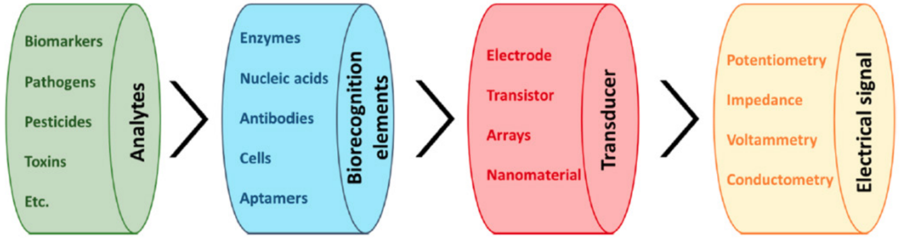
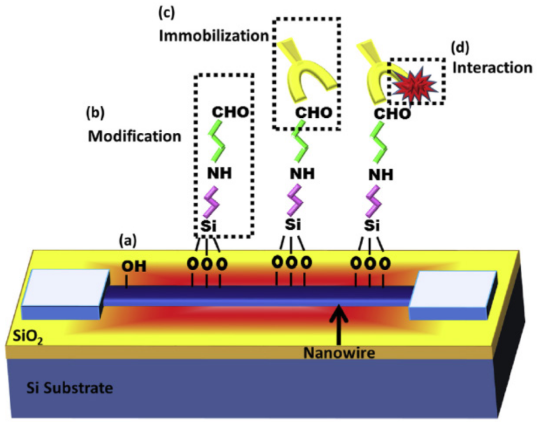
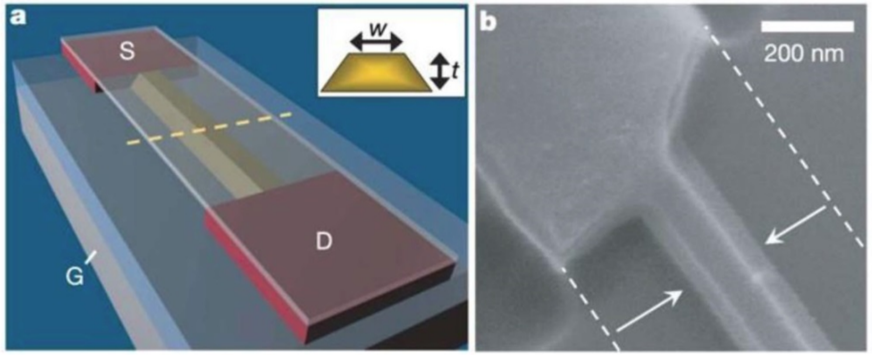
# Introduction to BioMEMS & Medical Microdevices

# *Biosensors*

Prof. Steven S. Saliterman, http://saliterman.umn.edu/



# Topics

- Biosensor Types & Classification
  - COVID-19 sensor example.
- Optimization
- Biological recognition elements.
- Transducers
  - Field effect transistors (FETs).
- Nanotechnology - nanoparticles
- Immobilization techniques.
  - APTES-GA (3-aminopropyltriethoxysilane) method.
  - Ab, Enzyme, Biotin/Avidin, Aptamer
- Electrical Sensing
  - Electrochemical techniques.

# Biosensors – Some Definitions

- *"A device that uses specific biochemical reactions mediated by isolated enzymes, immune systems, tissues, organelles or whole cells to detect chemical compounds by electrical, thermal or optical signals."*
- A biological or biologically derived *sensitive recognition element* usually is *immobilized* on a *transducer* to measure one or more *analytes*.

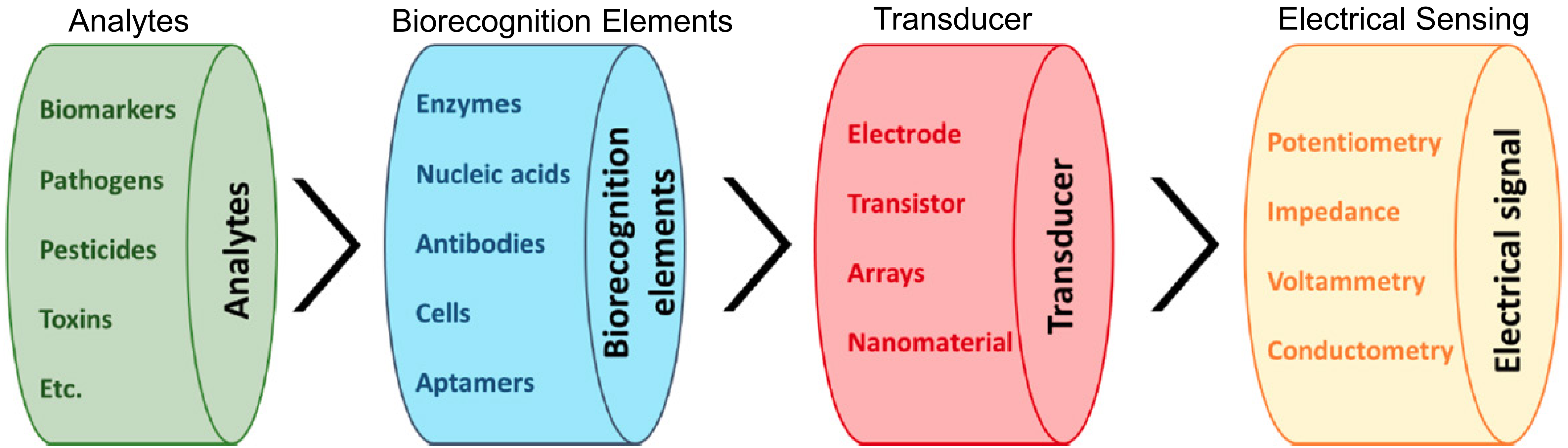

Int. Union of Pure and Applied Chemistry
Syu YC, Hsu WE, Lin CT. Review-Field-Effect Transistor Biosensing: Devices and Clinical Applications. *Ecs Journal of Solid State Science and Technology*. 2018;7(7):Q3196-Q3207.
Luka G, Ahmadi A, Najjaran H, et al. Microfluidics Integrated Biosensors: A Leading Technology towards Lab-on-a-Chip and Sensing Applications. *Sensors*. 2015;15(12):30011-30031.

# Uses of Biosensors…

- Commonly combined with microfluidic systems for:
  - High throughput processing,
  - Enhanced transport for controlling the flow conditions,
  - Increased mixing rate of different reagents,
  - Reduced sample and reagent volumes (down to nanoliter), increase sensitivity of detection, and utilizing the same platform for both sample preparation and detection.
  - Portability, disposability, real-time detection, unprecedented accuracies, and simultaneous analysis of different analytes in a single device.

Syu YC, Hsu WE, Lin CT. Review-Field-Effect Transistor Biosensing: Devices and Clinical Applications. *Ecs Journal of Solid State Science and Technology.* 2018;7(7):Q3196-Q3207.
Luka G, Ahmadi A, Najjaran H, et al. Microfluidics Integrated Biosensors: A Leading Technology towards Lab-on-a-Chip and Sensing Applications. *Sensors.* 2015;15(12):30011-30031.

# Biosensing…



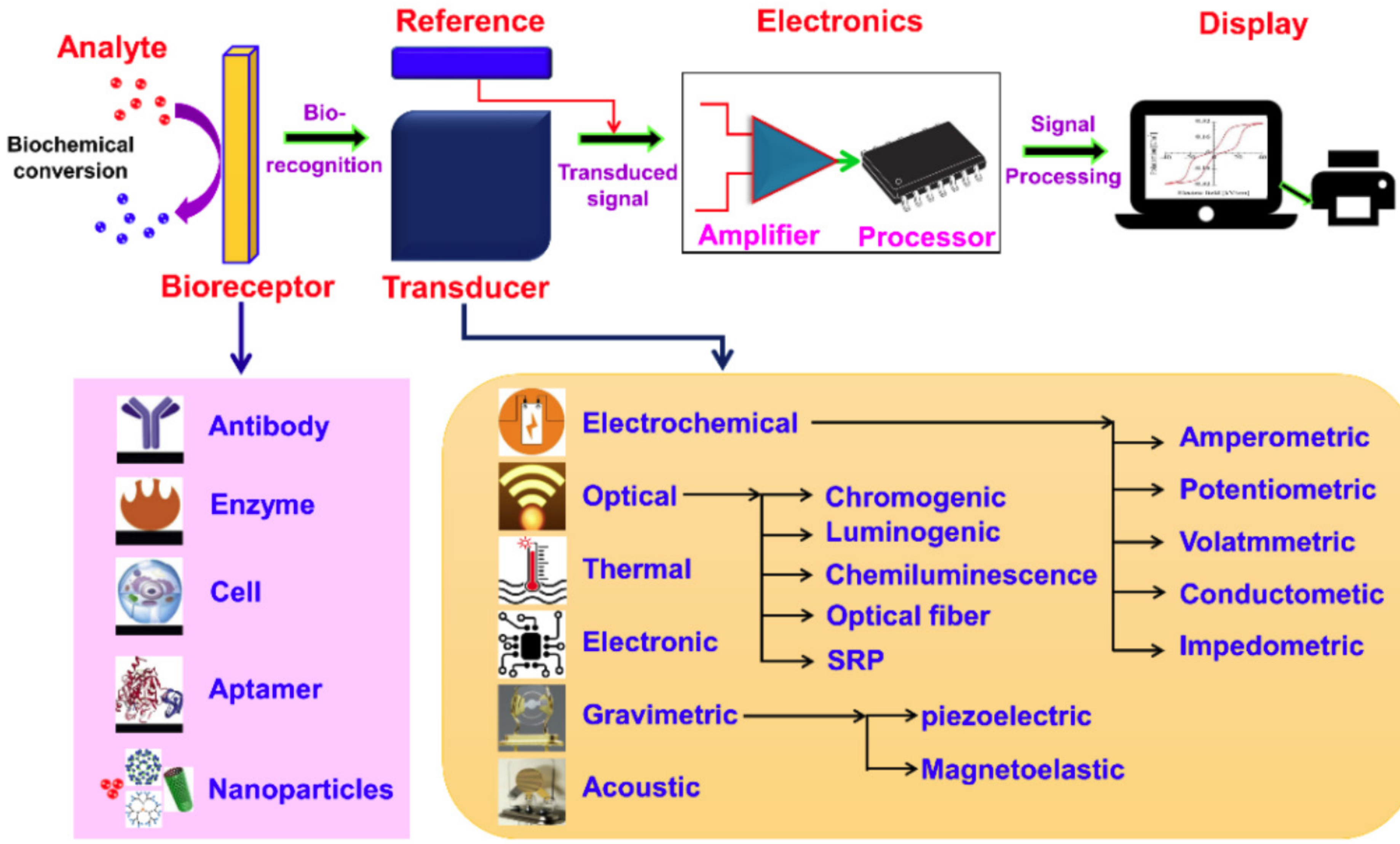

Sample preparation – filtration, extraction, purification and enrichment.

Ideally retained in direct contact. The biomolecules change physical quantities, such as charge, mass or photons.

