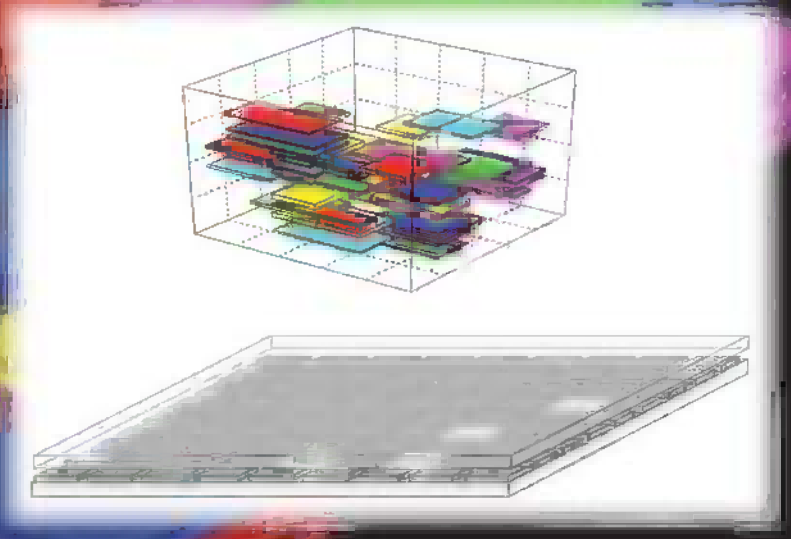
DIGITAL MICROFLUIDIC BIOCHIPS

SYNTHESIS, TESTING, AND RECONFIGURATION TECHNIQUES



Krishnendu Chakrabarty
Fei Su

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Preface

Microfluidic biochips are soon expected to revolutionize clinical diagnostics, massively parallel DNA analysis, and other laboratory procedures involving molecular biology. In contrast to continuous-flow systems, a new generation of microfluidic biochips, referred to as *digital microfluidic biochips,* offers dynamic reconfigurability and system scalability that facilitate large-scale bioassay applications. As more bioassays are executed concurrently on a biochip, system integration and design complexity are expected to increase dramatically. Moreover, as many digital microfluidic biochips will be used in safety-critical applications, defect/fault tolerance is also expected to emerge as important design consideration.

Current full-custom design techniques for digital microfluidic biochips may not scale well for large design, and they do not easily handle yield/reliability issues. There is a need to deliver the same level of computer-aided design (CAD) support to the biochip designer that the semiconductor industry now takes for granted. This book describes a design automation framework that addresses key issues in the synthesis, testing, and reconfiguration of digital microfluidic biochips.

Part I of this book presents synthesis techniques for digital microfluidic biochips. It first presents an architectural-level synthesis methodology that addresses the optimization problem of bioassay scheduling under resource constraints. Following architectural-level synthesis, two key problems in geometry-level synthesis, namely microfluidic module placement and droplet routing, are investigated. A unified synthesis methodology that integrates operation scheduling, resource binding, and module placement in one synthesis procedure is also presented.

Part II of this book investigates testing techniques for digital microfluidic biochips. A cost-effective test methodology, whereby faults can be detected by electrically controlling and tracking the motion of test droplets, is first proposed. Based on the proposed detection mechanism, the problems of test planning and test resource optimization are further investigated. A concurrent testing methodology is also developed to allow the detection of catastrophic faults and normal bioassays to run simultaneously on a biochip. A defect-oriented testing and diagnosis method is presented to handle different types of defects in microfluidic arrays.

Reconfiguration techniques are the focus of Part III of the book. Different reconfiguration techniques and the corresponding defect/fault tolerance approaches are presented. Two defect/fault tolerance schemes based on space redundancy and graceful degradation, respectively, are analyzed. The proposed reconfiguration techniques are expected to increase the yield and reliability of digital microfluidic biochips. The proposed design automation tools are evaluated using a set of real-life bioassays.

This book grew out of an ongoing research project on CAD for biochips at Duke University. The results of this research have been published as papers in a number

of journals and conference proceedings. The chapters in this book present all these results as a research monograph in a single volume. It can be used as a reference book for academic and industrial researchers in the areas of digital microfluidic biochips and electronic design automation.

In summary, this book provides an important bridge between the electronic design automation and microfluidic biochip research communities. This work is expected to reduce human effort and enable high-volume productions and applications of microfluidics-based biochips. The insights gained from this work are expected to pave the way for the integration of microfluidic components in the next generation of system-on-chip/system-in-package designs.

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Dedication

To Kamalika and Arunangshu
Krishnendu Chakrabarty

To my parents, sister, and my dear wife Min
Fei Sui

Part I

Synthesis Techniques

As more bioassays are executed concurrently on a digital microfluidics-based biochip, system integration and application complexity are expected to increase steadily. Thus system-level design automation tools (e.g., synthesis tools) are needed to handle increasing biochip design complexity. Synthesis research for digital microfluidic biochips can benefit from classical CAD techniques, which is a well-studied problem, and advances in synthesis techniques for integrated circuits continue even today.

As stated in Section 1.3.2, we envisage that the synthesis of a digital microfluidic biochip can be divided into two major phases, referred to as architectural-level synthesis (i.e., high-level synthesis) and geometry-level synthesis (i.e., physical design), respectively. A behavioral model (e.g., sequencing graph model) for a biochemical assay is first obtained from the protocol for that assay. Note that by using discrete unit-volume droplets, a microfluidic function can be reduced to a set of repeated basic operations (i.e., moving one unit of fluid over one unit of instance). This "digitization" method facilitates the implementation of many well-defined protocols for nano- and microscale bioassays on a microchip. A generic class of microdroplet-based bioassay protocols that can be applied to digital microfluidic biochips usually consists of the following steps:

1. Dispensing sample/reagent droplets into the microfluidic array
2. Transporting the droplets to some locations on the array for assays operations (e.g., mixing, dilution or optical detection)
3. Finally, moving the droplets of assay products or wastes out of the microfluidic array.

