About OMICS Group

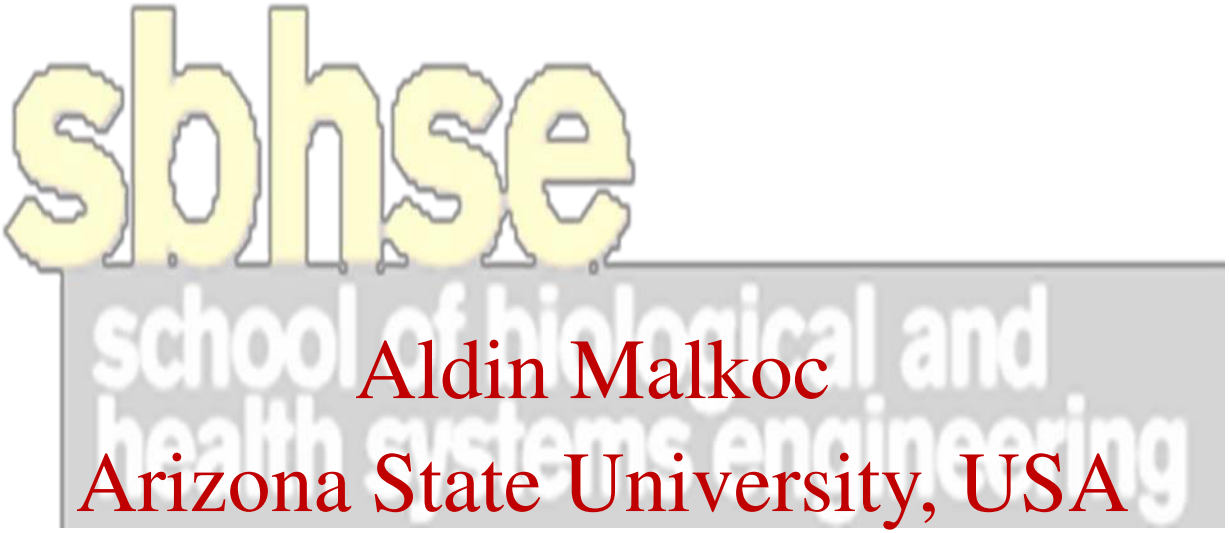
OMICS Group International is an amalgamation of Open Access publications and worldwide international science conferences and events. Established in the year 2007 with the sole aim of making the information on Sciences and technology 'Open Access', OMICS Group publishes 400 online open access scholarly journals in all aspects of Science, Engineering, Management and Technology journals. OMICS Group has been instrumental in taking the knowledge on Science & technology to the doorsteps of ordinary men and women. Research Scholars, Students, Libraries, Educational Institutions, Research centers and the industry are main stakeholders that benefitted greatly from this knowledge dissemination. OMICS Group also organizes 300 International conferences annually across the globe, where knowledge transfer takes place through debates, round table discussions, poster presentations, workshops, symposia and exhibitions.

About OMICS Group Conferences

OMICS Group International is a pioneer and leading science event organizer, which publishes around 400 open access journals and conducts over 300 Medical, Clinical, Engineering, Life Sciences, Pharma scientific conferences all over the globe annually with the support of more than 1000 scientific associations and 30,000 editorial board members and 3.5 million followers to its credit.

OMICS Group has organized 500 conferences, workshops and national symposiums across the major cities including San Francisco, Las Vegas, San Antonio, Omaha, Orlando, Raleigh, Santa Clara, Chicago, Philadelphia, Baltimore, United Kingdom, Valencia, Dubai, Beijing, Hyderabad, Bengaluru and Mumbai.

The development of an oligonucleotide, label-free electrochemical impedance based point-of-care technology

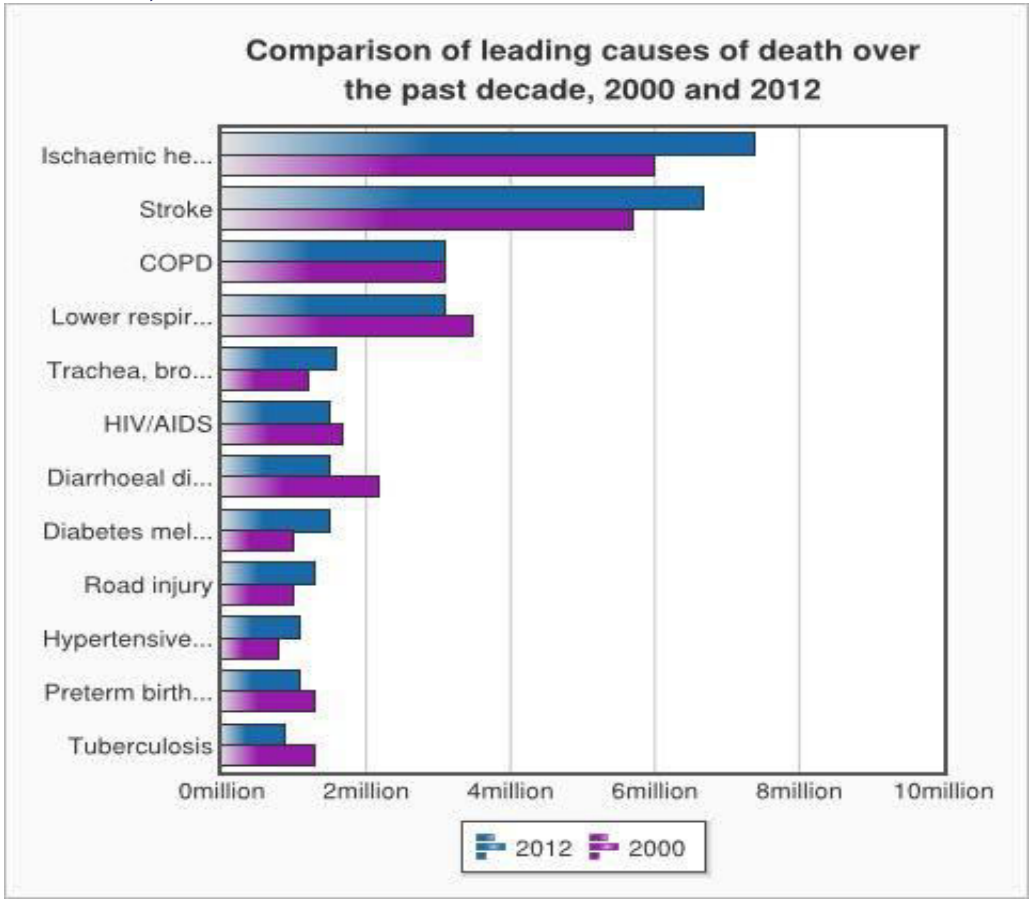


Aldin Malkoc
Arizona State University, USA

Jeffrey T. La Belle, Ph. D
Michael Caplan, Ph. D

Background: Bacterial Infectious Disease (BID) Statistics

- BIDs are responsible for at least 4.6 million deaths in the world
- Annual Costs
  - 100 billion dollars annual worldwide



"WHO (2008) The Burden of Tuberculosis: Economic Burden. Geneva, Switzerland: World Health Organization."
"The top 10 causes of death. (n.d.). WHO. Retrieved July 13, 2014,"
"Gallup JL, Sachs JD (2000) The Economic Burden of Malaria. Cambridge, MA, USA: Center for International Development at Harvard University"