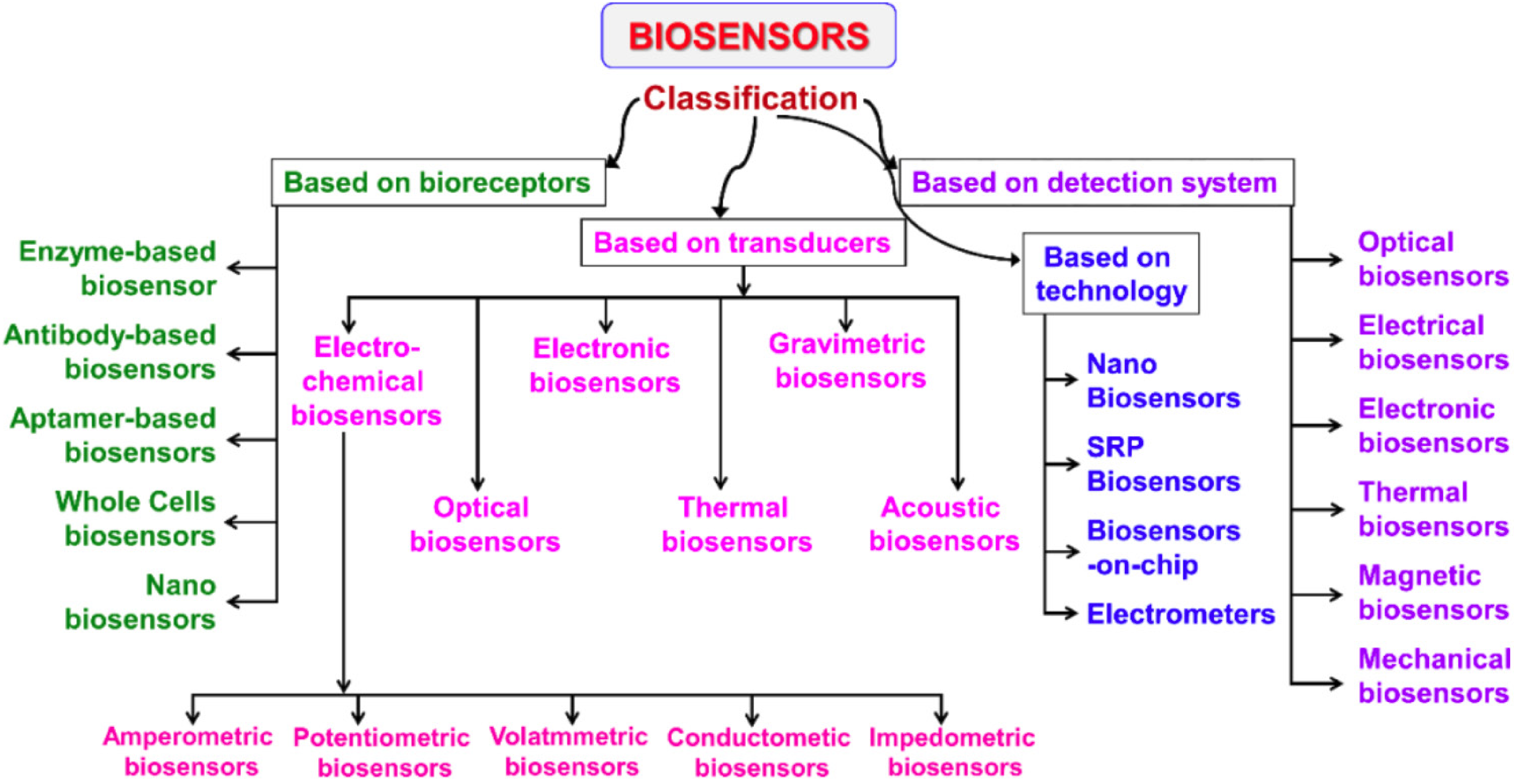
Measurement of current, potential, or field effect, realized through label-free or labeled approaches.



Wongkaew N, Simsek M, Griesche C, Baeumner AJ. Functional Nanomaterials and Nanostructures Enhancing Electrochemical Biosensors and Lab-on-a-Chip Performances: Recent Progress, Applications, and Future Perspective. *Chemical Reviews.* 2019;119(1):120-194.

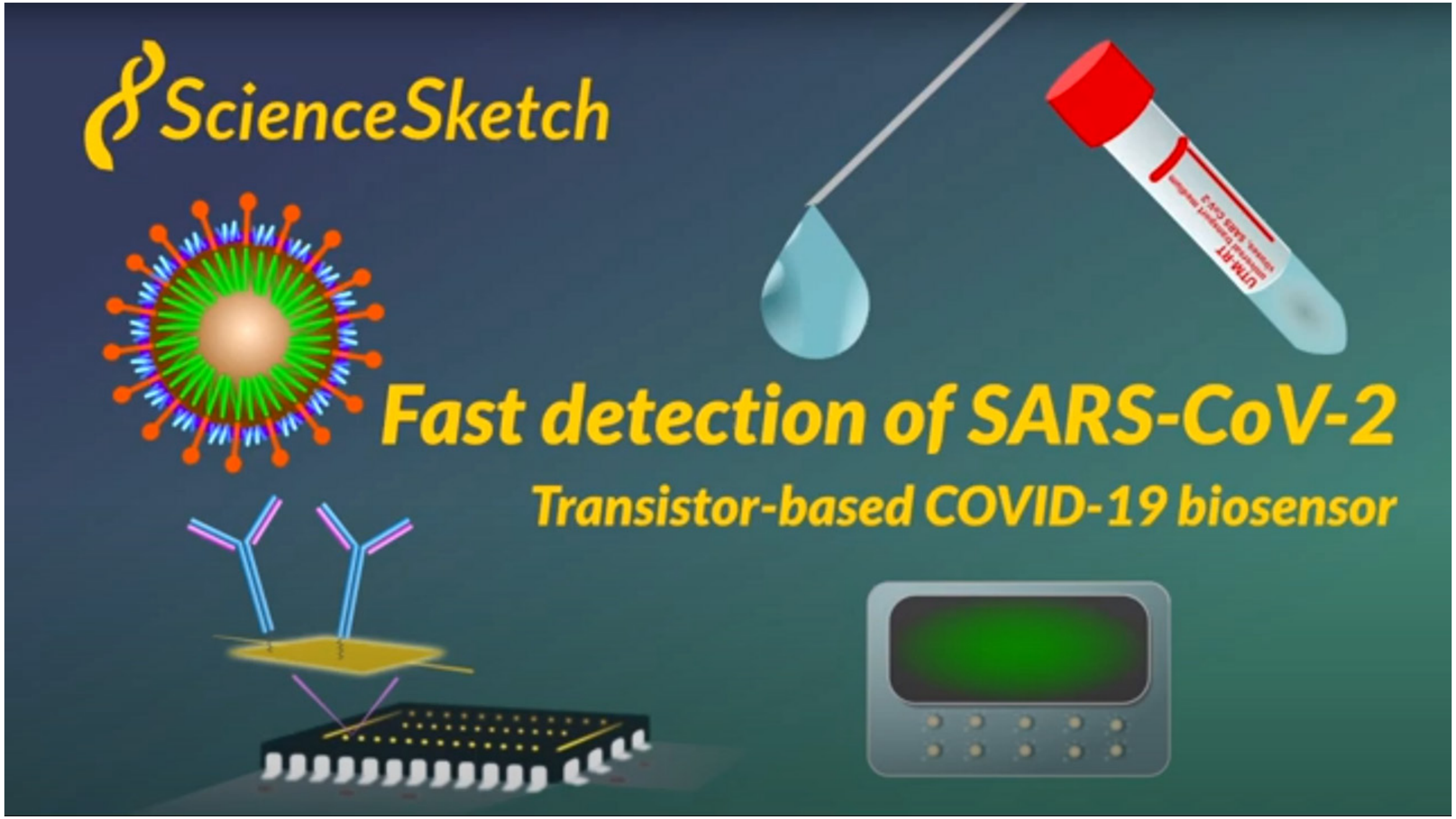



Naresh V, Lee N. A Review on Biosensors and Recent Development of Nanostructured Materials-Enabled Biosensors. *Sensors*. Feb 2021;21(4)1109. doi:10.3390/s21041109




Naresh V, Lee N. A Review on Biosensors and Recent Development of Nanostructured Materials-Enabled Biosensors. *Sensors*. Feb 2021;21(4)1109. doi:10.3390/s21041109

# Example: COVID-19 Detection…






https://youtu.be/gmd5gSvZzV8

# Optimization of Biosensors

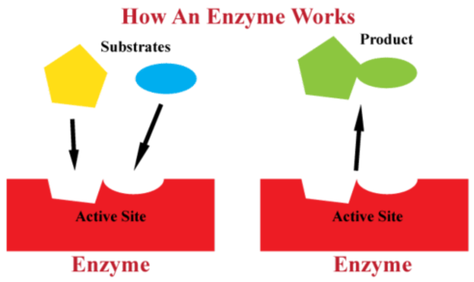
- Selectivity – detect the analyte of interest.
- Sensitivity – minimum amount of analyte that can be detected.
- Linearity - better linearity means the higher the substrate concentration detection.
- Response Time – time to obtain 95% of the test result.
- Reproducibility – precision (repeatability) and accuracy (generating a mean value close the actual value).
- Stability – affected by the affinity of the bioreceptor and its degradation over time.



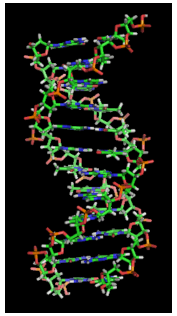
Naresh V, Lee N. A Review on Biosensors and Recent Development of Nanostructured Materials-Enabled Biosensors. *Sensors*. Feb 2021;21(4)1109. doi:10.3390/s21041109

# Biological Recognition Elements

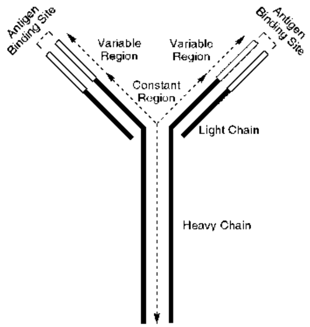
- Enzymes
  - *Catalyst* for biochemical reactions that act upon *substrate* molecules producing a *product.*

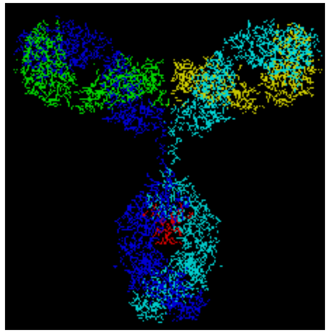



- Nucleic acids
  - DNA, RNA – composed of nucleotides. Adenine, Thymine, Guanine, Cytosine (DNA) or Uracil (RNA). A-T, G-C or G-U.



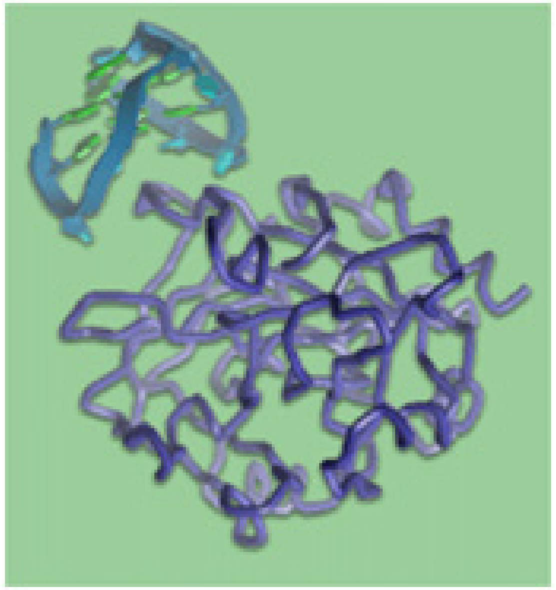
- Antibodies (Immunoglobulins)
  - Proteins produced in response to and *counteracting* a specific *antigen* (eg. bacteria, virus, or foreign substance). IgG, IgM, IgA, IgE & IgD.



