengthX/2 - $ReservoirLengthX/2 - $AccelerationBuffer) F($NonWriteSpeed)
G90 Y(+$TotalChipLengthY/2 - $WGTotalLengthY - $WG Separation_from_Reservoir - $ReservoirLengthY1 - $FLAG Write PC*($PCLengthY + $ChannelLengthY2) - $ReservoirLengthY) F($NonWriteSpeed)
G90 Z(-$ChannelLengthZ/2) F($NonWriteSpeed)

PSOCONTROL X RESET
PSOWINDOW X 1 INPUT 6
PSOOUTPUT X WINDOW
PSOWINDOW X 1 RANGE ($AccelerationBuffer) ($AccelerationBuffer + $Reservoir-LengthX) UNITS
PSOWINDOW X 1 ON INVERT
PSOCONTROL X ARM
G4 F($WaitTimeAOM)
FOR $ForLoopCounterYAxisLines = 1 TO ($ReservoirNumSectionsY - 1) // Loop for number of lines in Y direction
G91 Y(+$ReservoirSectionLengthY) F($NonWriteSpeed)
FOR $ForLoopCounterZAxisLines = 1 TO $ReservoirNumLinesZFace // Loop for number of lines in Z direction
G91 X(+$AccelerationBuffer + $ReservoirLengthX + $AccelerationBuffer) F($ReservoirScanSpeed)
G91 Z(+$ReservoirSpacingZ) F($NonWriteSpeed)
G91 X(-$AccelerationBuffer - $ReservoirLengthX - $AccelerationBuffer) F($ReservoirScanSpeed)
G91 Z(+$ReservoirSpacingZ) F($NonWriteSpeed)
NEXT $ForLoopCounterZAxisLines
NEXT $ForLoopCounterYAxisLines
G90 Z(-$ChannelLengthZ/2) F($NonWriteSpeed)
NEXT $ForLoopCounterYAxisLines

PSOCONTROL X ON
G90 X0 Y0 Z0 F($NonWriteSpeed) // Move back to the original position
FILEWRITE $PATHNAME "\#F3 \#F" "Finished writing interior of Reservoir 2: (#DT #TS)"
ELSE
FILEWRITE $PATHNAME "\#F3 \#F" "Did NOT write Reservoir 2: (#DT #TS)"
ENDIF

////////////////////////////////////////////////////////////////////////////
// FINISHING PROGRAM
//
PSOCONTROL X ON
G90 UU $LowPower E$UU_Change_Speed // Set power to very low for finishing the program
MSGLAMP3 WHITE "Prgm Terminating"

FILEWRITE $PATHNAME "\#F3 \#F" "The ending time of the program is: (#DT #TS)"
FILEWRITE $PATHNAME ""

FILEWRITE $PATHNAME "WG Parameters"
FILEWRITE $PATHNAME "\#F" "WG_LengthX = " $WG_LengthX
FILEWRITE $PATHNAME "\#F" "WG_Power = " $WG_Power
FILEWRITE $PATHNAME "\#F" "WG_ScanSpeed = " $WG_ScanSpeed
FILEWRITE $PATHNAME "\#F" "WG_Separation_DistanceY = " $WG_Separation_DistanceY
FILEWRITE $PATHNAME "\#D" "WG_Multiplier = " $WG_Multiplier
FILEWRITE $PATHNAME "\#F" "WG_Separation_from_Reservoir = " $WG_Separation_from_Reservoir
FILEWRITE $PATHNAME "\#F" "WGAOM_Distance_from_Channel_side = " $WGAOM_Distance_from_Channel_side
FILEWRITE $PATHNAME "\#F" "WGAOM_Distance_from_PC_side = " $WGAOM_Distance_from_PC_side
FILEWRITE $PATHNAME ""

FILEWRITE $PATHNAME "PC Parameters"
;FILEWRITE $PATHNAME "\#F" "PC_Power = " $PC_Power
FILEWRITE $PATHNAME "\#F" "PC_PeriodicityA = " $PC_PeriodicityA
FILEWRITE $PATHNAME "\#F" "PC_PeriodicityC = " $PC_PeriodicityC
FILEWRITE $PATHNAME "\#F" "PC_WriteSpeed = " $PC_WriteSpeed
FILEWRITE $PATHNAME "\#F" "PC_LengthX = " $PC_LengthX
FILEWRITE $PATHNAME "\#F" "PC_LengthY = " $PC_LengthY
FILEWRITE $PATHNAME "\#F" "PC_LengthZ = " $PC_LengthZ
FILEWRITE $PATHNAME ""