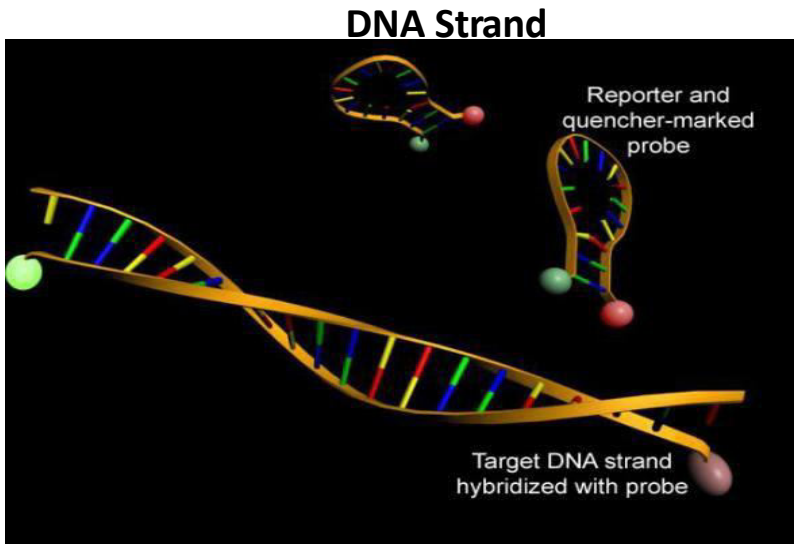
Current SOTA (State of the Art) BID Sensors

SOTA Sensor	Pros	Cons	Price
Plate Culture	Simplest setup for bacterial detection	Takes 3-5 days with low sensitivity.	>10$
GRAM Stain	Identifies gram negative or positive microbes	Insensitivity of Gram smear	>20$
PCR-gel Electrophoresis	Multiplication of DNA rather quickly, simple set-up	Long time, False Positives 9-57%	>50$
Fermentation Test	Will produce high accuracy result	Takes few days to receive result	>100$
Molecular Beacon	High signal to noise ratio and high specificity	Post analysis due to mutations	>500$

http://www.biomerieux-diagnostics.com/servlet/srt/bio/clinical-diagnostics/dynPage?open=CNL_CLN_PRD&doc=CNL_PRD_CPL_G_PRD_CLN_11&pubparams.sform=4&lang=en
http://www.vintessential.com.au/resources/articles/malolactic-fermentation-monitoring.html

What to Measure for in BIDs

- Why Nucleotide
  - Earlier/Accurate detection
- Goal
  - EIS and Molecular Beacon
- Ideal Method
  - Low cost
  - High specificity and sensitivity
  - Quick response time
-

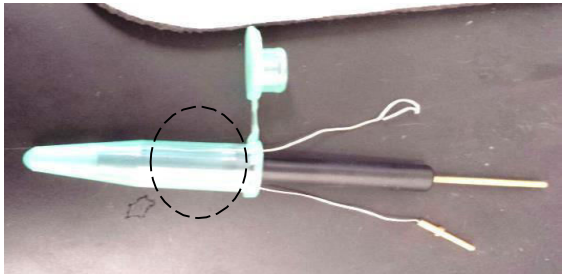




Experimental Setup

- Using TP combined with EIS immobilization chemistry for detection
  - Nucleotide-nucleotide interactions
- EIS
  - Three electrode set-up
- Hot Plate
  - Optimal binding temperature

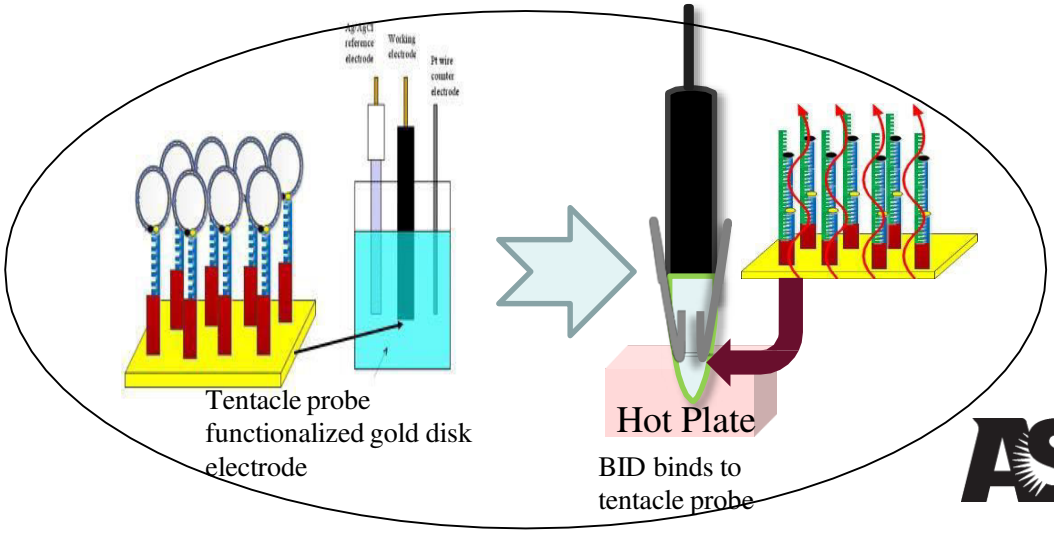




Target Sequence

WT: ATTATTACTTTACTATATTAGCTTTTCCGCCATCTAAAATTCTATT

SNP: ATTATTACTTTACTATATTATCTTTTCCGCCATCTAAAATTCTATT

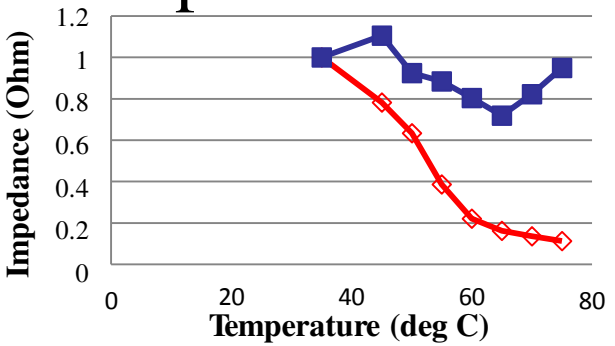


Data Collected

- Lower Limit Detection

Electrochemical Technique	Pros	Cons	LLD (M)
Electrochemical Impedance Spectroscopy (EIS) [5]	Detection range is 10^{-9}-10^{-15} M. The technique takes 90 seconds and does not require a label.	Requires immobilization of protein recognition elements.	$10^{-9} – 10^{-15}$
EIS with Tentacle Probe	ANOVA shows a P-value = 1.93E-06 WT and SNP are have significantly different impedance	Requires immobilization of protein recognition elements.	$20*10^{-9}$

- Replication of Data



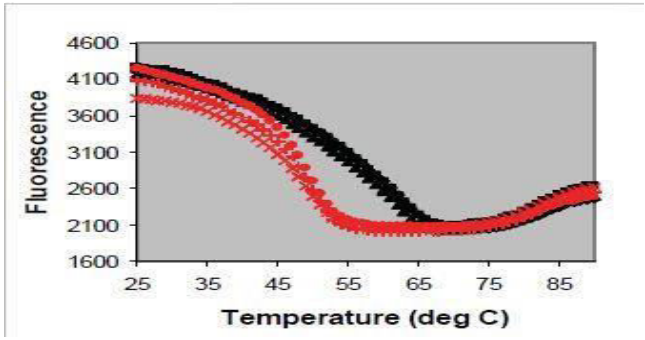
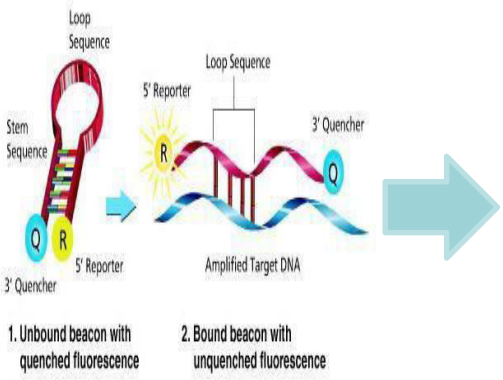


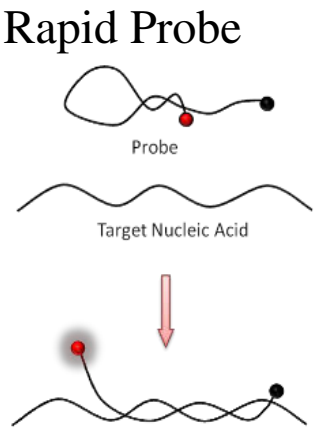
Figure at Left. A Temperature gradient of using a molecular beacon. Immobilized to Gold surface.