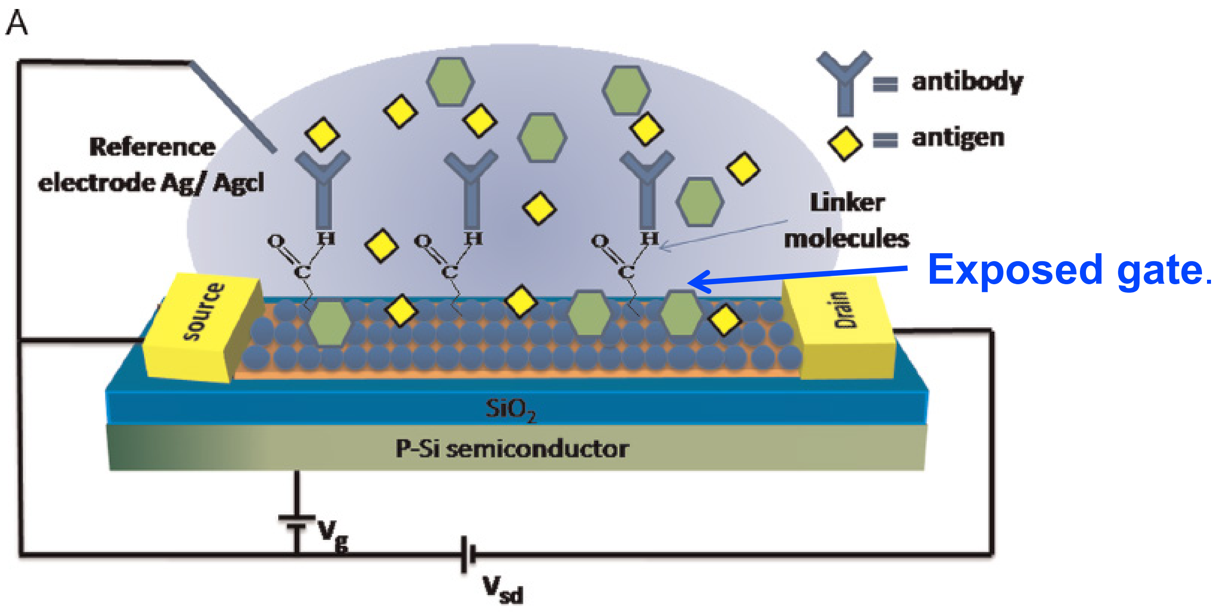



Enzyme illustration courtesy of Wilson, Z CK-12 Foundation
DNA courtesy of Wikimedia.org.
Antibody molecule courtesy of http://www.umass.edu/microbio/rasmol/padlan.htm.
Aptamer image Image courtesy of Archemix.

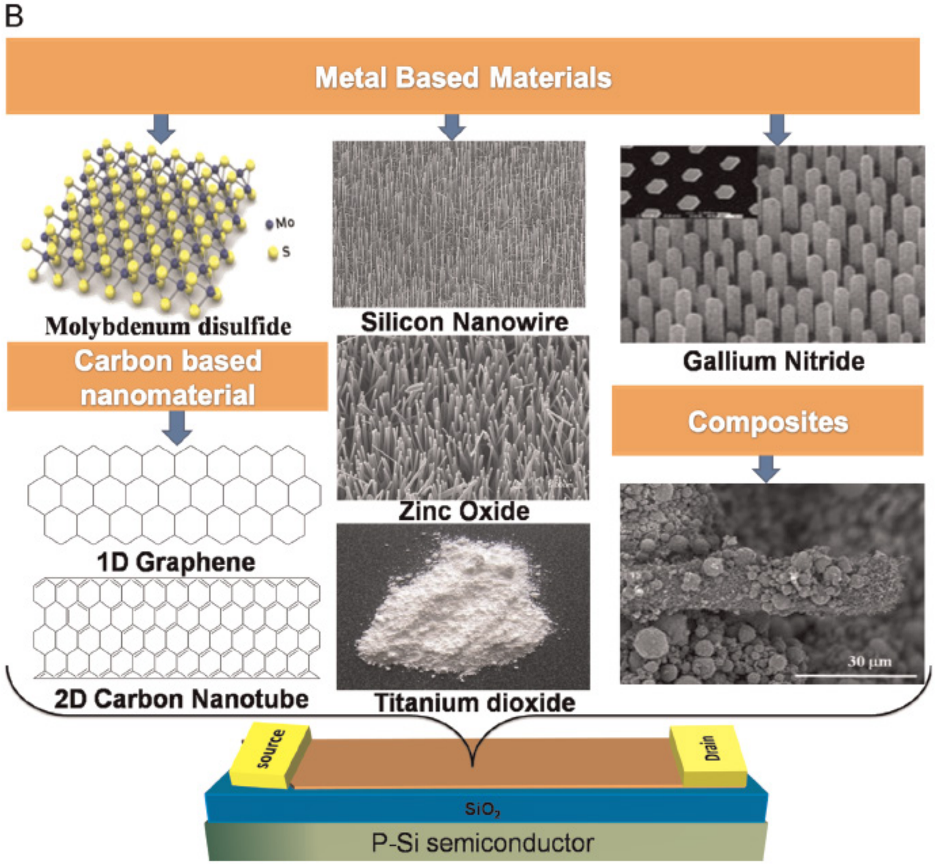
- Aptamers
  - These are *artificial nucleic acid ligands or peptide molecules* that can be generated against amino acids, drugs, proteins and other molecules. Function similar to antibodies.
  - They are single-stranded DNA or RNA (ssDNA or ssRNA) molecules that *to bind to various molecular targets* such as small molecules, proteins, nucleic acids, and even cells, tissues and organisms.
    - They bind because they fit their target, and by non-covalent interactions.
    - *Peptide aptamers* can bind cellular protein targets and exert biological effects, including interference with the normal protein interactions of their targeted molecules with other proteins.



# Transducers – eg. Field Effect Transistor




FET-based biosensor, based on complimentary metal oxide semiconductor (CMOS) technology.



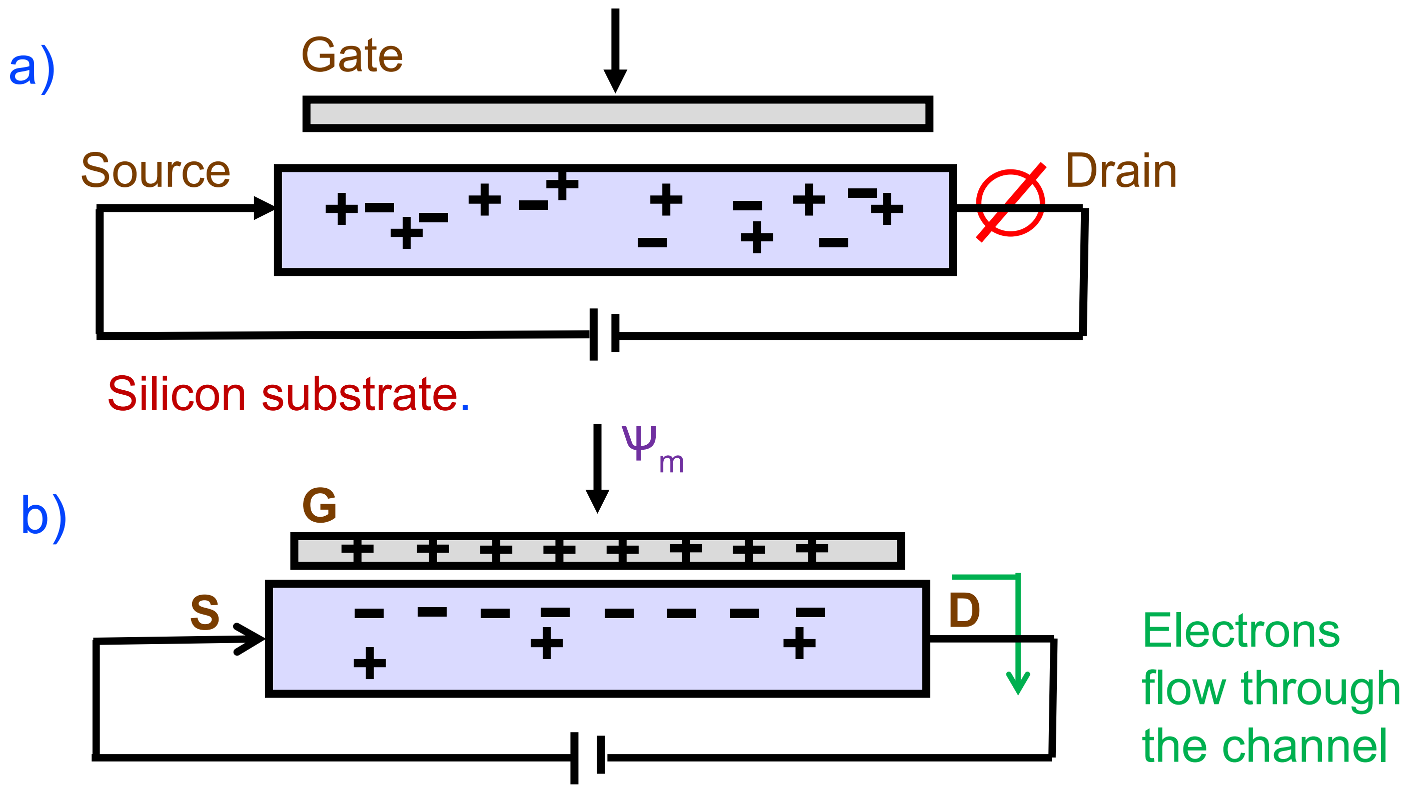

Different nanomaterials embedded onto the gated region of the FET-based biosensor.



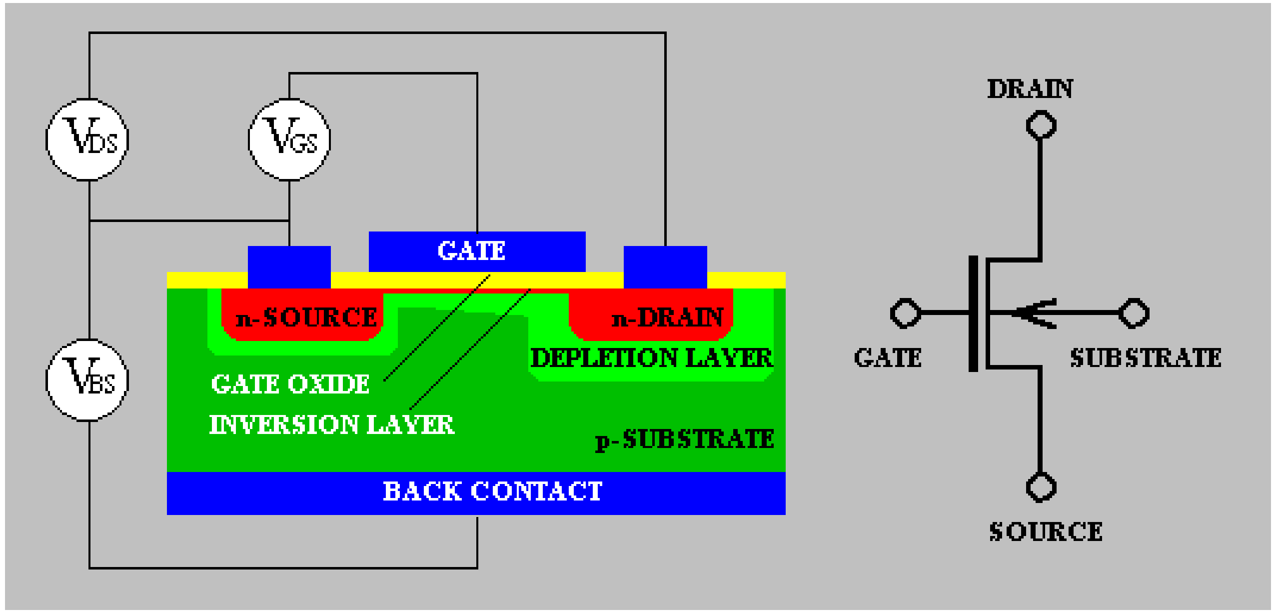
Nehra A, Singh KP. Current trends in nanomaterial embedded field effect transistor-based biosensor. *Biosensors & Bioelectronics.* 2015;74:731-743.

# *FET Conceptually…*

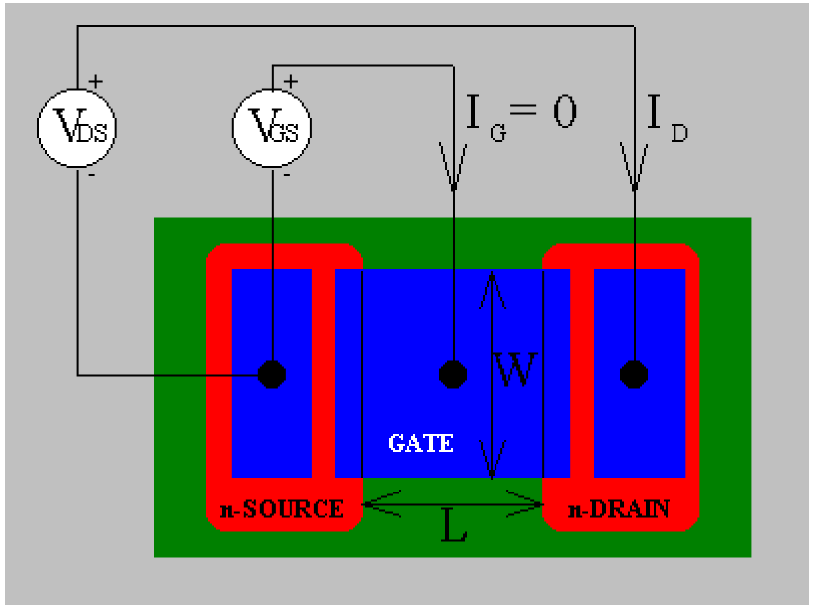
- When the gate metal potential ($\Psi_m$- psi) is changed, the electric field induces the "band bending" of the semiconductor channel accordingly.
- This results in channel carrier concentration changes, such as *inversion, depletion or accumulation (negative gate).*
- The gate potential can be given by other factors such as pH, ions or charge of biomolecules.



