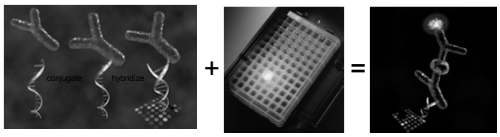
Oligonucleotide tethering of proteins for multiplex assay migration to mobile applications

Robert S. Matson, Ph.D., FACB
President

QuantiScientifics LLC, Irvine, CA

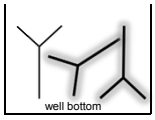
Our Approach

A^2



Creation of multiplex immunoassays by the self assembly of oligo-antibody conjugates

Protein Immobilization-a stochastic process



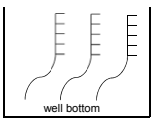
e.g. ELISA plate

The Physical-Chemical Nature of Proteins

- different molecular sizes
- isoelectric points
- relative hydrophobicity
- tertiary structure
- surface denaturation
- printer related issues: adsorption; carryover
- differences in protein to protein spot densities
- variation in binding or capture efficiency

Printing proteins-leads to variation

Oligonucleotide Tethering- well defined

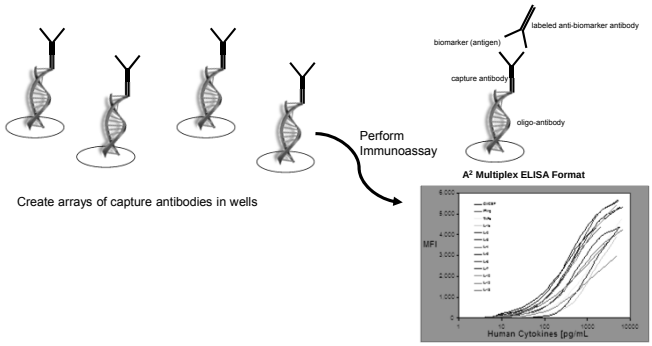


e.g. Oligonucleotide Array

The Thermodynamics of DNA

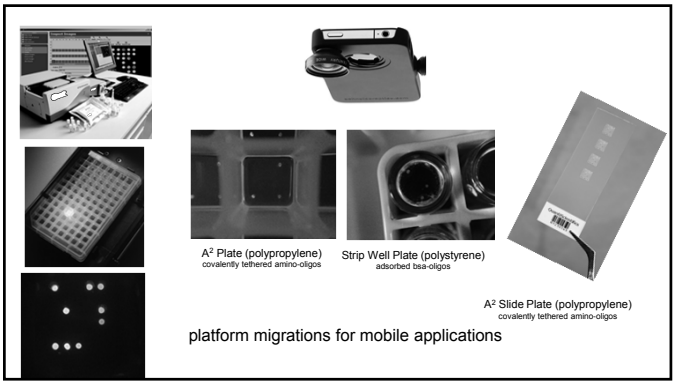
- similar molecular size
- no significant charge or hydrophobicity effects
- 3' or 5' tethering provides orientation
- thermodynamic-based sequence design
- stable
- hybridization controlled spot density

Printing DNA- robust and reproducible



Create arrays of capture antibodies in wells

Multiplex ELISA



A² Plate (polypropylene)
covalently tethered amino-oligos

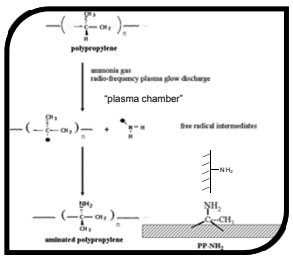
Strip Well Plate (polystyrene)
adsorbed bsa-oligos

A² Slide Plate (polypropylene)
covalently tethered amino-oligos

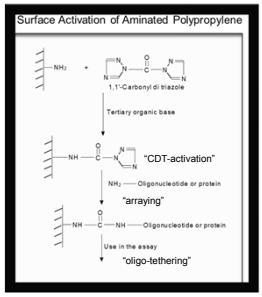
platform migrations for mobile applications

Modification of Thermoplastics for Immunoassay Development

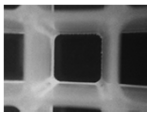
Cold Gas Plasma Surface Modification of Polypropylene



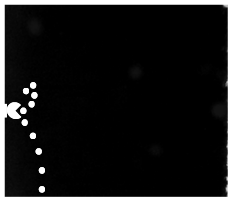
Amine Substrate Stability ≥ 7 years, real-time shelf life

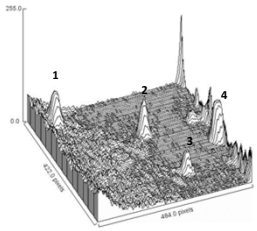


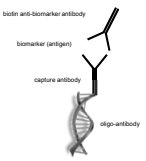
CDT Activated Substrate Stability ≥ 5 years, 2°-8°C
Oligo Tethered Substrates ≥ 5 years, 2°-8°C
≥ 1 year, room temp.



iPhone 4G Image of A² Plate Wells under Long Wavelength UV



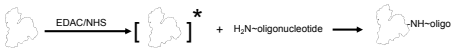




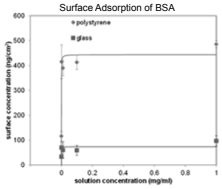
Sensitivity, pg/mL range

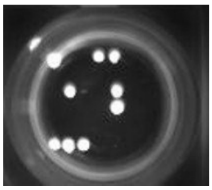
A² Multiplex ELISA Format

Migration to Strip Wells



Surface Adsorption of BSA

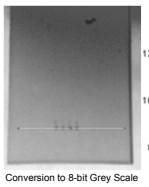




BSA-Capture Oligonucleotide Arrayed on Polystyrene Strip Well

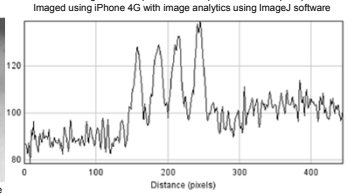
Migration to Plastic Slides

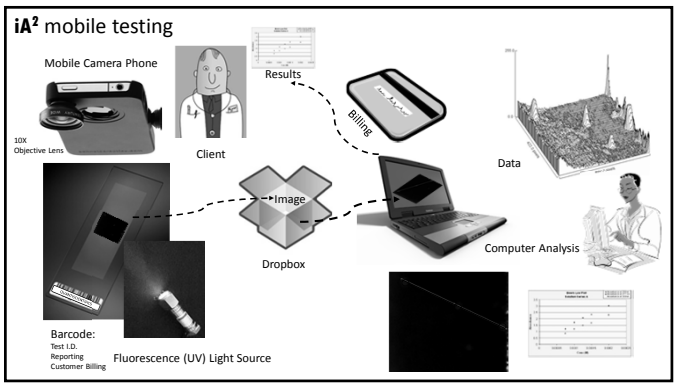
iPhone 4G Image of PP Slide



Conversion to 8-bit Grey Scale

Fluorescent Signal Development with streptavidin alkaline phosphatase
Imaged using iPhone 4G with image analytics using ImageJ software




iA² mobile testing
Mobile Camera Phone
10X
Objective Lens
Client
Results
Billing
Data
Image
Dropbox
Computer Analysis
Barcode:
Test I.D.
Reporting
Customer Billing
Fluorescence (UV) Light Source