Satterfield, B. C., M. R. Caplan, and J. A. A. West. "Tentacle probe sandwich assay in porous polymer monolith improves specificity, sensitivity and kinetics."*Nucleic Acids Research* 36.19 (2008): e129-e129. Print.
[5] Bhavasar, 2009

Future Work & Conclusion

- Rapid probe (RP)
  - Replace TP with molecular beacon that is truly cost efficient.
- Human blood testing
  - Complex Solution
- Multiplexing
  - Detection for multiple BID from one sample
- Real time testing
  - Point-of-care EIS/RP device







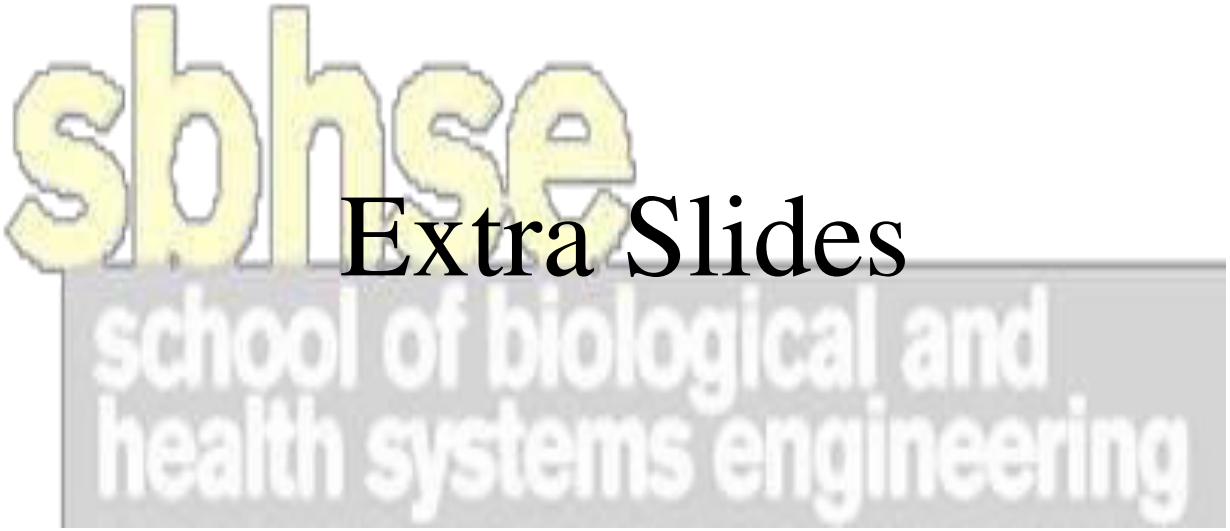
'Co-Diagnostics. (n.d.). Co-Diagnostics. Retrieved July 16, 2014, from http://www.codiagnostics.com'
'http://www.ysinhhocphantu.com/training/ky-thuat-real-time-pcr/8/'
'http://www.walgreens.com/marketing/library/contents.jsp?docid=100220&doctype=13'



Thank You

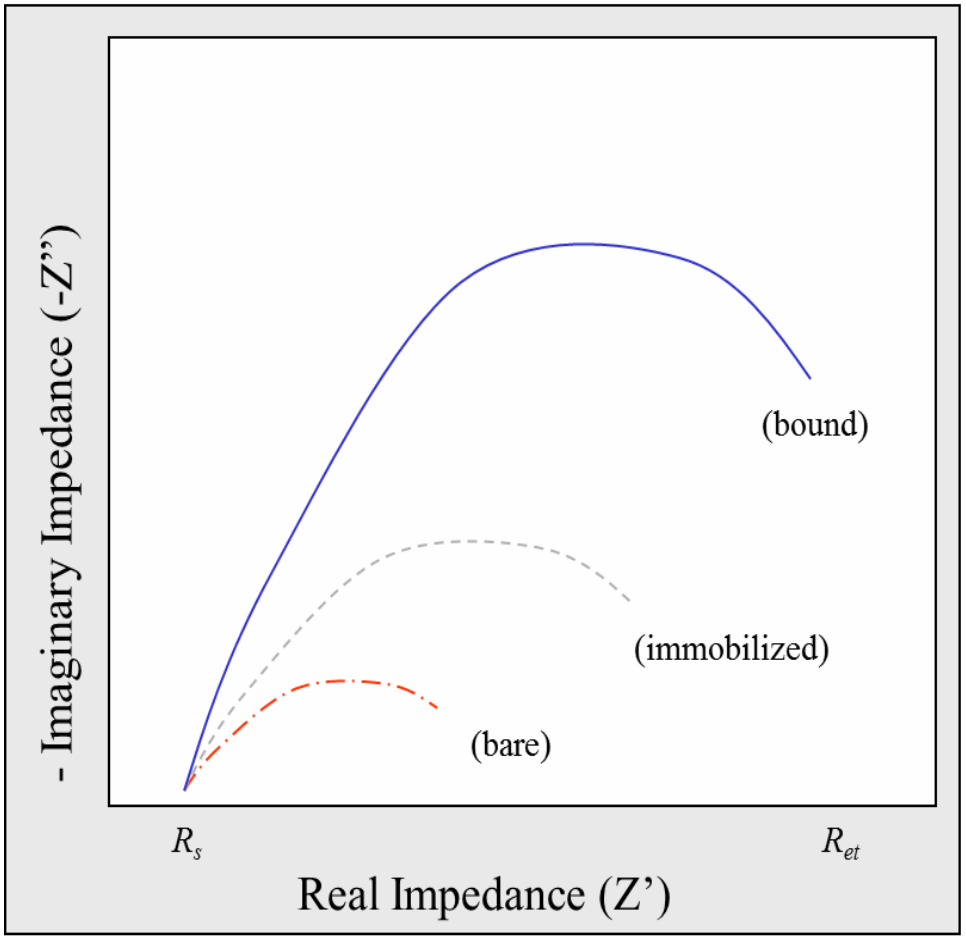
Questions?