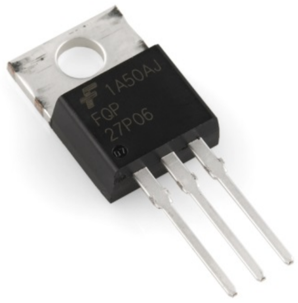

Syu YC, Hsu WE, Lin CT. Review-Field-Effect Transistor Biosensing: Devices and Clinical Applications. *Ecs Journal of Solid State Science and Technology.* 2018;7(7):Q3196-Q3207




Metal Oxide Semiconductor Field Effect Transistor




Top View




Package

- A metal or polysilicon gate covers the region between the source and drain.
- These electrons form a conducting channel between the source and the drain, called the ***inversion layer***.
- The flow of electrons from the source to the drain is controlled by the voltage applied to the gate.

Bart J. Van Zeghbroeck https://ecee.colorado.edu/~bart/book/mosintro.htm

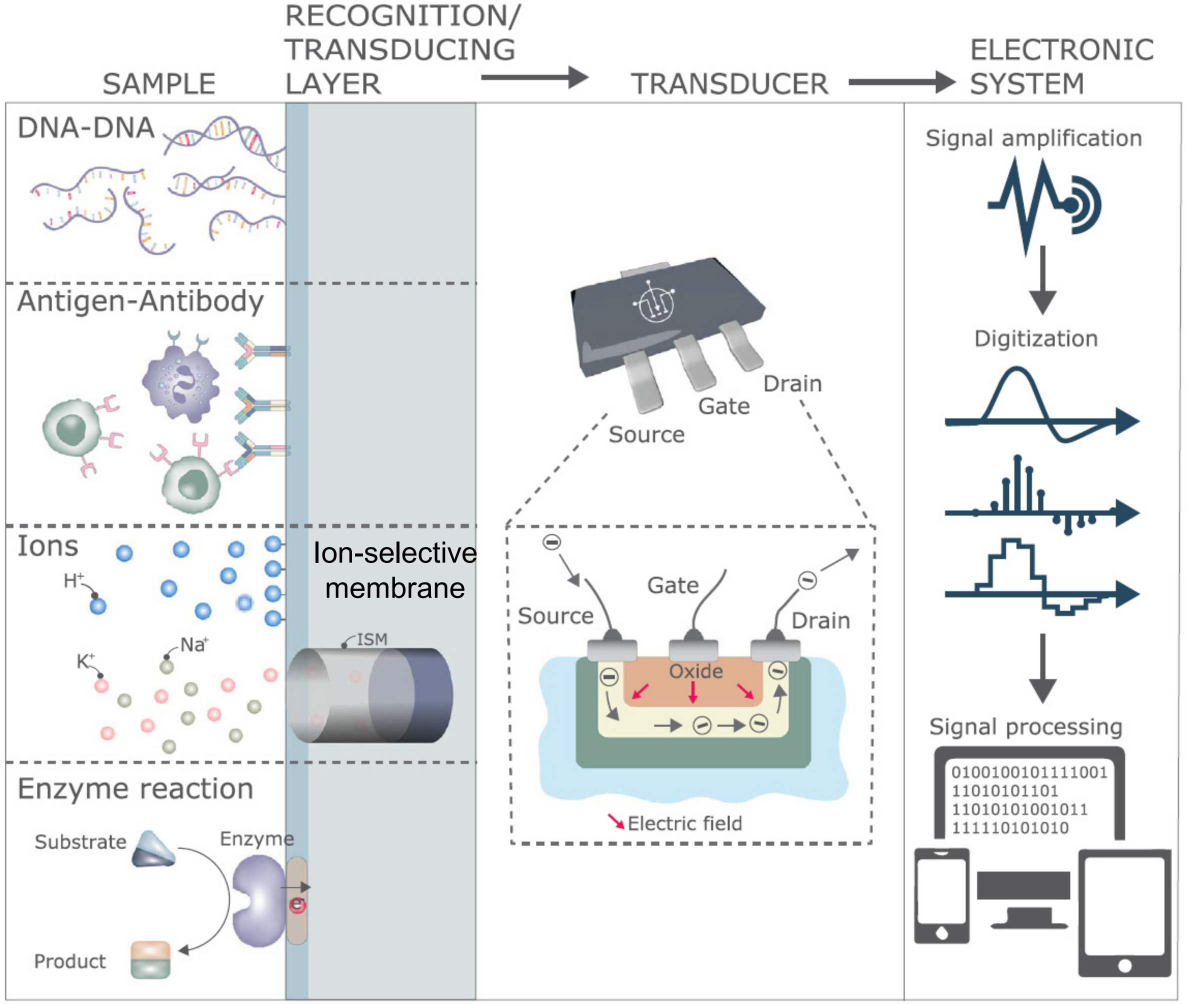
# Types & Advantages of FET

- Types:
  - Ion selective FET (ISFET) – conventional and double gate.
  - Silicon nanowire biosensors.
  - Organic FET and graphene FET biosensors.
- Miniature, ultra sensitive and fast response time.
- Respond to electrostatic charges *and* potential changes.
  - Detection of nucleotides, amino acids, cells (e.g. bacterial and viruses).
- Arrays may allow parallel processing.
- Suitable for integration with other electronics.
- Excellent for point-of-service devices of the future.

Syu YC, Hsu WE, Lin CT. Review-Field-Effect Transistor Biosensing: Devices and Clinical Applications. *Ecs Journal of Solid State Science and Technology.* 2018;7(7):Q3196-Q3207

# *Biological & Chemical FET Sensor…*

- The conventional ion-selective FET (ISFET) is comprised of a MOSFET *with the metal gate replaced by a dielectric layer as a sensing membrane.*
- This dielectric is normally silicon dioxide.




*Steven S. Saliterman*

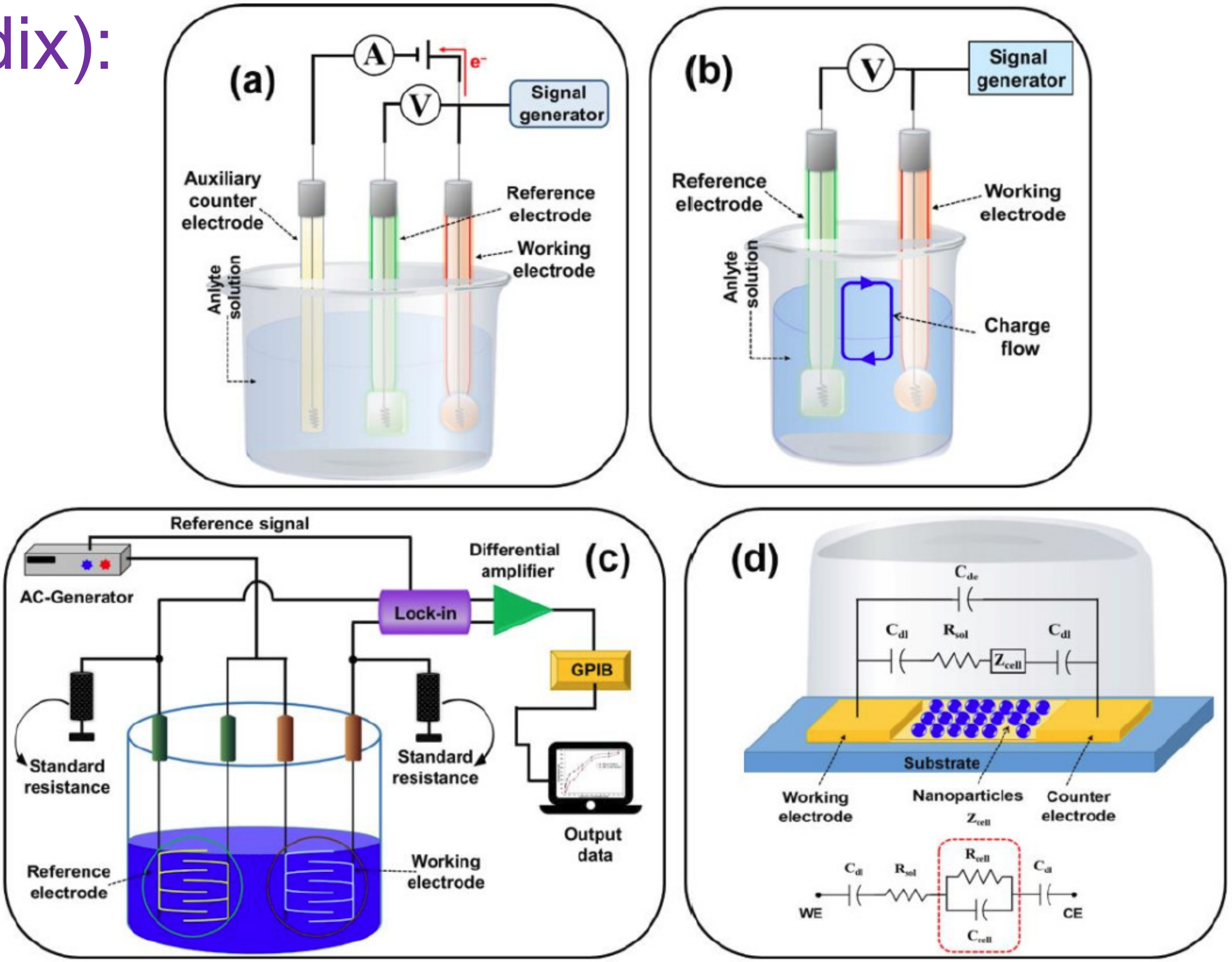
Kaisti M. Detection principles of biological and chemical FET sensors. *Biosensors & Bioelectronics.* 2017;98:437-448.

