Nanoreactor amplification scheme for electrochemical, enzymatic biosensors

A collaboration between:

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BioCure (Schweiz) GmbH

- BioCure (Schweiz) GmbH is the Swiss subsidiary from BioCure Inc., Norcross, GA USA
- BioCure Inc. is a Spin-off from Ciba Vision / Novartis
  - Exclusive biomaterials license excludes “eye”
- 1st rd funding 1999 $9 mio; 2nd rd 2006 $1.5 mio
- Current headcount: 14 employees
BioCure’s Core Strengths

- Polymer chemistry
- Product development
- Pre-clinical testing
- Regulatory affairs / quality systems
- Sterile manufacturing since 2002
- Products/Projects: *In situ* forming implants, hydrogels beads
- Technologies: Hydrogels, coatings, polymeric vesicles, nanoreactors
Nanoreactors for Immunoassay

Ratio AG/Enzyme: 1:1 – 1:5

1:300 (– 10’000?)
Electrochemical Biosensors

Benefits:
1. Signal-Amplification
2. Low non-specific binding
   → Much improved S/N ratio
   → Higher sensitivity, reduced time to measure, simplified device
Nanoreactor - Formation

Size: 80 nm – 1 μm
QCM-D - Adsorption Polymeric Vesicles

Quartz crystal microbalance with dissipation monitoring (QCM-D)

Blue: 450 nm diameter vesicles
Red: 1000 nm diameter vesicles

Neutravidin (NA), polymeric vesicles
EC Signal from Enzymes in Polymeric Vesicles
Chronoamperometry with Ferrocyanide

With increasing concentration of vesicles also an increased signal
Biosensor

Vesicle

NA with biotin

2nd antibody with biotin

Antigen

BSA (brown)
1st antibody (green)
Polymeric Vesicles in Biosensor - QCM-D -

See also: D. Grieshaber et al., Sensors 2008, 8, 7894-7903
Polymeric Vesicles in Biosensor
Chronoamperometry with Ferroicyanide

The sandwich set-up shows activity
## Current Detection Limits

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<th>QCM-D</th>
<th>Chronoamperometry with Ferrocyanide</th>
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<tbody>
<tr>
<td>Model assay with NA</td>
<td>0.2 pM</td>
<td>1-2 pM</td>
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<tr>
<td>Sandwich assay</td>
<td>Feasible (pM)</td>
<td>Feasible (pM)</td>
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Conclusion

• New amplification scheme for Immunoassays
  – Signal amplification, low non-specific adsorption
    → Improved S/N
• Simple EC detection scheme
  → Suited for hand-held diagnostic system
• Control in QCM-D
• Polymeric vesicles - the stable liposomes...
Next steps

- Correlation of the number of encapsulated enzymes to the signal per antigen
- Optimize amplification
- Optimize kinetics
- Quantify non-specific binding – Antibody optimization
- Fluorescent Applications (i.e. Microarrays)
Acknowledgements

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Questions?