FILEWRITE $PATHNAME "Channel Parameters"
FILEWRITE $PATHNAME "\#D" "Channel_Number_Boundary_Lines = " $Channel_Number_Boundary_Lines
berBoundaryLines

FILEWRITE $PATHNAME "#F" "ChannelPowerBoundary = " $ChannelPowerBoundary
FILEWRITE $PATHNAME "#F" "ChannelPowerInner = " $ChannelPowerInner
FILEWRITE $PATHNAME "#F" "ChannelScanSpeed = " $ChannelScanSpeed
FILEWRITE $PATHNAME "#F" "ChannelSpacingX = " $ChannelSpacingX
FILEWRITE $PATHNAME "#F" "ChannelSpacingZ = " $ChannelSpacingZ
FILEWRITE $PATHNAME "#F" "ChannelLengthX = " $ChannelLengthX
FILEWRITE $PATHNAME "#F" "ChannelLengthY1 = " $ChannelLengthY1
FILEWRITE $PATHNAME "#F" "ChannelLengthY2 = " $ChannelLengthY2
FILEWRITE $PATHNAME "#F" "ChannelExtensionFractionIntoReservoir = " $ChannelExtensionFractionIntoReservoir

FILEWRITE $PATHNAME "Reservoir Parameters"
FILEWRITE $PATHNAME "#F" "ReservoirPowerInner = " $ReservoirPowerInner
FILEWRITE $PATHNAME "#F" "ReservoirPowerBoundary = " $ReservoirPowerBoundary
FILEWRITE $PATHNAME "#F" "ReservoirScanSpeed = " $ReservoirScanSpeed
FILEWRITE $PATHNAME "#F" "ReservoirSpacingX = " $ReservoirSpacingX
FILEWRITE $PATHNAME "#F" "ReservoirSpacingY = " $ReservoirSpacingY
FILEWRITE $PATHNAME "#F" "ReservoirSpacingZ = " $ReservoirSpacingZ
FILEWRITE $PATHNAME "#D" "ReservoirNumBoundaryLayers = " $ReservoirNumBoundaryLayers
FILEWRITE $PATHNAME "#D" "ReservoirNumSectionsX = " $ReservoirNumSectionsX
FILEWRITE $PATHNAME "#D" "ReservoirNumSectionsY = " $ReservoirNumSectionsY
FILEWRITE $PATHNAME "#D" "ReservoirNumSectionsZ = " $ReservoirNumSectionsZ
FILEWRITE $PATHNAME "#F" "ReservoirLengthX = " $ReservoirLengthX
FILEWRITE $PATHNAME "#F" "ReservoirLengthY = " $ReservoirLengthY
FILEWRITE $PATHNAME "#F" "ReservoirLengthZ = " $ReservoirLengthZ

FILEWRITE $PATHNAME "Vertical Throughports PC Parameters"
FILEWRITE $PATHNAME "#F" "PCVerticalPortPower = " $PCVerticalPortPower
FILEWRITE $PATHNAME "#F" "PCVerticalPortScanSpeed = " $PCVerticalPortScanSpeed
FILEWRITE $PATHNAME "#D" "PCNumVerticalPortsX = " $PCNumVerticalPortsX
FILEWRITE $PATHNAME "#F" "PCVerticalPortSpacingX = " $PCVerticalPortSpacingX
FILEWRITE $PATHNAME "#F" "PCVerticalPortSpacingY = " $PCVerticalPortSpacingY
FILEWRITE $PATHNAME "#F" "PCVerticalPortsLengthZ = " $PCVerticalPortsLengthZ
FILEWRITE $PATHNAME ""