# Common Sensing Methods

## Electrochemical Sensing (see Appendix):

a) Amperometric/Voltametric
b) Potentiometric
c) Conductometric biosensors
d) Equivalent circuit of the impedimetric biosensor

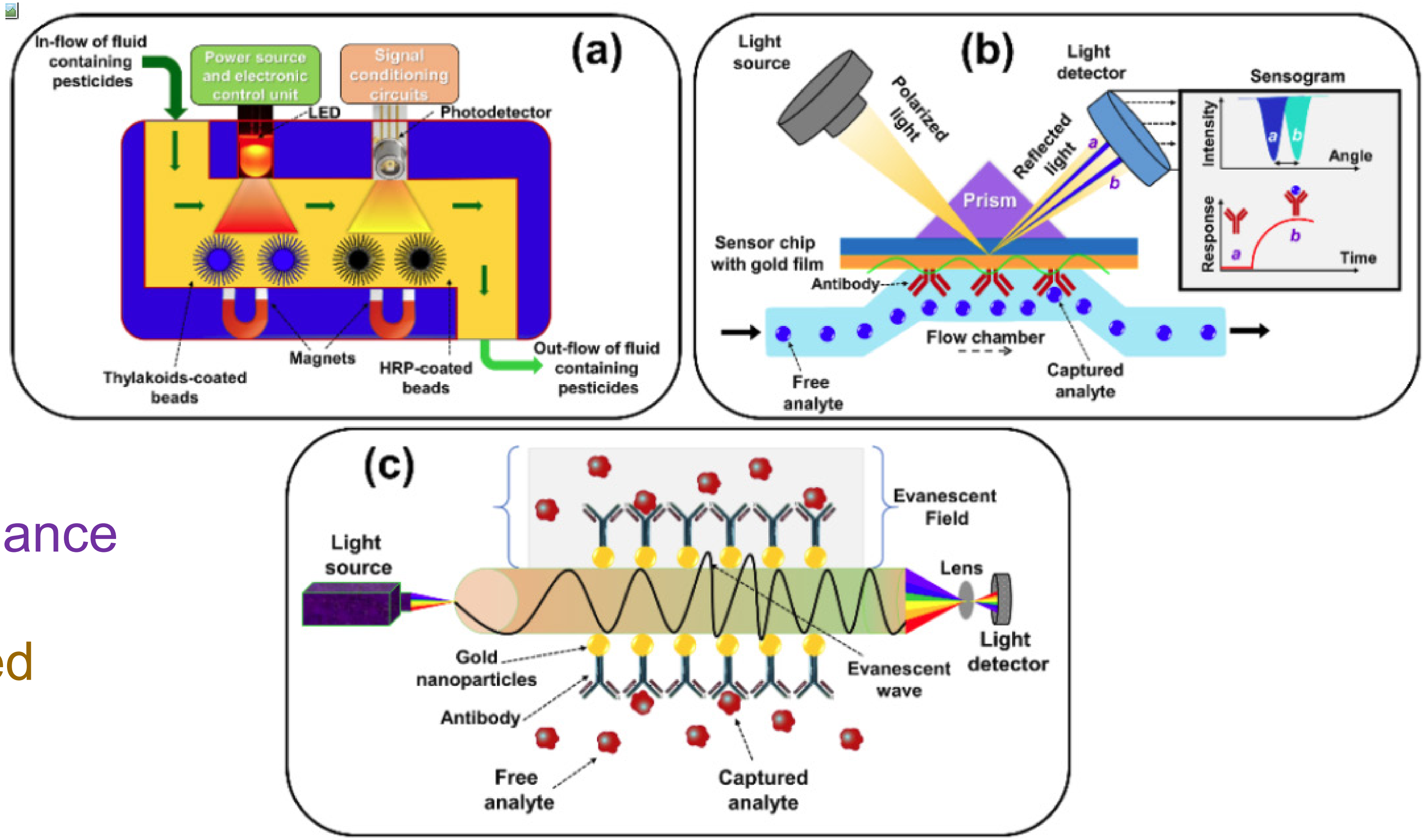
(Cdl = double-layer capacitance of the electrodes, Rsol = resistance of the solution, Cde = capacitance of the electrode, Zcell = impedance introduced by the bound nanoparticles, and Rcell and Ccell are the resistance and capacitance in parallel).




*Steven S. Saliterman*

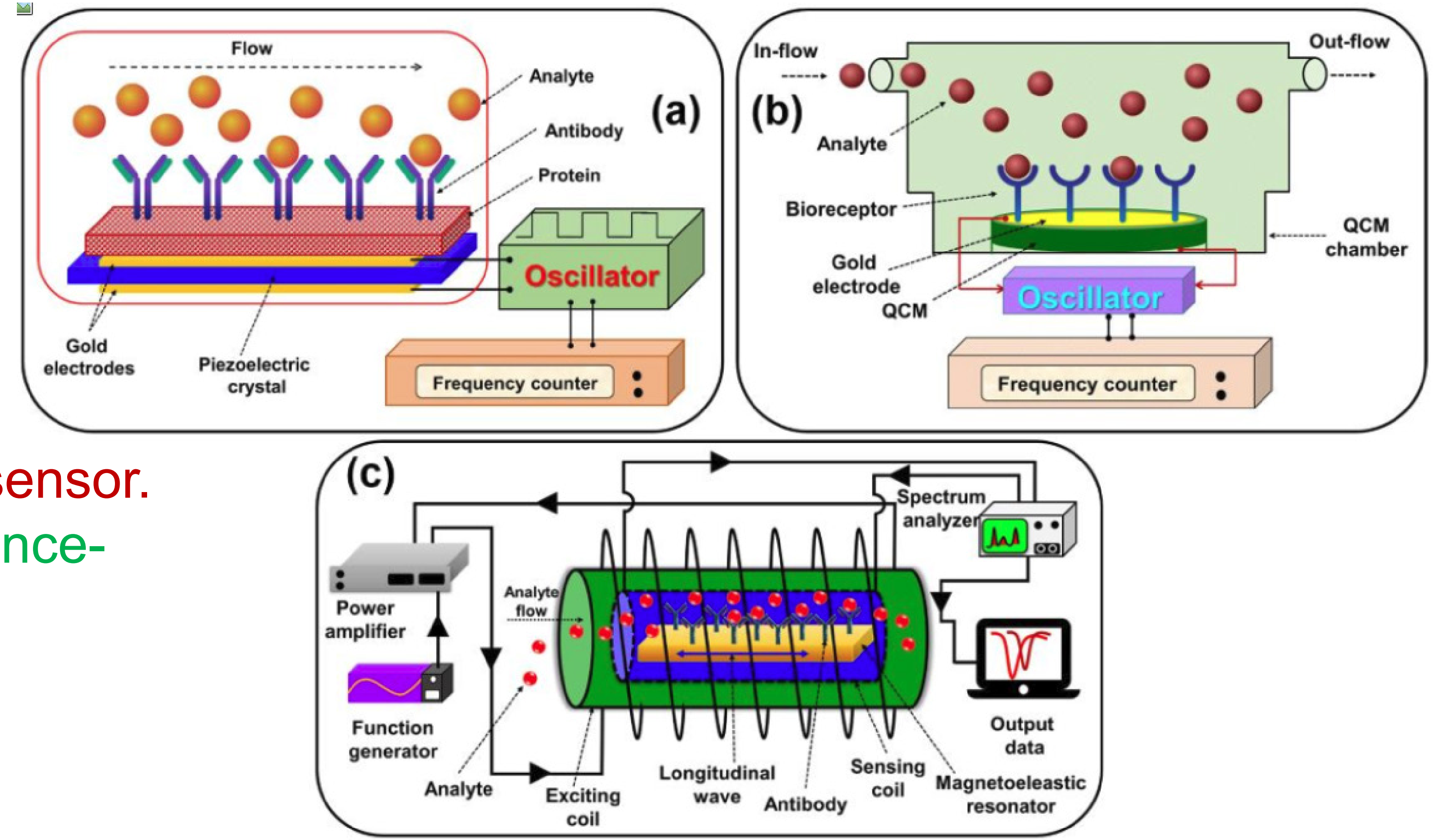
Naresh V, Lee N. A Review on Biosensors and Recent Development of Nanostructured Materials-Enabled Biosensors. *Sensors*. Feb 2021;21(4)1109. doi:10.3390/s21041109

# More Sensing Methods…




a) Chemiluminescence biosensor.
b) Surface plasmon resonance (SPR) biosensor.
c) Evanescent wave-based optical fiber biosensor.

Naresh V, Lee N. A Review on Biosensors and Recent Development of Nanostructured Materials-Enabled Biosensors. *Sensors*. Feb 2021;21(4)1109. doi:10.3390/s21041109

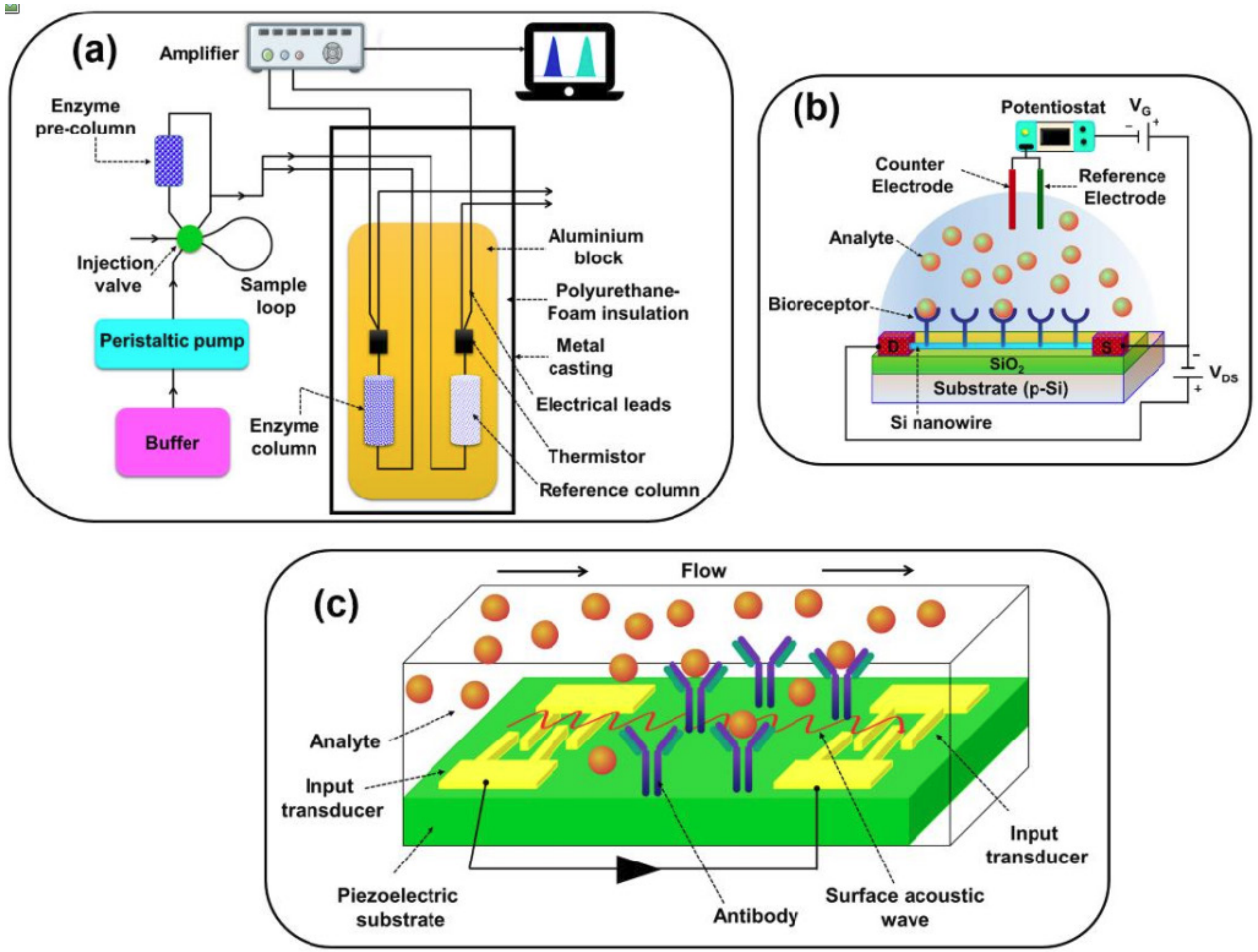



a) Piezoelectric-based biosensor.
b) Quartz crystal microbalance-based biosensor.
c) Magnetoelastic-based biosensor.



Naresh V, Lee N. A Review on Biosensors and Recent Development of Nanostructured Materials-Enabled Biosensors. *Sensors*. Feb 2021;21(4)1109. doi:10.3390/s21041109

a) Enzyme thermistor-based biosensor.
b) Si nanowire-based field-effect transistor (FET) (D is drain and S is the source) biosensor.
c) Surface acoustic wave (SAW)-based biosensor.