Based on the generated sequencing graph model, architectural-level synthesis is used to generate a macroscopic structure of the biochip; this structure is analogous to a structural register-transfer level (RTL) model in electronic CAD. This macroscopic model provides an assignment of assay functions to biochip resources, as well as a mapping of assay functions to time steps, based in part on the dependencies between them. Finally, geometry-level synthesis creates a physical representation at the geometrical level (i.e., the final layout of the biochip) consisting of the configuration of the microfluidic array, locations of reservoirs and dispensing ports, droplet routes, and other geometric details.

The goal of a synthesis procedure is to select a design that minimizes a certain cost function under resource constraints. For example, architectural-level synthesis for microfluidic biochips can be viewed as the problem of scheduling assay functions and binding them to a given number of resources so as to maximize parallelism, thereby decreasing response time. On the other hand, geometry-level synthesis addresses the placement of resources and the routing of droplets to satisfy objectives such as area or throughput. Defect/fault tolerance can also be included as a critical objective in the proposed synthesis method.

In architectural-level synthesis, both scheduling and resource-binding problems are addressed to generate a structural view of biochip design. As in the case of high-level synthesis for integrated circuits, resource binding in the biochip synthesis flow refers to the mapping from bioassay operations to available functional resources. Scheduling determines the start times and stop times of all assay operations, subject to the precedence constraints imposed by the sequencing graph. In Chapter 2, we present the proposed architectural synthesis methodology based on integer linear programming and heuristic techniques for scheduling assay operations under resource constraints. Resource-binding problem is also investigated in this chapter.

A key problem in the geometry-level synthesis of biochips is the placement of microfluidic modules such as different types of mixers and storage units. Based on the results obtained from architectural-level synthesis (i.e., a schedule of bioassay operation, a set of microfluidic modules, and the binding of bioassay operations to modules), placement determines the locations of each module on the microfluidic array in order to optimize some design metrics. Chapter 3 presents a simulated annealing-based methodology to solve the microfluidic module problem in a computationally efficient manner.

In Chapter 4, we further propose a synthesis methodology that unifies operation scheduling, resource binding, and module placement. This method allows architectural design and physical design decisions to be made simultaneously. Moreover, the proposed technique, which is based on parallel recombinative simulated annealing, can also be used after fabrication to bypass defective cells in the microfluidic array.

Chapter 5 investigates another key problem in biochip physical design (i.e., droplet routing between modules and between modules and on-chip reservoirs). It follows architectural-level synthesis and module placement in the proposed synthesis flow. We present the first systematic routing method for digital microfluidic biochips; the proposed approach attempts to minimize the number of cells used for droplet routing, while satisfying constraints imposed by throughput considerations and fluidic properties.

1 Introduction

Recent advances in microfluidics technology have generated tremendous interest in the design and implementation of miniaturized devices for biochemical analysis [1,2,3,4]. These composite microsystems, referred to interchangeably in the literature as microfluidic biochips, lab-on-a-chip, and bioMEMS, offer a number of advantages over conventional laboratory procedures. They automate highly repetitive laboratory tasks by replacing cumbersome equipment with miniaturized and integrated systems, and they enable the handling of small amounts (e.g., nanoliters) of fluids. Thus they are able to provide ultrasensitive detection at significantly lower costs per assay than traditional methods, and in a significantly smaller amount of laboratory space.

Microfluidic biochips promise to revolutionize enzymatic analysis (e.g., glucose and lactate assays), DNA analysis (e.g., PCR and nucleic acid sequence analysis), proteomic analysis involving proteins and peptides, immunoassays, and toxicity monitoring [5,6]. An emerging application area for microfluidic biochips is clinical diagnostics, especially immediate point-of-care diagnosis of diseases [5,6]. Microfluidics can also be used for countering bioterrorism threats [7,8]. Microfluidics-based devices, capable of continuous sampling and real-time testing of air/water samples for biochemical toxins and other dangerous pathogens, can serve as an always-on "bio-smoke alarm" for early warning.

The so-called *first generation* microfluidic biochips were based on continuous liquid flow through fabricated microchannels, and they were designed for simple biochemical analyses or assays [3,4]. Recently, a *second-generation* paradigm has emerged that manipulates liquids as discrete nanoliter droplets [9,10]. Following the analogy of digital electronics, this technology is referred to as *digital microfluidics.* In contrast to continuous-flow biochips, digital microfluidic biochips offer a scalable system architecture based on a two-dimensional microfluidic array of identical basic cells. Since each droplet (or groups of droplets) can be controlled independently, these "digital" systems also have dynamic reconfigurability, whereby groups of cells in a microfluidic array can be reconfigured to change their functionality during the concurrent execution of a set of bioassays. Due to their inherent properties of dynamic reconfigurability and architectural scalability, digital biochips can be used as programmable "microfluidic processors," especially for massively parallel DNA analysis, automated drug discovery, and real-time biomolecular detection.

As the use of digital microfluidic biochips increases, their complexity and integration scale are expected to become significant due to the need for multiple and concurrent assays on the chip, as well as more sophisticated control for resource management. Time-to-market and fault tolerance are also expected to emerge as design considerations. As a result, current full-custom design techniques will not