Chitosan — a Biomaterial Interface that can be Selectively Deposited onto Micropatterned Surfaces and Conjugated to Sensing Biomolecules

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## Motivation

**INTEGRATING MICROFABRICATION WITH BIOLOGY**

**Biosensors:** Disease Diagnosis, Contaminant Detection & Drug Discovery

**Microfabrication:** Surface Patterning for Spatial Selectivity

**Biology:** Molecule Recognition for Biosensing

**Challenges:** Coupling Labile Biomolecules to Inorganic Surface

**Solutions:** Exploit Polysaccharide Chitosan as Interface Material
Chitosan: An Interface Material

- Electrochemical Deposition
  - pH-Dependent Electrostatic Behavior
  - pH-Dependent Solubility

\[
\text{Soluble} \quad \leftrightarrow \quad \text{Insoluble} \quad + \quad 2n \ H^+ 
\]
Electrochemical Deposition of Chitosan

--- Mechanism

1. Proton Consumption Creates Localized pH Gradient

2. pH-Dependent Solubility Allows Chitosan to Deposit on Cathode Surface
Chitosan Deposition is Voltage Programmable & Spatially-Selective

- Patterned Gold Surfaces
- Fluorescently-Labeled Chitosan
- Applied Voltage

Fluorescently-Labeled Chitosan

125 µm
Deposition of Fluorescently-labeled Chitosan

Microscope

Before Dep.  After Dep.

Optical

Fluorescence

Magnification 20×  Magnification 8×

Chitosan: An Interface Material

- Electrochemical Deposition
  - Voltage- Programmability
  - Spatial Selectivity

- Chemical Modification
  - Reactive with Standard Chemistries
  - High Resolution / Repeatability
Repeatability of Chitosan Deposition & Reaction

Chitosan Deposition

Amine-Reactive Fluorescent Tag

Chitosan: An Interface Material

- **Electrochemical Deposition**
  - Voltage- Programmability
  - Spatial Selectivity

- **Chemical Modification**
  - Reactive with Standard Chemistries
  - High Resolution / Repeatability

- **Bio-functionality**
  - Nucleic Acid (DNA) Coupling
  - Protein Coupling
Surface Activation by Standard Chemistry

Coupling Agent: Glutaraldehyde

Activated Surface

Chitosan

Gold

ssDNA

Protein
Activated Surface

Immobilization & Hybridization of Nucleic Acid (DNA)

The “Chip”
Sensitive, Selective & Repeatable DNA-DNA Hybridization

Target 0.25nmol

Chitosan: An Interface Material

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- **Bio-functionality**
  - Nucleic Acid (DNA) Coupling
    - Molecular Recognition
    - Spatial Resolution
  - Protein Coupling
3-D Structure of Green Fluorescent Protein (GFP)
Immobilization of GFP Protein onto Spatially Templated Chitosan

Results:
Immobilization of GFP Protein in Microfluidic Channel

Dimensions:

Microfluidic Channel: \(3.2\text{mm} \times 50\mu\text{m} \times 100\mu\text{m}\)

Reservoir: \(\phi = 0.8\text{mm}\)
Covalent Immobilization of GFP onto Micro-fluidic Channels

Deposit Chitosan → Activate Chitosan

Couple GFP

Chitosan at the Interface

- **Electrochemical Deposition**
  - Voltage- Programmability
  - Spatial Selectivity

- **Chemical Modification**
  - Reactive with Standard Chemistries
  - High Resolution / Repeatability

- **Bio-functionality**
  - Nucleic Acid (DNA) Coupling
    - Molecular Recognition
    - Spatial Resolution
  - Protein Coupling
    - Retain Protein Structure
    - Microfluidic Environment
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