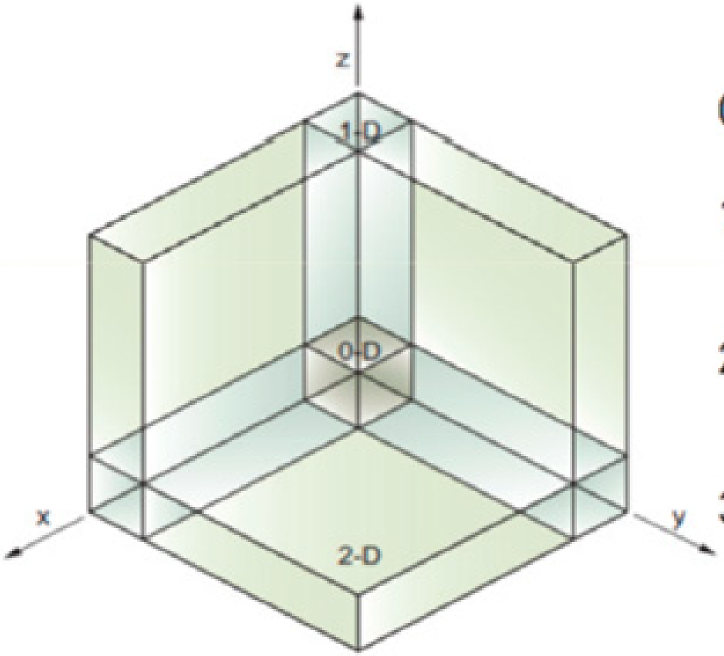





Naresh V, Lee N. A Review on Biosensors and Recent Development of Nanostructured Materials-Enabled Biosensors. *Sensors*. Feb 2021;21(4)1109. doi:10.3390/s21041109

# Nanotechnology

## Classification of Nanomaterial Dimensions

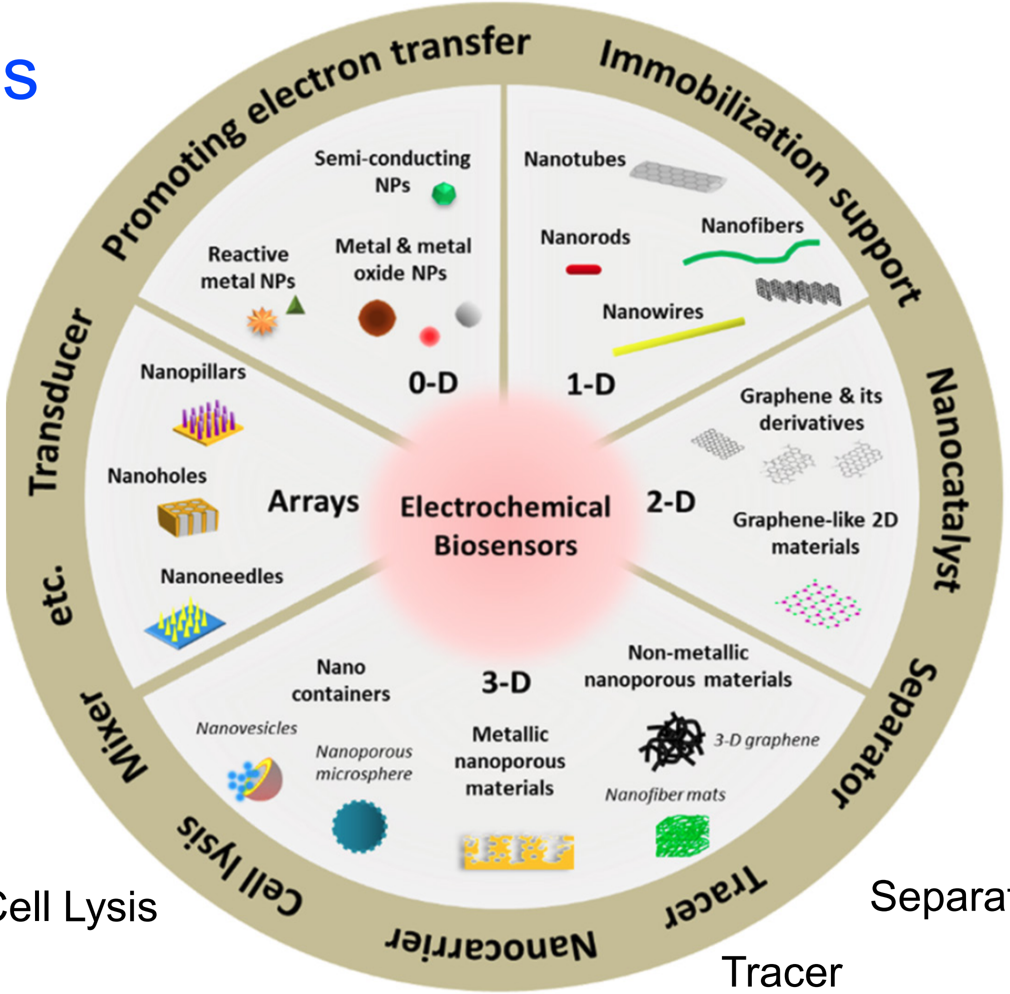



0-D: All dimensions at the nanoscale

1-D: Two dimensions at the nanoscale, one dimension at the macroscale

2-D: One dimension at the nanoscale, two dimensions at the macroscale

3-D: No dimensions at the nanoscale, all dimensions at the macroscale




Mixer

Cell Lysis

Nanocarrier

Tracer

Separator



Schodek, D.; Ferreira, P.; Ashby, M. Nanomaterials, Nano-technologies and Design; Butterworth-Heinemann: 2009.

# “Enzyme-Like” Activity of Nanoparticles…

- Nanomaterials like gold nanoparticles (AuNPs), $Fe_3O_4$, Pd, NiO, $TiO_2$ have *intrinsic enzyme-like activity*.
- Metals and metal oxides are well-known catalysts driving many catalytic reactions.
- They are also widely used in electrochemical biosensors enabling *nonenzymatic detection* of metabolites such as sugars and reactive oxygen species - enabling catalyst-enhanced signal amplification.

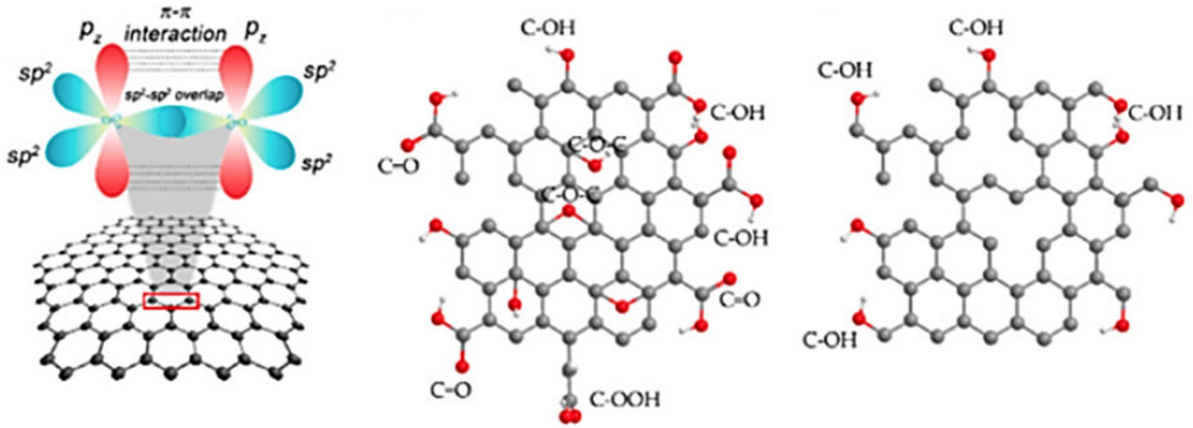
Nanoparticle fluid dispersions in the University of Minnesota Nano-Bio Lab.



Wongkaew N, Simsek M, Griesche C, Baeumner AJ. Functional Nanomaterials and Nanostructures Enhancing Electrochemical Biosensors and Lab-on-a-Chip Performances: Recent Progress, Applications, and Future Perspective. *Chemical Reviews.* 2019;119(1):120-194.

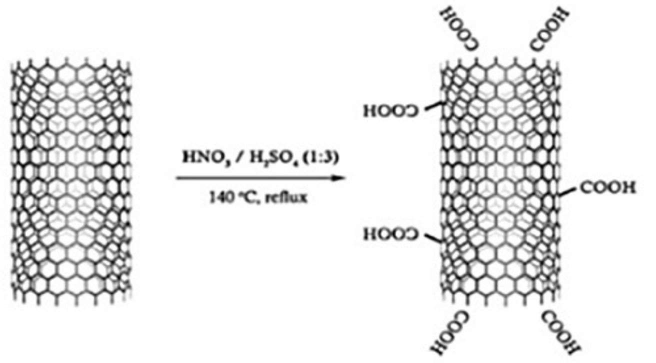
# Carbon Nanomaterials…

- Graphene, carbon nanotubes (CNTs), and carbon nanofibers (CNFs), have been extensively studied and applied in electrochemical sensors.




Graphene    Graphene oxide    Reduced graphene oxide

(1)

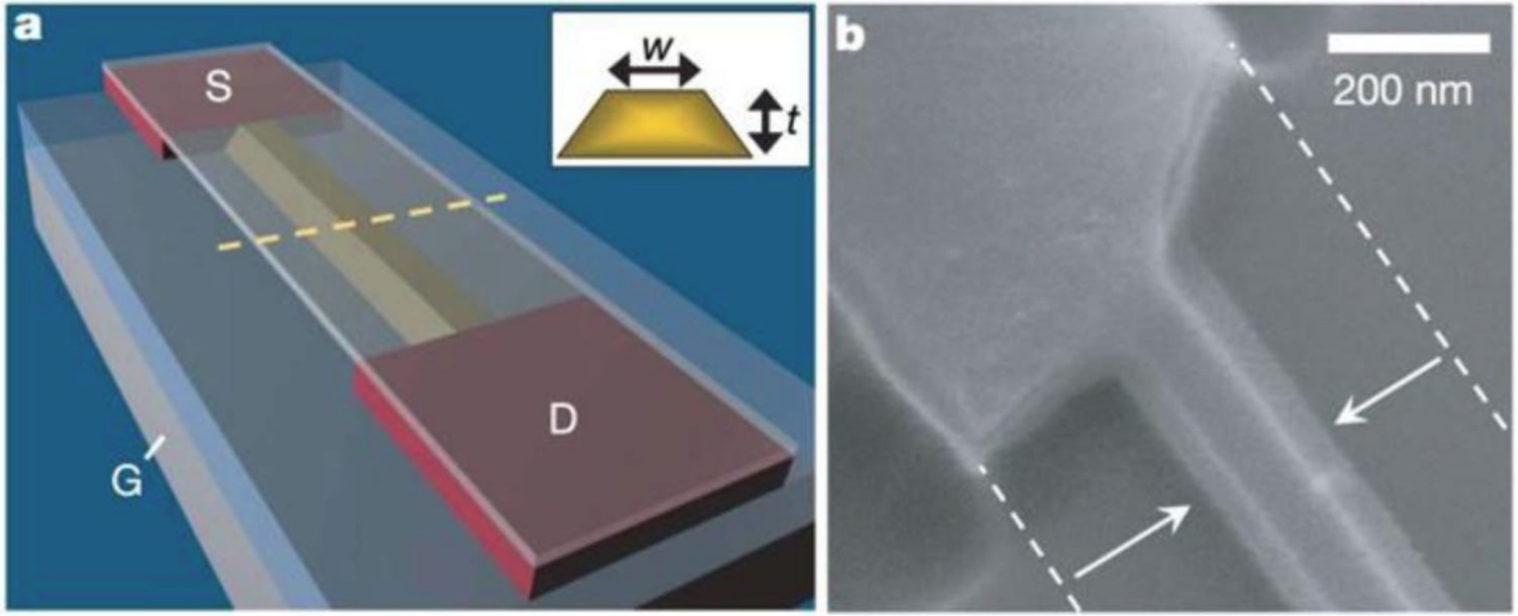



Carbon nanotube    Acid-treated carbon nanotube

(2)

Suvarnaphaet, P.; Pechprasarn, S. Graphene-Based Materials for Biosensors: A Review. Sensors 2017, 17, 2161. Bikiaris, D.; Vassiliou, A.; Chrissafis, K.; Paraskevopoulos, K. M.; Jannakoudakis, A.; Docoslis, A. Effect of Acid Treated Multi-Walled Carbon Nanotubes on the Mechanical, Permeability, Thermal Properties and Thermo-Oxidative Stability of Isotactic Polypropylene. Polym. Degrad. Stab. 2008, 93, 952−967.