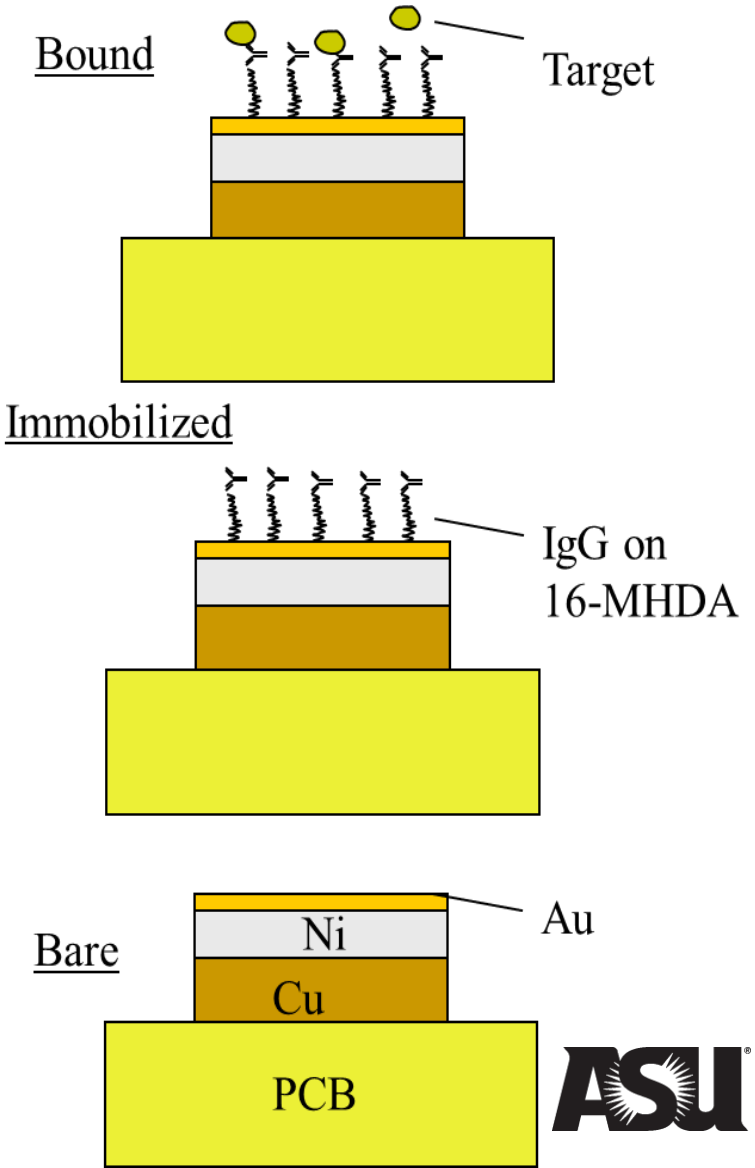
Funding from
ASU Fulton Undergraduate Research Initiative
Labelle's Army



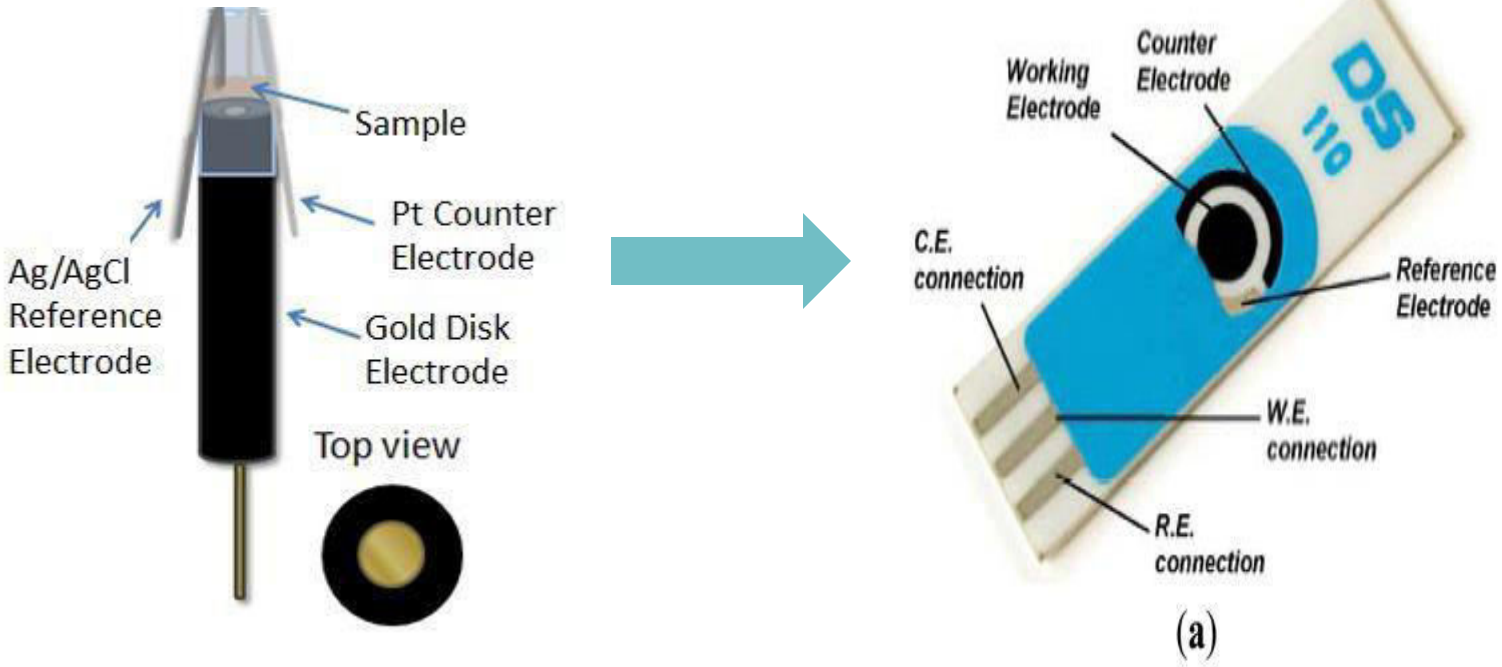
Extra Slides

Immobilization





Strip Electrodes



bioMerieux

- Identifies gram negative or positive microbes
- Load and culture the cupules and inoculate for 24-48 hrs
- A distinct pattern (20 codes) reduces into a 7 digit code you give to the company that identifies the bacteria (genus and species ID)

Metabolic analysis



↑API® strips

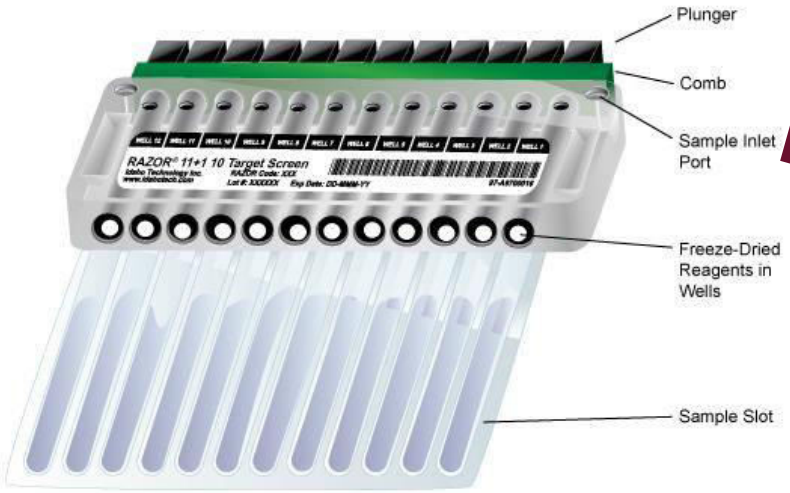
http://www.biomerieux-diagnostics.com/servlet/srt/bio/clinical-diagnostics/dynPage?open=CNL_CLN_PRD&doc=CNL_PRD_CPL_G_PRD_CLN_11&pubparams.sform=4&lang=en

JBAIDS



The 10 Pouch Format

The 10 pouch tests for ten pathogens (Anthrax, *Brucella* spp, Botulism A, *Coxiella*, *E. coli* 0157, Tularemia, Ricin, *Salmonella*, Smallpox, and Plague) from one sample using just two syringes.

