Dissertation

Fluidic Microsystems for Biochemical Analysis

ausgeführt zum Zwecke der Erlangung des
akademischen Grades eines Doktors der technischen
Wissenschaften von

Gabriel Hairer

eingereicht an der Technischen Universität Wien
Fakultät für Elektrotechnik und Informationstechnik

2009

The research presented in this thesis was carried out at the Industrial Sensor Systems group of the Institute of Sensor and Actuator Systems, Vienna University of Technology, Austria.

Erstbegutachter	Univ.-Prof. Dr. Michiel J. Vellekoop Vienna University of Technology, Austria
Zweitbegutachter	Assoc.-Prof. Dr. André Bossche Delft University of Technology, The Netherlands

Schriftenreihe des Instituts für Sensor- und Aktuatorsysteme
Technische Universität Wien

Gabriel Hairer

Fluidic Microsystems for Biochemical Analysis

Shaker Verlag
Aachen 2009

Bibliographic information published by the Deutsche Nationalbibliothek
The Deutsche Nationalbibliothek lists this publication in the Deutsche Nationalbibliografie; detailed bibliographic data are available in the Internet at http://dnb.d-nb.de.

Zugl.: Wien, TU, Diss., 2009

Printed in Germany.

ISBN 978-3-8322-8736-8
ISSN 1866-1181

Shaker Verlag GmbH • P.O. BOX 101818 • D-52018 Aachen
Phone: 0049/2407/9596-0 • Telefax: 0049/2407/9596-9
Internet: www.shaker.de • e-mail: info@shaker.de

Contents

Abstract

The concept of miniaturised total analysis systems (μTAS) came up in the early 1990s. This approach implicates the integration of a (bio)chemical sensor into a microsystem combined with on-chip reagent processes and microfluidics. Such miniaturised analysis systems allow to carry out measurements from small volumes of complex fluids with high efficiency and speed.

In this thesis, a modular design of a self-contained, cost-efficient and miniaturised system for fast DNA analysis is presented. The system consists of an on-chip DNA amplification unit based on the PCR method and of a capacitive microsensor for label-free DNA detection. The DNA detection principle relies on a capacitance change at the surface of interdigitated submicron electrode pairs during the DNA hybridisation process caused by the accumulation of complimentary DNA strands. Fast on-chip DNA amplifications and the performance of the label-free sensing principle have been successfully demonstrated. Both devices are fabricated as separate chips, which are finally combined with a fluidic system allowing the reagent to be transported from one chip to the other. A portable control-measurement unit has been realised, which allows thermocycling at predefined temperature steps for the PCR and to measure capacitance changes on the surface of several electrode pairs in parallel. The combination of the PCR chip, the DNA sensor, the fluidic system and the control-measurement unit composes a cost-efficient and miniaturised analysis system, which is suitable for the amplification and detection of specific DNA sequences in a point-of-care format for biochemical analysis.

Further this thesis deals with the issue of microfluidics, which is a main research field of μTAS. Here, the flow behaviour of a sample stream inside a microchannel is studied, which is focused by vertical and horizontal neighbouring sheath flows. This so-called hydrodynamic focusing technique offers the possibility to adapt the dimensions of the sample stream and its position inside the microchannel by varying the flow rate ratio between the sample liquid and the individual adaptable sheath liquids. Two microfabricated flow-cells have been realised and characterised; one for non-coaxial sheath flow (sample stream is surrounded at three sides, while at the fourth side the sample moves along the channel bottom) and one for coaxial sheath flow (sample stream is squeezed on all four sides). Experiments have been carried out with both devices, where the size, shape and position of the sample stream have been established by laser scanning confocal microscopy and dye intensity analysis. In microchannels (typically 25×40 µm^2) the creation of sample stream diameters that were five to ten times smaller than the channel dimensions were achieved, and the smallest measured sample stream width was 1.5 µm. The mechanism to control the sample stream inside the microchannel allows the sample dimensions to be adapted over a wide range. A comparison of the experimental results with fluidic simulations show excellent agreements. In combination with (integrated) sensors, these microfluidic devices are suitable for on-chip single cells analysis.

Nomenclature

0.1 List of Abbreviations

Symbol	Description
16S rDNA	DNA coding (16S subunit of prokaryotic ribosomes)
2D	two-dimensional
3D	three-dimensional
A	adenine
AC	alternating current
ADC	analogue-digital-converter
Ag	silver
bp	basepair
BSA	bovine serum albumin
C	cytosine
CDC	capacitance-to-digital converter
CFD	computational fluid dynamic
C_3H_8O	isopropyl alcohol
$C_6H_{12}O$	methylisobutylketone
Cy3	cyanine 3
DC	direct current
DEP	dielectrophoretic
DI	de-ionised
DNA	deoxyribonucleic acid
dNTPs	deoxyribonucleotide triphosphates
DRIE	deep reactive ion etching

Symbol	Description
dsDNA	double-stranded DNA
DVD	digital versatile disc
FACS	fluorescence activated cell sorter
FEM	finite element method
G	guanine
GPIB	general purpose interface bus
H_2O_2	hydrogen peroxide
H_2SO_4	sulfuric acid
IC	integrated circuit
I^2C	inter-integrated circuit
kb	kilo base
KCl	potassium chloride
KOH	potassium hydroxide
LC	liquid crystal
LED	light-emitting diode
LPCVD	low pressure chemical vapour deposition
M	molar
MEMS	micro-electro-mechanical system
$MgCl_2$	magnesium chloride
$Na_2S_2O_8$	sodium peroxodisulfate
PBS	phosphate buffered saline
PCB	printed circuit board
PCR	polymerase chain reaction
PDE	partial differential equations
PDMS	polydimethylsiloxane
PEB	post exposure bake
PGMEA	1-methoxy-2-propyl-acetat
PMMA	polymethylmethacrylate

Symbol	Description
POM	polyoxymethylene
Pt	platinum
PWM	pulse-width modulation
RIE	reactive ion etching
RTD	resistance temperature detector
rpm	rounds per minutes
rRNA	ribosomal ribonucleic acid
Si_3N_4	silicon nitride
SiO_2	silicon dioxide
SLR	single-lens reflex
ssDNA	single-stranded DNA
T	thymine
Taq	thermus aquaticus
US	ultrasonic
USB	universal serial bus
UV	ultraviolet
w/v %	weight/volume percentage
µC	microcontroller
µTAS	miniaturised total analysis system

0.2 List of Constants

Constant	Description	Unit
α	temperature coefficient	$3.85 \times 10^{-3}\,\mathrm{K}^{-1}$
π		3.14159
ε_0	permittivity of vacuum	$8.8542 \times 10^{-12}\,\mathrm{As/Vm}$

0.3 List of Variables

Symbol	Description	Unit
A	area	m^2
b	width	m
B	analogous to a capacitance	F
C	capacitance	F
D	diffusion coefficient	$m^2\,s^{-1}$
f	frequency	Hz
h	height	m
I	DC current	A
k	variable	–
l	length	m
L	characteristic length	m
n	number	–
P	perimeter	m
Pe	Péclet number	–
R	electrical resistance	Ω
Re	Reynolds number	–
T	temperature	$^\circ C$
ΔT	temperature difference	K
U	DC voltage	V
U_{pp}	peak-to-peak voltage	V
v	average velocity	$m\,s^{-1}$
Z	impedance	Ω
ε_r	relative dielectric constant	–
η	dynamic viscosity	$Pa \cdot s$
ω	angular frequency	$rad\,s^{-1}$

Symbol	Description	Unit
ρ	mass density	$\mathrm{kg\,m^{-1}}$