# Silicon Nanowire Biosensor…



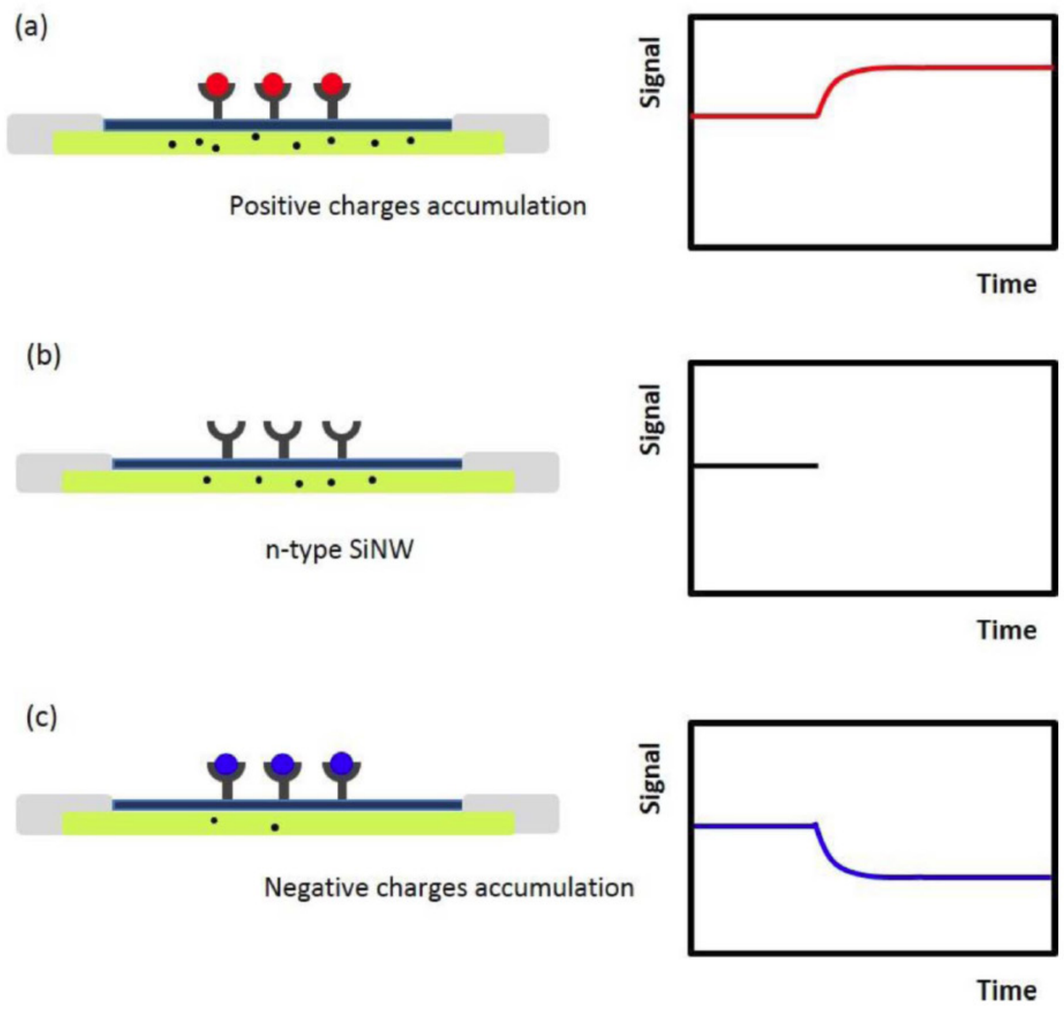

a) An illustration of silicon nanowire; a wire-like channel connects to source and drain electrodes.
b) The SEM image of a silicon nanowire.

Eric Stern, James F. Klemic, David A. Routenberg, Pauline N. Wyrembak, Daniel B. Turner-Evans, Andrew D. Hamilton, David A. LaVan, Tarek M. Fahmy, and Mark A. Reed, Nature, 445, 519 (2007).
Syu YC, Hsu WE, Lin CT. Review-Field-Effect Transistor Biosensing: Devices and Clinical Applications. *Ecs Journal of Solid State Science and Technology.* 2018;7(7):Q3196-Q3207

a) Positive charges accumulate on the surface. The electrostatic attraction force to electron carriers results in higher conductance.

b) The original state of SiNW.

c) Negative charges accumulate on the surface. The electrostatic repulsion force to electron carriers results in lower conductance.



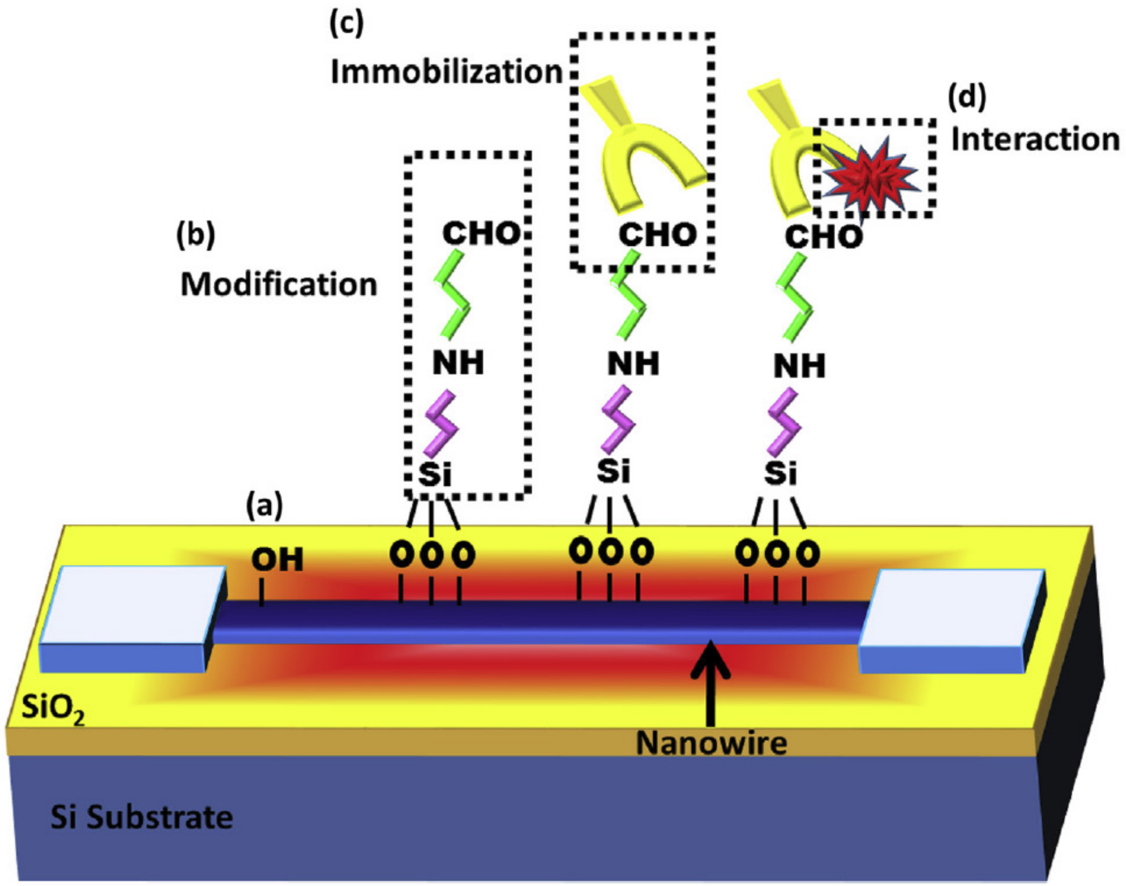
# Biomolecule Detection…

a) Attachment of hydroxide groups on the surface.

b) Surface modification of the first linker from the silane group and the second linker from the carboxyl group.

c) Antibody immobilization.

d) Binding of antigen. Measurements occur between the source and drain.






Adzhri R, Arshad KM, Gopinath SCB, et al. High-performance integrated field-effect transistor-based sensors. *Analytica Chimica Acta.* 2016;917:1-18.

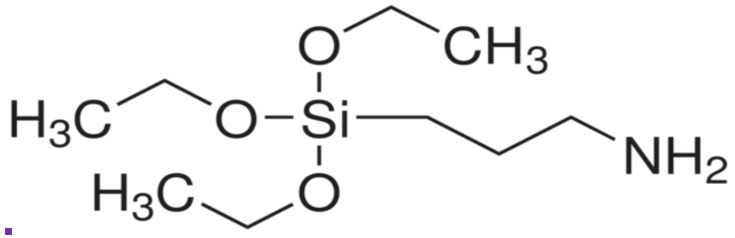
# Immobilization Methods

- The functionalization process required more complex process than immobilizing the probes only.
  - This is because the functional groups of biomolecules need cross-linkers to form covalent bonds with the sensing film (dielectric layer) of FET-based biosensors.
- The most commonly used cross-linking process for ISFET and SiNW (ie. the oxide-based sensing dielectric) is the **APTES-GA** method. (**3-Aminopropyl-tri-ethoxysilane**).



Syu YC, Hsu WE, Lin CT. Review-Field-Effect Transistor Biosensing: Devices and Clinical Applications. *Ecs Journal of Solid State Science and Technology.* 2018;7(7):Q3196-Q3207

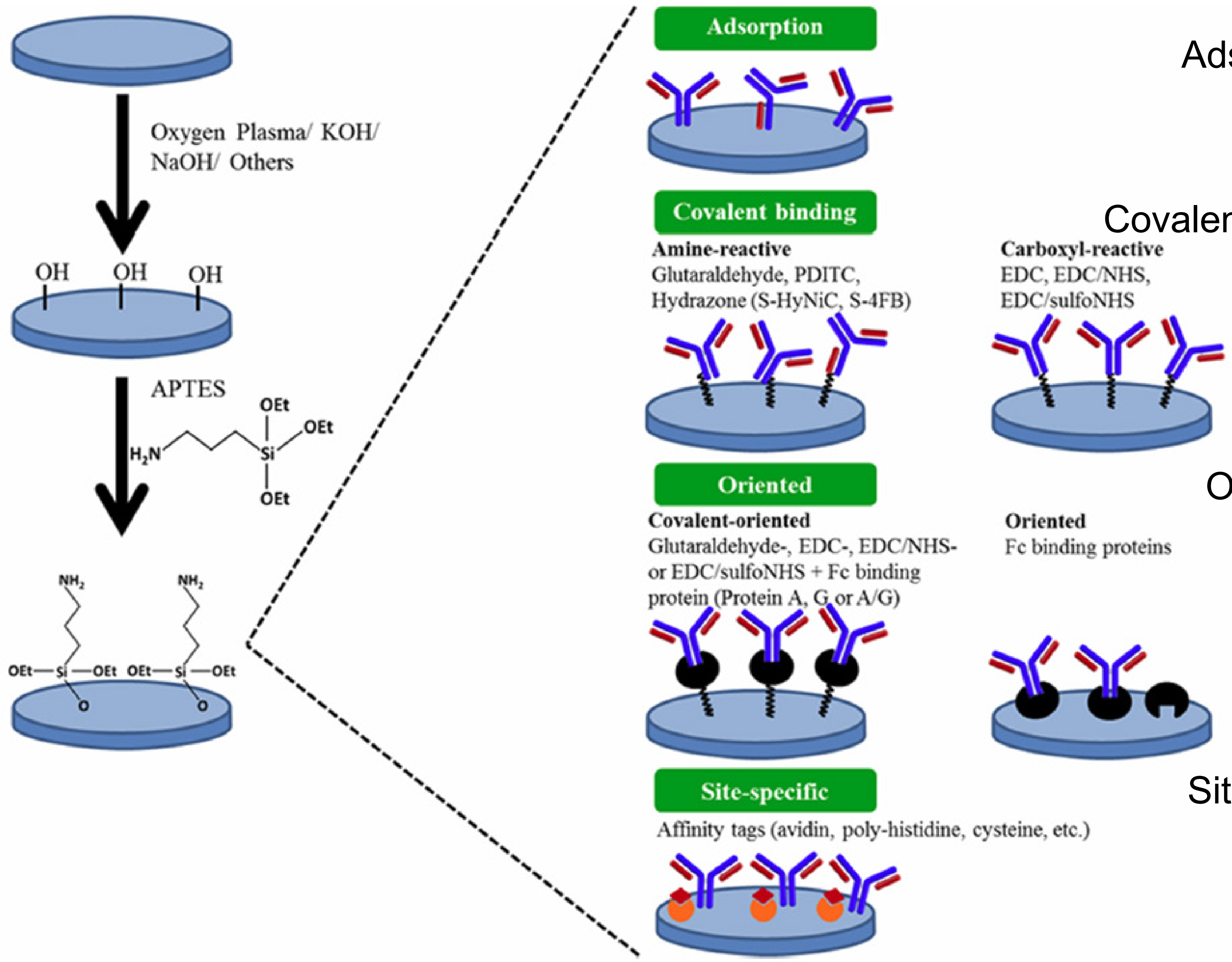
# APTES




- *APTES is a silane molecule*,
  - Bonds with sensor dielectric surface, such as $SiO_2$.
  - It can be deposited on solid materials, electrode materials, nanomaterials, and nanocomposites.
- This self-assembled organic monolayer forms on the sensor surface and provides a good platform for the second stage linkers by its amine group in the other end.
- The second stage cross-linker is *glutaraldehyde*, which is a bifunctional reagent connecting the APTES and bio-probe by imide bonds.