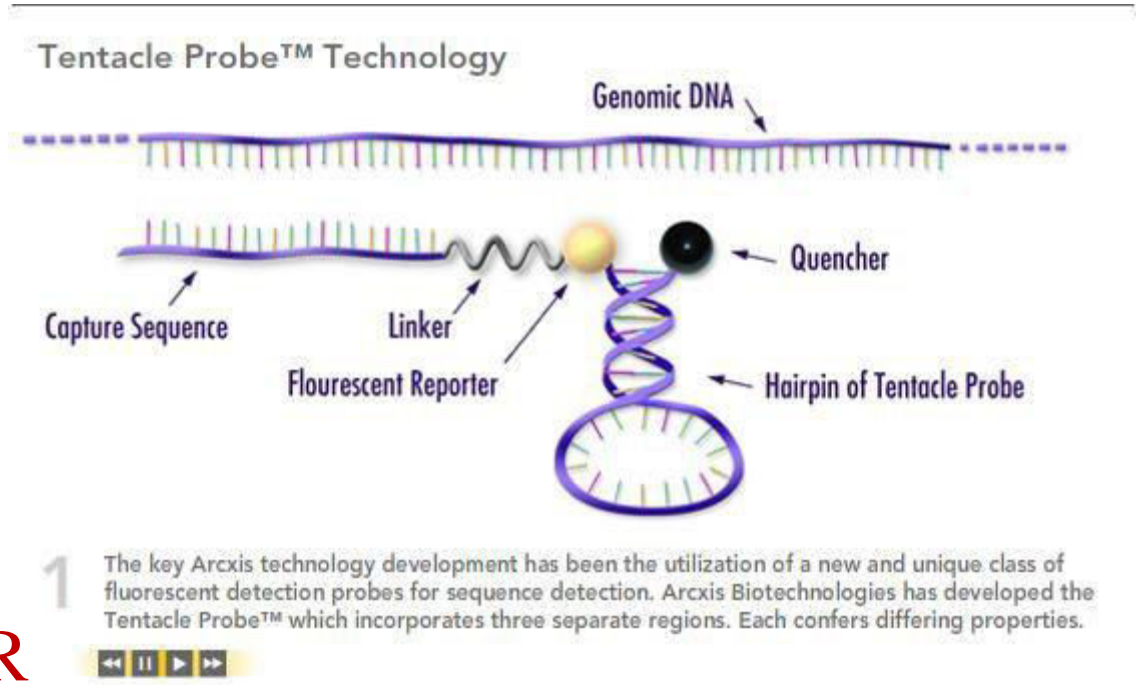


- Joint Biological Agent Identification and Diagnostic System (JBAIDS) Plague Detection kit
- *Yersinia pestis*
- Idaho Technology, Inc. Salt Lake City, UT
- Anthrax, *Brucella* spp, Botulism A, *Coxiella*, *E. coli* 0157, Tularemia, Ricin, *Salmonella*, Smallpox, and Plague
- Real time PCR

RAZOR pouches are defense articles controlled for export under the International Traffic in Arms Regulations (ITAR), administered by the U.S. Department of State, Directorate of Defense Trade Controls. The transfer of any hardware or technical data controlled by the ITAR to any unauthorized persons, requires prior written approval for from the U.S. Department of State.

Solutions – Tentacle Probe

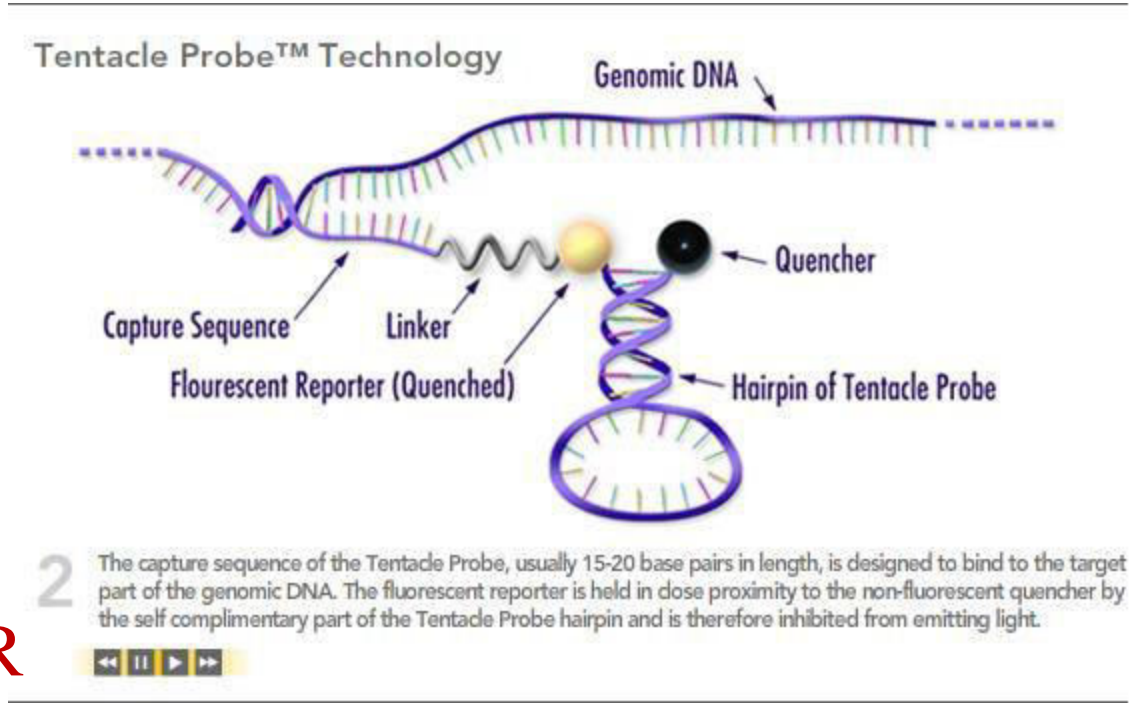
- Tentacle Probe™
- Capture Region
- Detection Region
- High Specificity
- High Sensitivity
- With or with out PCR



Arcxis Biotechnology,

Solutions – Tentacle Probe

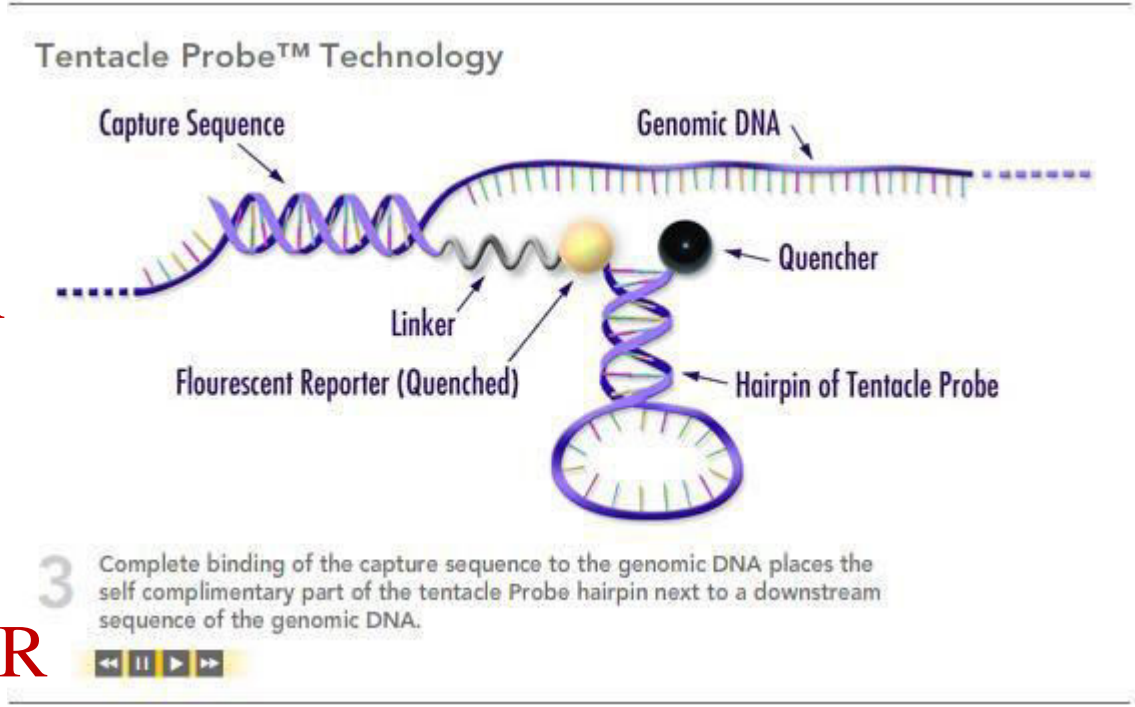
- Tentacle Probe™
- Capture Region
- Detection Region
- High Specificity
- High Sensitivity
- With or with out PCR



