Ultra-low-light CMOS biosensor helps tackle infectious diseases

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Enable Point-of-care Molecular Diagnostics

Rapid, precise diagnosis of infectious pathogens, so doctors can respond quickly with life-saving drugs and treatment
Today: Molecular Diagnostics (MDx) accurate, but not timely and accessible

- Sample Collection
- Transportation & Preservation
- Batch Testing in Centralized Lab

Time to result: days to weeks
Point-of-care (POC) + MDx = Fast and actionable diagnostics

On-site detection w/portable instrument,
Time to result: 30 min ~ 1.5 hours

Actionable diagnostics means:
- Distinguish bacterial vs. viral infections (flu, hepatitis)
- Detect drug-resistant mutations
- Quantitative when needed
- Fit into “symptom to treatment” timing window
Compact instrumentation is key…

- Current generation of MDx instruments are too bulky, expensive
  - Microfluidics alone fell short of enabling miniaturization
  - Compact instrumentation with integrated and compact read out electronics is important -> need better biosensors

Microfluidics allow diagnostic reactions in compact chip format

The instrument needs to be small too!

(Beckman Coulter’s Ampligrid System)
Landscape of molecular* bio-sensors

* Molecular means nucleic acid (DNA, RNA) or protein molecules.
Comparison of molecular sensor technologies

1. Low-light optical sensors for fluorescence or chemiluminescence-based detection
   - PMT
   - CCD, cooled-CCD
   - Avalanche diodes
   - CMOS

2. Electrochemical sensors, integrated with assay
   - “Surface chemistry” complicated and unstable.
   - Can be very sensitive, but specificity is poor.

3. Optical sensors integrated with assay
   - Better light collection efficiency, but increase per-use cost

Why low light sensitivity?
1. Molecular probes emits very low level of light
2. Signal of interest is very narrow band

Most successful electrochemical sensor is the Glucose sensor.
Introducing Anitoa ULS24 Ultra-low-light CMOS bio-imager

- **Ultra low-light sensitivity**
  - Detection threshold $\sim 3.0 \times 10^{-6}$ lux*
  - Low dark current, high SnR (>13dB at detection threshold)
  - Wide dynamic range (> 85dB)
  - 12-bit ADC Digital interface through Serial Peripheral Interface (SPI)
  - Built-in temperature sensor
  - 3.3V and 1.8V power supply, **30mW max** power
  - 150um pixels in 24 x 24 format

* @ 550nm, 20nm bandwidth, 3s integration time

PMT or Cooled-CCD level sensitivity in CMOS

Anitoa ULS24 CMOS Ultra-low-light bio-imager
Intelligent Dark Current Management

Intelligent Dark-current Management: Starts with high responsivity/low dark current photo-diodes. The readout circuit performs multimodal sensing to capture signal and noise information, the ADC and DSP takes advantage of the multi-modal information to achieve better noise cancellation.

(process improvement: Target raw sensitivity and dark current)
(analog circuitry innovation: Target elimination of reset noise and signal conditioning)
(novel DSP algorithm: Target reduction of readout noise and fixed pattern noise)

Intelligent Dark-current Management: Starts with high responsivity/low dark current photo-diodes. The readout circuit performs multimodal sensing to capture signal and noise information, the ADC and DSP takes advantage of the multi-modal information to achieve better noise cancellation.
Integration of Anitoa ULS24 in an embedded system

- ULS24 can directly interface with the host processor through an SPI interface
  - ULS24 just need a 12MHz clock and 3.3/1.8v supply
  - Easily support multi-channel configuration
- Alternatively, ULS24 can go through a dedicated uC to interface with the host system.
  - The dedicated uC provides timing control
Anitoa ULS24 application performance data: dsDNA quantification

DNA quantification test results: 500x more sensitive than absorption-based (A260) techniques

DNA quantification with Qubit® quantitation reagent (Life Technologies). Excitation light source: 470nm LED Filters: Chroma OD6 band pass filters

SnR vs signal strength vs integration time
Combining CMOS biosensor with Microfluidics

- CMOS biosensor and Microfluidics innovations enable **compact** molecular diagnostic **instrumentation**

- **Miniaturization** of **optoelectronic** sub-systems is the key
  - Ultra-low light CMOS biosensor complements Microfluidics
Putting it together - Anitoa’s portable Nucleic-Acid-Test (NAT) platform targeting infectious disease

Features and benefits

- Low-cost*, miniaturized design
- Reliable. Great reproducibility
- Low power, no moving parts, can be battery backed.
- High sensitivity, high level of integration
- Single chip* fluorescent and bioluminescent imaging