FILEWRITE $PATHNAME "Vertical Throughports Channel Parameters"
FILEWRITE $PATHNAME "#F" "ChannelVerticalPortPower = " $ChannelVerticalPortPower
FILEWRITE $PATHNAME "#F" "ChannelVerticalPortScanSpeed = " $ChannelVerticalPortScanSpeed
FILEWRITE $PATHNAME "#D" "ChannelNumVerticalPortsX = " $ChannelNumVerticalPortsX
FILEWRITE $PATHNAME "#F" "ChannelVerticalPortSpacingX = " $ChannelVerticalPortSpacingX
FILEWRITE $PATHNAME "#F" "ChannelVerticalPortSpacingY = " $ChannelVerticalPortSpacingY
FILEWRITE $PATHNAME "#F" "ChannelVerticalPortLengthZ = " $ChannelVerticalPortLengthZ
FILEWRITE $PATHNAME ""

FILEWRITE $PATHNAME "Other Parameters"
FILEWRITE $PATHNAME "#F" "NonWriteSpeed = " $NonWriteSpeed
FILEWRITE $PATHNAME "#F" "UU_Change_Speed = " $UU_Change_Speed
FILEWRITE $PATHNAME "#F" "GlobalZOffset = " $GlobalZOffset
FILEWRITE $PATHNAME "#F" "WaitTimeAOM = " $WaitTimeAOM
FILEWRITE $PATHNAME "#F" "AccelerationBuffer = " $AccelerationBuffer
FILEWRITE $PATHNAME "#F" "LowPower = " $LowPower
FILEWRITE $PATHNAME ""

FILEWRITE $PATHNAME "FLAGS"
FILEWRITE $PATHNAME "#D" "FLAG_Write_WGs_before_Reservoir = " $FLAG_Write_WGs_before_Reservoir
FILEWRITE $PATHNAME "#D" "FLAG_Write_WGs_through_Channel = " $FLAG_Write_WGs_through_Channel
FILEWRITE $PATHNAME "#D" "FLAG_WriteWGs_ALMOST_through_Channel = " $FLAG_WriteWGs_ALMOST_through_Channel
FILEWRITE $PATHNAME "#D" "FLAG_Write_WGs_through_PC = " $FLAG_Write_WGs_through_PC
FILEWRITE $PATHNAME "#D" "FLAG_WriteWGs_ALMOST_through_PC = " $FLAG_WriteWGs_ALMOST_through_PC
FILEWRITE $PATHNAME "#D" "FLAG_Write_WGs_x_minus_dir = " $FLAG_Write_WGs_x_minus_dir
FILEWRITE $PATHNAME "#D" "FLAG_Write_WGs_x_plus_dir = " $FLAG_Write_WGs_x_plus_dir
FILEWRITE $PATHNAME "#D" "FLAG_Write_PC = " $FLAG_Write_PC
FILEWRITE $PATHNAME "#D" "FLAG_Write_Channel_1 = " $FLAG_Write_Channel_1
" $FLAG_Write_Channel_1
FILEWRITE $PATHNAME "#D" "FLAG_Write_Channel_2 =
" $FLAG_Write_Channel_2
FILEWRITE $PATHNAME "#D" "FLAG_Write_Reservoir_1 =
" $FLAG_Write_Reservoir_1
FILEWRITE $PATHNAME "#D" "FLAG_Write_Reservoir_2 =
" $FLAG_Write_Reservoir_2
FILEWRITE $PATHNAME ""

FILECLOSE

G82 // G82 undoes a G92.
G90 X($OrigX) Y($OrigY) Z($OrigZ) F($NonWriteSpeed)
G91 Z(20) F($NonWriteSpeed) // Move away from the focal plane to avoid further modification

MSGLAMP1 YELLOW "%6.3f" "Total Time (Hrs): " (Clock.X - $ClockTotalStart) / 1000 / 60 / 60 // Calculates elapsed time for running program in hours
MSGLAMP3 WHITE "Prgm Terminating"
M2 // Program termination
Appendix D

Published Work

Journal Publications


**Conferences and Proceedings**


4. S. Ho, M. Haque, P. R. Herman, and J. S. Aitchison. Femtosecond laser-assisted etching of three dimensional woodpile micro-channel arrays in fused silica, *CLEO 2011, Baltimore, MD*, Talk CTuAA1, May 2011.


Bibliography