Arcxis Biotechnology,

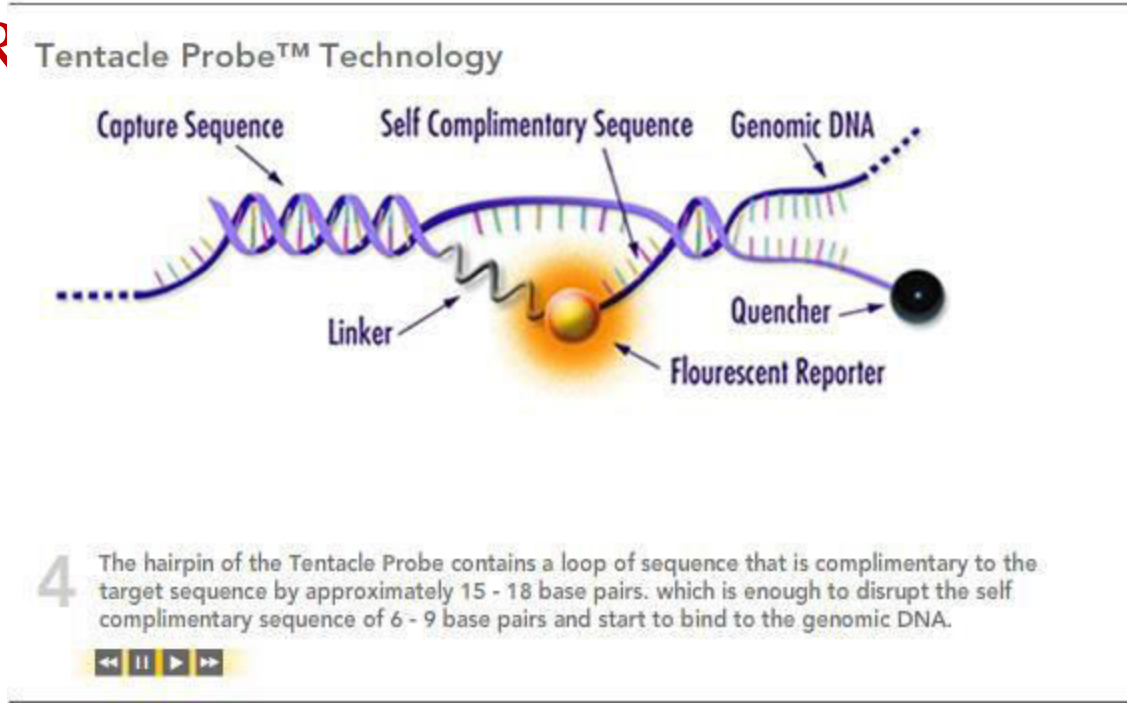
Solutions – Tentacle Probe

- Tentacle Probe™
- Capture Region
- Detection Region
- High Specificity
- High Sensitivity
- With or with out PCR



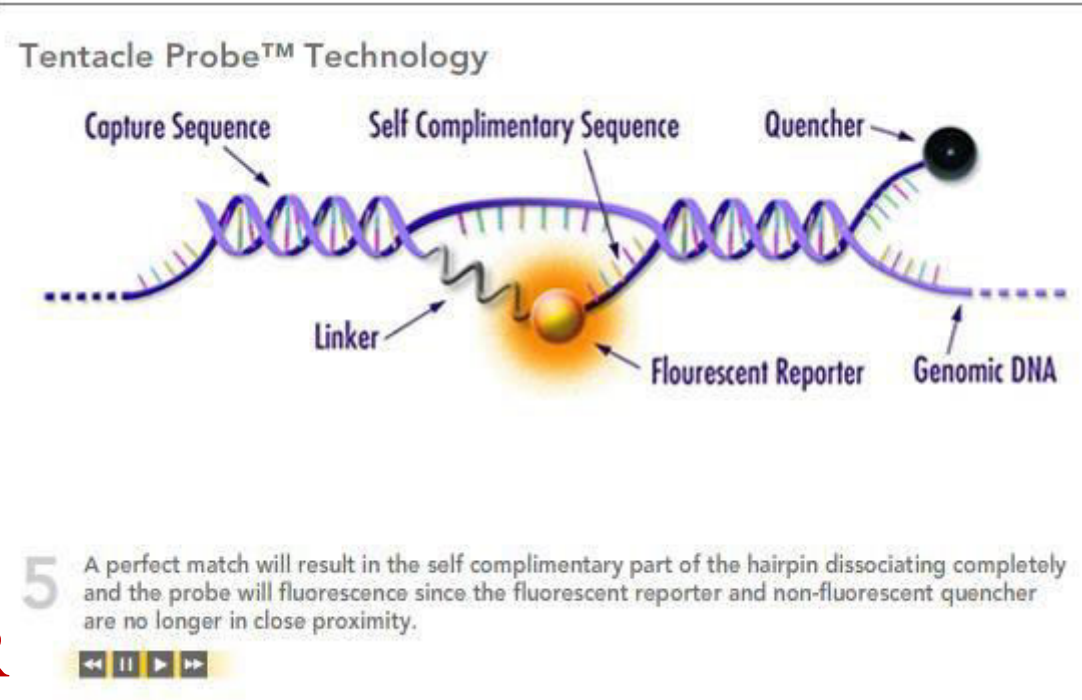
Solutions – Tentacle Probe

- Tentacle Probe™ R
- Capture Region
- Detection Region
- High Specificity
- High Sensitivity
- With or with out PC



Solutions – Tentacle Probe

- Tentacle Probe™
- Capture Region
- Detection Region
- High Specificity
- High Sensitivity
- With or with out PCR



Arcxis Biotechnology,

Solutions cont..

- Synthesized oligonucleotide
  - Capture – PEG – Hairpin
  - 18 Complementary BP – PEG – CALfluor 560-
    29 Complementary BP –BHQ1
- *B. anthracis – B.cereus*
  - *1* BP polymorphism NO FALSE POS
- *Y. pestis – Y. psudotuberculosis*
  - *25* deleted base*s* NO FALSE POS