(* low instrument cost and low consumable cost important, this means no active component in consumable)
Putting it together(2) – Integrated optoelectronics, thermal and fluidic system

Achieving portable MDx solution
- Multichannel fluorescent imaging
- Multi-channel LED based excitation source
- Miniature thermal cycler
- Support standard qPCR tube or microfluidic chip with flexible well format
- No internal moving parts*

* Unless motion control needed for fluidic pumps or valves (not shown)
Real time quantitative PCR with Anitoa ULS24 CMOS biosensor

qPCR test results: Detection and quantification of E. coli and HBV (incl. wild-type and drug-resistant variations: rtM204I, rtL180M) w/ 4 copies/reaction sensitivity, 10^9 dynamic range.
HBV drug-resistant mutations diagnostics with Anitoa ULS24 CMOS biosensor

- Important advantage of MDx is detection of drug-resistance mutations
  - ...and predict drug reaction
  - Avoid further development of drug-resistance
  - Fluorescent wavelength multiplex instrument offers advantage

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<th>HBV mutations</th>
<th>Lamivudine</th>
<th>Adefovir</th>
<th>Clevudine</th>
<th>Sebivo</th>
<th>Entecavir</th>
<th>Tenofovir</th>
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S: Sensitive    R: resistant I: Intermediate

(* There is also the Interferon method, which show broad sensitivity, but has more side effects, need injection.)

No more “shot-gun” approach in drug prescription.
POC MDx benefits to target applications

- Short symptom to treatment window (Actionable results on site)
- ICU urgent need (Avoid life threatening complications)
- Short Viral Sample Life (Use sample right away to Avoid false negative)

- Influenza A,B, H1N1 Swine Flu
- MRSA
- Hepatitis
## Business case of POC MDx powered by Anitoa CMOS biosensor

<table>
<thead>
<tr>
<th></th>
<th>CMOS biosensor enabled POC MDx</th>
<th>1st gen POC (e.g. Cepheid GeneXpert, Biofire etc.)</th>
<th>Traditional MDx (Life Technology, Roche, Qiagen)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Deployment</td>
<td>Point of Care</td>
<td>Point of Care</td>
<td>Reference Lab</td>
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<tr>
<td>Size</td>
<td>Handheld</td>
<td>Bench top</td>
<td>Central Lab Equip.</td>
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<td>Equip Cost</td>
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<td>Sample to result</td>
<td>30min - 1.5hr</td>
<td>1-2 Hours</td>
<td>days to weeks</td>
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Costs important for adoption

- Per-test cost has to be competitive with centralized lab model
  - Labs reduce cost/test by high throughput batch operation
- Hospitals and doctors want a share of revenue from consumable use
  - Especially in developing countries.
- Instrument BOM needs to be low
  - Instrument supplier can achieve profitability without significant dependence on consumable profit
- Bio-informatics applications and services potential revenue source
Other identified applications of ultra-low light CMOS biosensor

1. Fluorescence Images Guided Surgery (FIGS)
2. Fluorescence or chemiluminescence-based Immunoassay/ELISA
3. Food safety, environment safety or bio-threat detection.
4. DNA or Protein microarray
5. Pyro-sequencing
6. Capillary electrophoresis
7. Cell sorting/Imaging flow cytometry /Circulating tumor cell detection
Development timeline and status

**Dec. 2013**: ULS24 MPW taped out

**Feb. 2014**: Validated with dsDNA quantification using Qubit® assay.

**June, 2014**: Validated with qPCR using SYBR Green Chemistry for E Coli detection.


**May, 2015**: Validated with qPCR with Hepatitis B, C including drug resistance strands, with 4 copies sensitivity, using Taqman® chemistry.

**Up to now**: 3rd party evaluation started in areas (FIGs, ELIZA, Cell sorting etc. Quantum dots measurement).

Summary and future plan

**Summary**
- Ultra-low-light CMOS biosensor enables compact and low-cost instrumentation for point-of-care molecular diagnostics.

**Future plan**
1. Further miniaturization of opto system
   - Smaller camera system for mobile integration
   - Direct coating and patterning of thin film filters on chip to achieve truly single chip multi-channel fluorescent imaging.
2. Create high speed variation of the chip
   - Targeting cell sorting and cancer screening applications
3. Further refinement of integrated opto-thermal-fluidic system platform
   - For handheld sample-to-answer MDx system
THANK YOU!

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