Integration of Chip-based Sample Preconcentration into the µChemLab™/CB Analysis Platform

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Sandia is a multiprogram laboratory operated by Sandia Corporation, a Lockheed Martin Company, for the United States Department of Energy under contract DE-AC04-94AL85000.
μChemLab™ Separation Platform

- Aerosol Collector
- Sample Prep
- μChemLab™/CB Liquid Analysis Module
- Water Analysis
- Microfluidic Delivery
  - Application of electric fields moves fluids
- Dual Channel Microchip Separations
- Laser Induced Fluorescence Detection
μChemLab™ Separation Platform

Two Channel Device

Capillary Gel Electrophoresis (CGE) Channel
- Beckman 14-200 SDS Gel
- Reversed polarity

Capillary Zone Electrophoresis (CZE) Channel
- 10 mM phytic acid, pH 9.5 containing 2 mM DAPS (zwitterionic detergent)
- Normal polarity

Fluorescamine labeling
- Fluorogenic dye
- Fast-reacting, amine specific
- Ex/Em 390 nm/480nm

Detection limits
- nM for CGE
Approaches to Preconcentration

• **Solid Phase Extraction** (on-going efforts for the miniaturization of SPE)

• **Electrokinetic Trapping** *(P411-T: Novel Miniaturized Protein Preconcentrator Based On Electrokinetic Trapping, Anup K. Singh; Daniel J. Throckmorton; Brian J. Kirby)*

• **Salt-Bridge**

Integration into the µChemLab™/CB

Silicate layer Approach:

- Sol-gel silicate layer allows current to pass
- Large molecules (i.e. proteins) retained
- Etched channels

Alternative Approach:

- Eliminate silicate layer
- Makes use of narrow gap and natural surface roughness
- Conduction via very small bore channels
- Large molecules (i.e. proteins) retained
- Etched channels

Chip Layout

Close-up of the Preconcentrator Region
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**Detection limits**
- nM for CGE
Imaging Normal and Preconcentrated Injections in CGE

Normal Mode

P1 Preconcentration Mode

P2 Preconcentration Mode

S → SW

S → P1

S → P2
CGE Separations Using Normal and Preconcentrated Injections

20 nM Lactalbumin, 20 nM Ovalbumin

60 s preconcentration
No preconcentration
Peak Areas in CGE Separations Increase as a Function of Preconcentration Duration

50 nM Lactalbumin, 50 nM Ovalbumin

120 s preconcentration

60 s preconcentration

No preconcentration

Fluorescence

Time (s)
Sub-Nanomolar Detection Limits Using CGE Separations and Preconcentration

500 pM LAC
500 pM OVA

Fluorescence
Time (s)
Characterization of Performance as a Function of Gap Width

Gap width
- Isotropic etch depth used to generate gaps of different widths
- For comparison purposes, all preconcentrations were held to 60s

*Single Batch Data

*Single Batch Data
μChemLab™ Separation Platform

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Preconcentration Using CZE

Elimination of EOF is required

• no bulk flow of fluid through the gap
  \[\therefore\] residual EOF could lead to pressure generation

Coating with linear polyacrylamide

SAM application
UV light polymerization

*Julie Fruetel, Victoria VanderNoot, Jay West, Brian Kirby, Ernest Hasselbrink and Timothy Shepodd, Laser-polymerized thin-film coating for protein analysis by CGE in a microchip, HPCE 2002*
Imaging Normal and Preconcentrated Injections in CZE

P1

SW

S

P2

Normal Mode
S → SW

P1 Preconcentration Mode
S → P1
CZE Separations Using Normal and Preconcentrated Injections

Running buffer: 5 mM phosphate, pH 8 containing 5 mM CAS-U zwitterionic detergent
Reverse polarity
Summary

• Preconcentration has been incorporated in the chip design with no additional processing steps during chip fabrication

• Successfully demonstrated with both CGE and CZE
  ▪ Preconcentration factors of 10-20x are routinely achievable in only 60s
  ▪ Coating and/or elimination of EOF is essential for CZE separations
  ▪ Buffer conditions in CZE will need to be optimized to achieve both good separation efficiency and the elimination of EOF
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