Immobilization of Tentacle Probe™

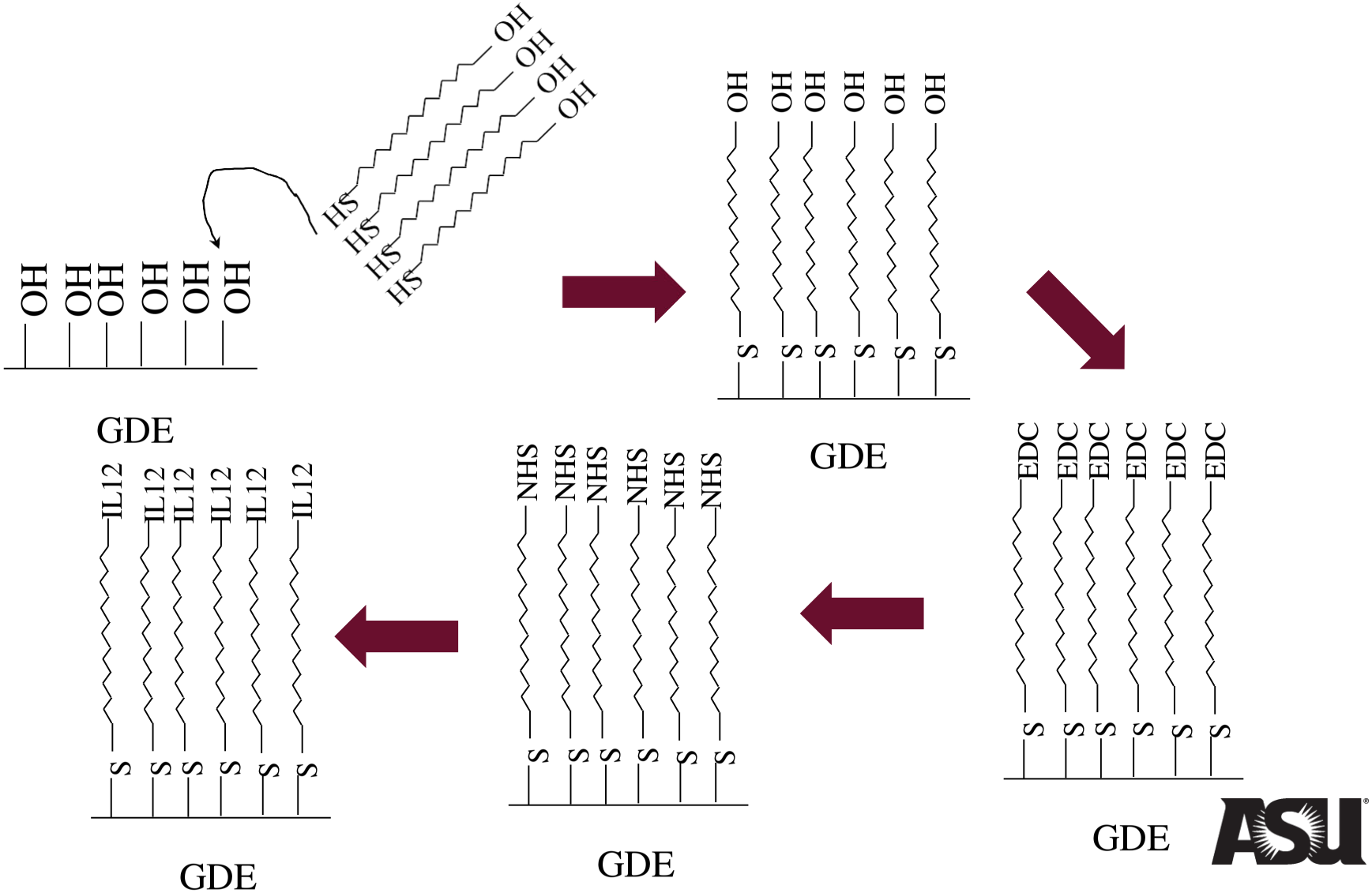


Table 1

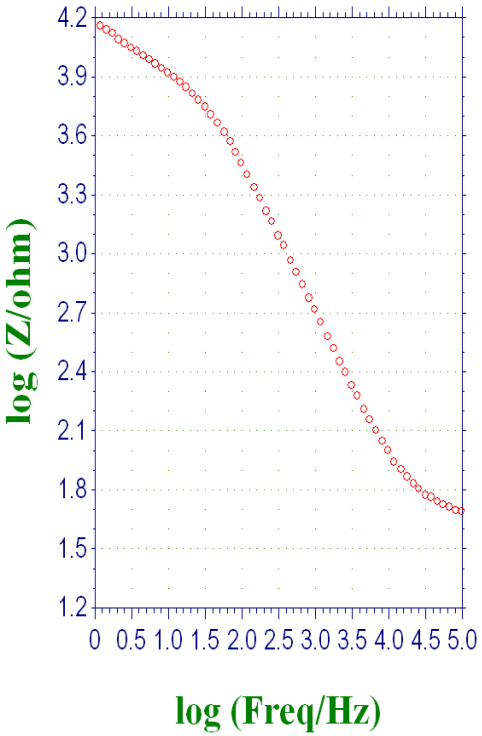
Table 1 – list of descriptions, pros and cons for common electrochemical techniques

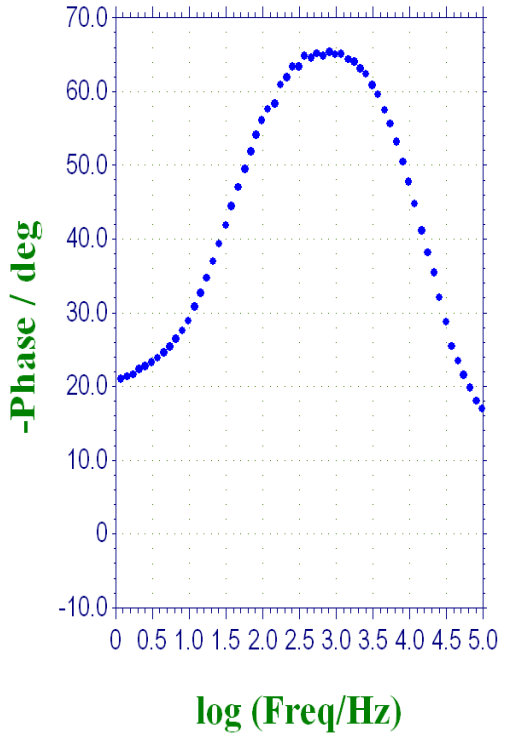
Electrochemical Technique	Description	Pros	Cons
[1]			
Amperometric i-t (AMP-it) [2]	A method in which one voltage is applied to the solution system and current over time is measured. This is very popular and well-developed to measure blood glucose levels.	Intrinsically has continuous time abilities and chosen voltage. This technique is well known.	The detection range is 10^{-4}-10^{-6} M.
[3]			
Square Wave Voltammetry (SWV) [4]	Essentially, a more sensitive CV using differential voltage sweeping, but does only oxidation or only reduction, as it sweeps voltage in one direction.	A more sensitive determination of oxidation or reduction voltages of the sample.	Can only sweep voltage in one direction and detection range is 10^{-6}-10^{-9} M.
[5]			

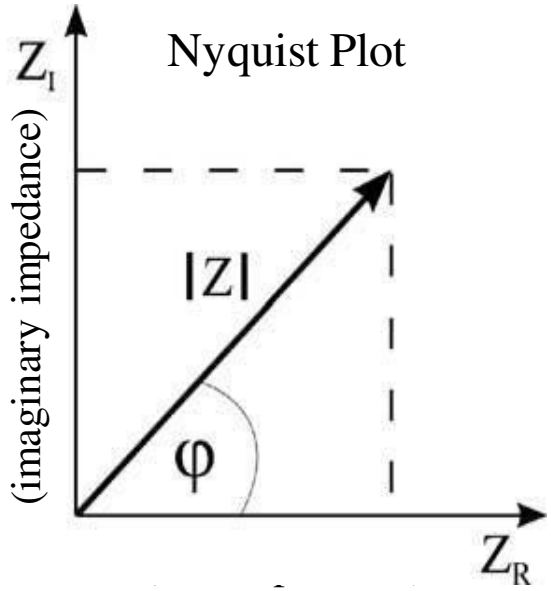
Wang, 2006 [2] Bishop 2010 [3] Ie, 2008 [4] Cai 2009 [5] Bhavsar, 2009

Electrochemical Impedance Spectroscopy (EIS) Background

Graphically







Lisdat et al, Bioanal. Chem, 2008

Mathematically

$$Z_R = Z\cos(\varphi)$$ (real impedance)

$$Z_i = Z\sin(\varphi)$$ (imaginary impedance)

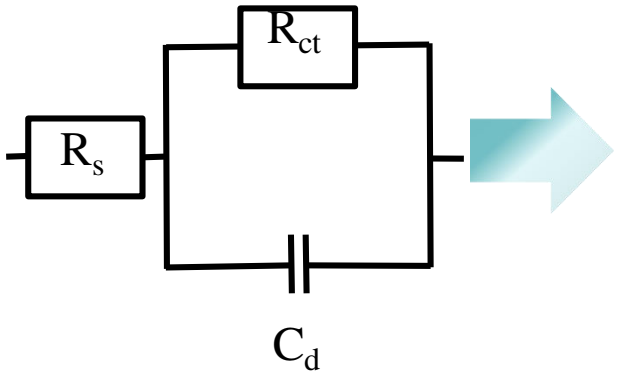
$$Z(j\omega) = \frac{U(j\omega)}{I(j\omega)} = Z_r(\omega) + jZ_i(\omega)$$

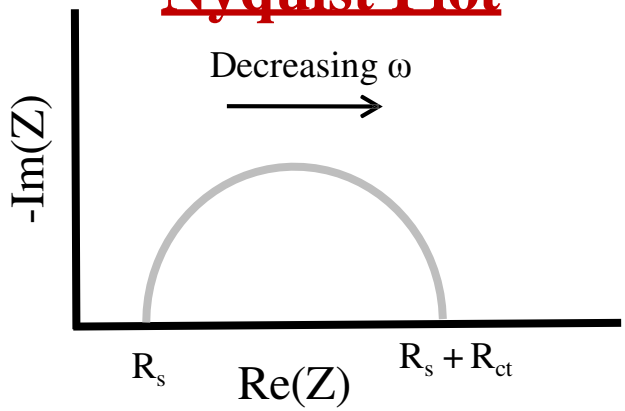
Background: EIS Equivalent Circuit Models

Circuit Model

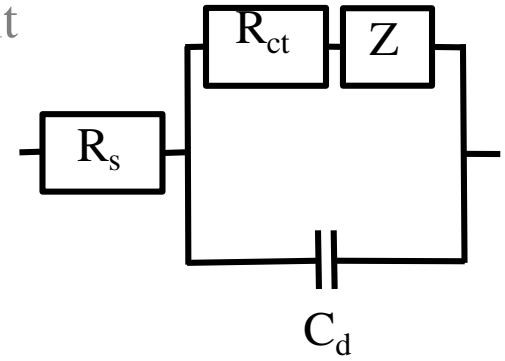
Nyquist Plot

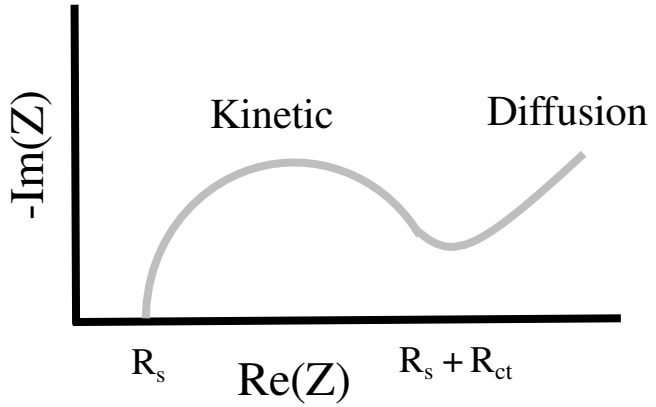
Randles Circuit





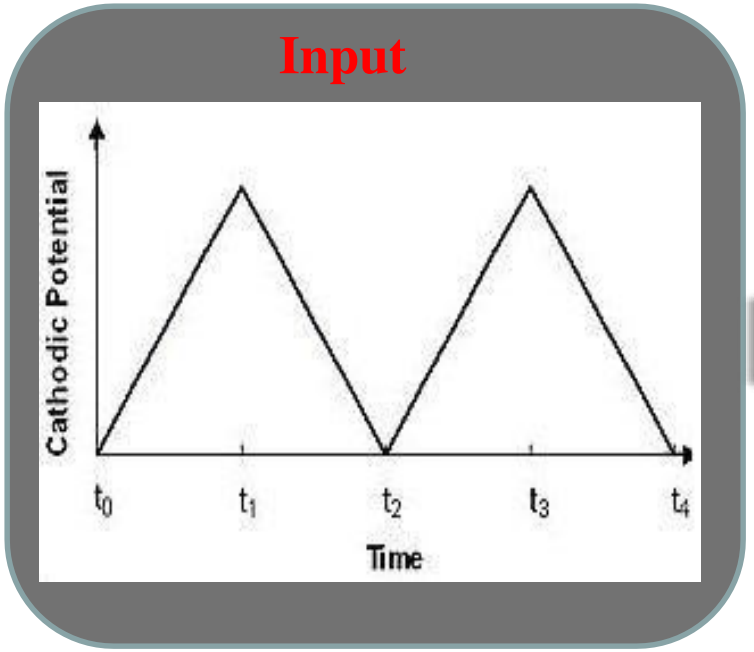
Warburg Circuit

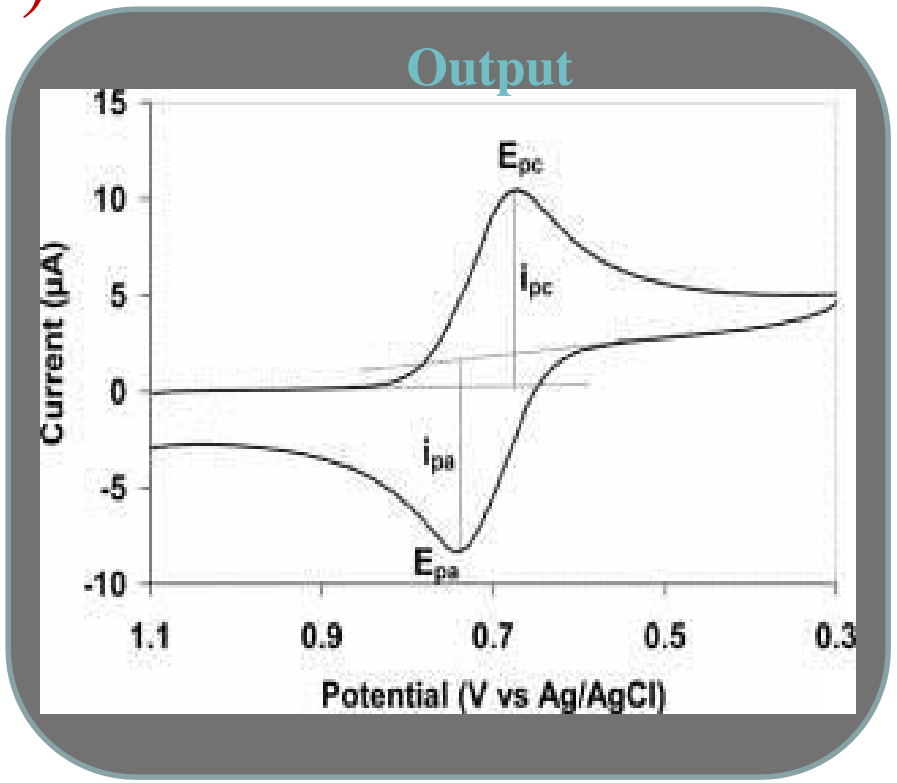




Electrochemical Techniques

Cyclic Voltammetry (CV)





Useful for other techniques:

SWV

Amp i*t

EIS

Formal Potential: Average of Reduction and Oxidation Peak Voltages

La Belle, E. Chem. 101. 2011.

Let Us Meet Again

We welcome you all to our future conferences of OMICS Group International

Please Visit:

www.omicsgroup.com

www.conferenceseries.com

www.pharmaceuticalconferences.com