# Antibody Immobilization

Scheme 2. Schematic of Most Commonly Used Ab Immobilization Strategies on APTES-Functionalized Bioanalytical Platforms




Adsorption

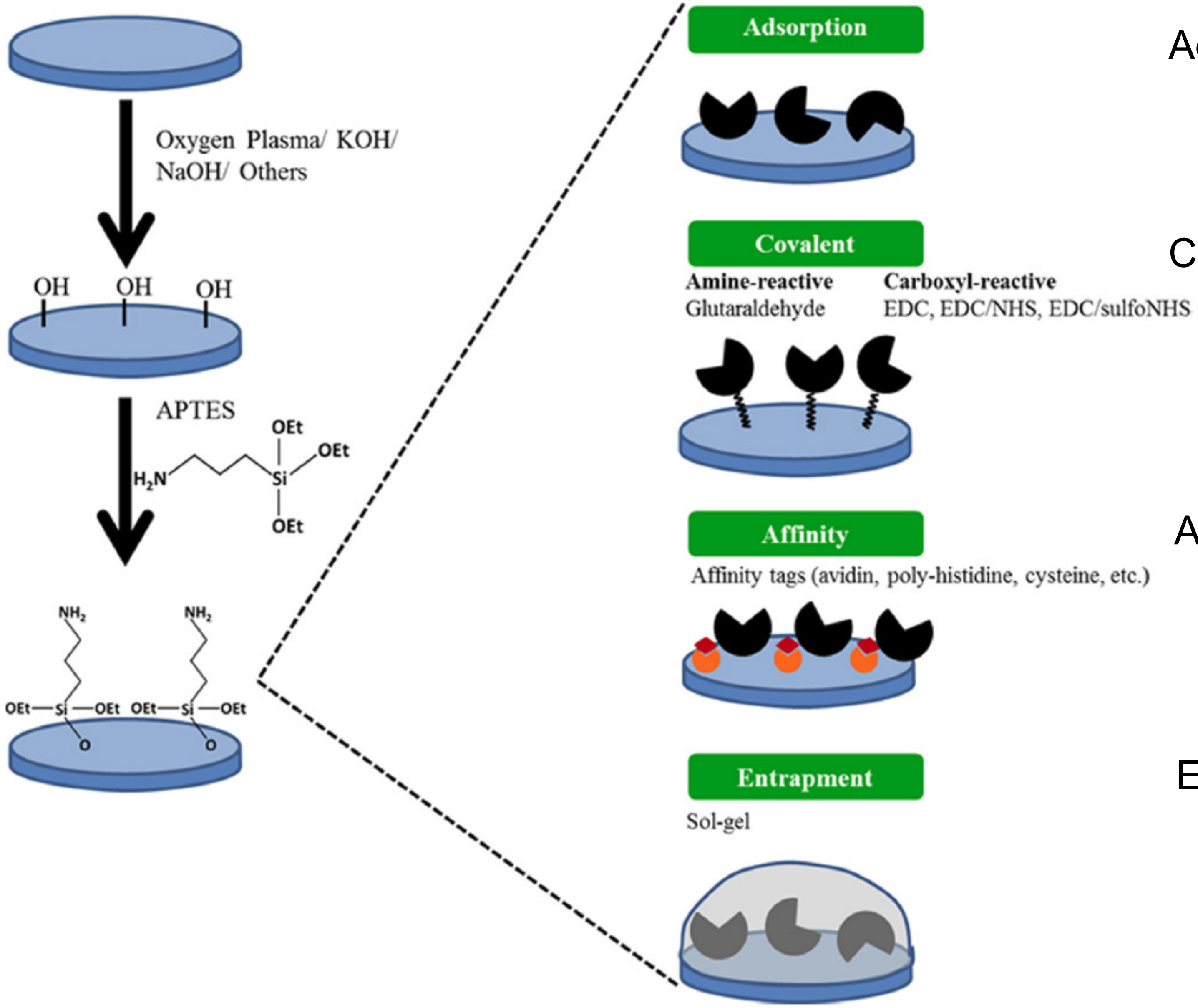
Covalent Bonding

Oriented

Site Specific

Vashist SK, Lam E, Hrapovic S, Male KB, Luong JHT. Immobilization of Antibodies and Enzymes on 3-Aminopropyltriethoxysilane-Functionalized Bioanalytical Platforms for Biosensors and Diagnostics. *Chemical Reviews.* 2014;114(21):11083-11130.

# Enzyme Immobilizatior




Adsorption

Covalent

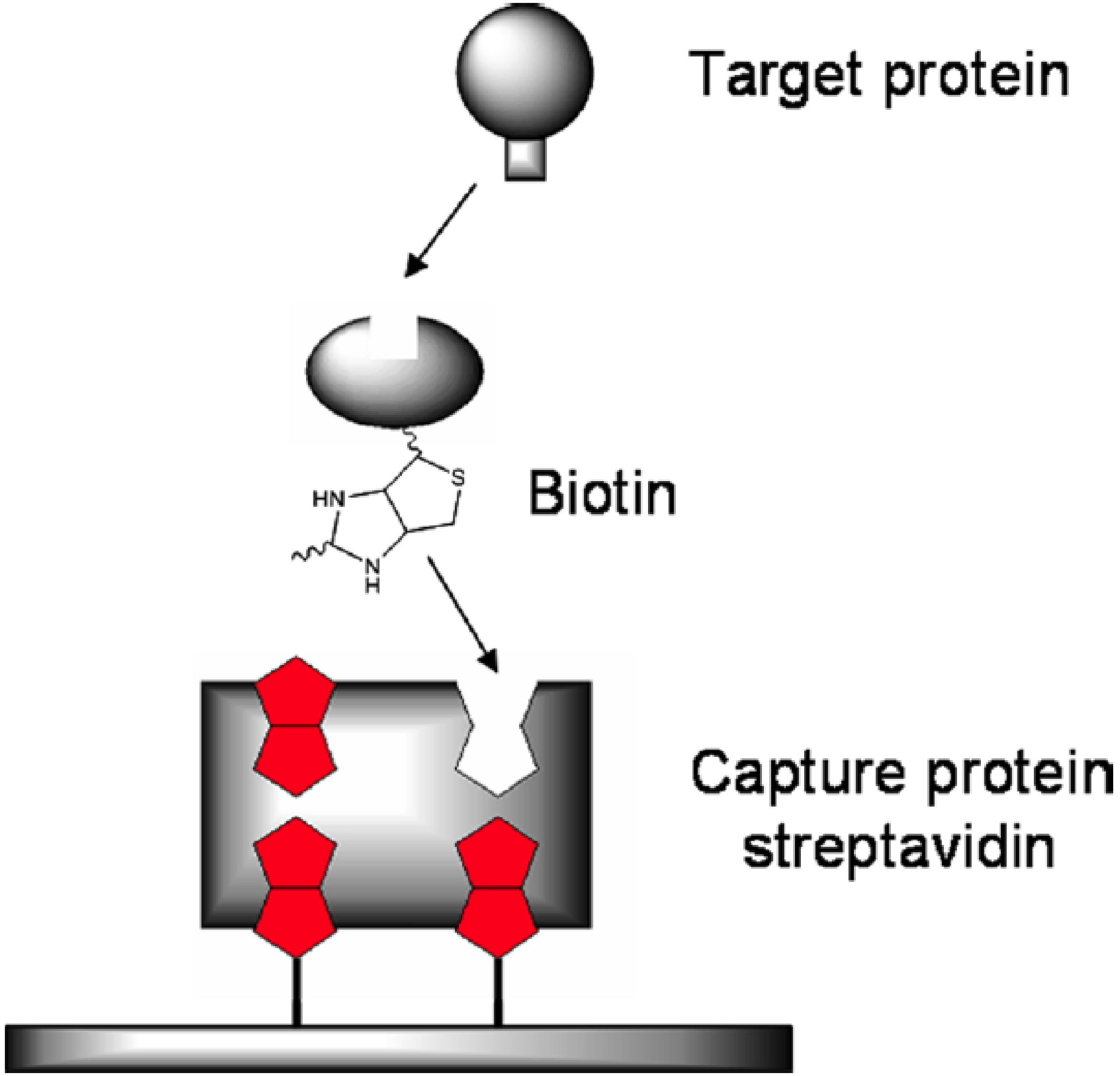
Affinity

Entrapment

Vashist SK, Lam E, Hrapovic S, Male KB, Luong JHT. Immobilization of Antibodies and Enzymes on 3-Aminopropyltriethoxysilane-Functionalized Bioanalytical Platforms for Biosensors and Diagnostics. *Chemical Reviews.* 2014;114(21):11083-11130.

**Scheme 5. Immobilization of a Protein by Affinity through a Streptavidin/Biotin Interaction**

# Protein Immobilization




Vashist SK, Lam E, Hrapovic S, Male KB, Luong JHT. Immobilization of Antibodies and Enzymes on 3-Aminopropyltriethoxysilane-Functionalized Bioanalytical Platforms for Biosensors and Diagnostics. *Chemical Reviews.* 2014;114(21):11083-11130.

# Further Study…

sensors

MDPI

*Review*

## A Review on Biosensors and Recent Development of Nanostructured Materials-Enabled Biosensors

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2021

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Analytica Chimica Acta 917 (2016) 1–18

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Review

# High-performance integrated field-effect transistor-based sensors

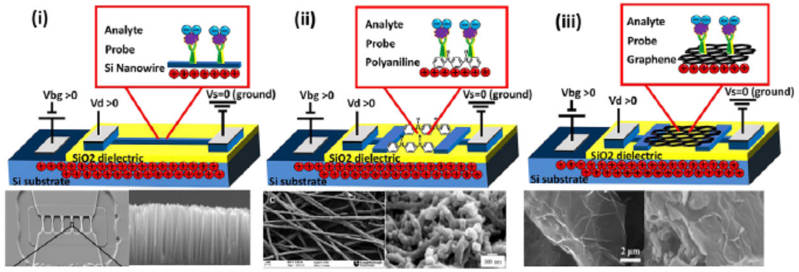
R. Adzhri [a], M.K. Md Arshad [a, b, *], Subash C.B. Gopinath [a, c], A.R. Ruslinda [a], M.F.M. Fathil [a], R.M. Ayub [a], M. Nuzaihan Mohd Nor [a], C.H. Voon [a]

[a] Institute of Nano Electronic Engineering (INEE), Universiti Malaysia Perlis (UniMAP), Kangar, Perlis, Malaysia
[b] School of Microelectronic Engineering (SoME), Universiti Malaysia Perlis (UniMAP), Kangar, Perlis, Malaysia
[c] School of Bioprocess Engineering (SBE), Universiti Malaysia Perlis (UniMAP), Arau, Perlis, Malaysia

## HIGHLIGHTS

- Performance of FET-based biosensors for the detection of biomolecules is presented.
- Silicon nanowire, polyaniline and graphene are the highlighted nano-scaled materials as sensing transducers.
- The importance of surface material interaction with the surrounding environment is discussed.
- Different device structure architectures for ease in fabrication and high sensitivity of sensing are presented.

## GRAPHICAL ABSTRACT




Steven S. Saliterman

CHEMICAL REVIEWS

Review

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# Immobilization of Antibodies and Enzymes on 3-Aminopropyltriethoxysilane-Functionalized Bioanalytical Platforms for Biosensors and Diagnostics

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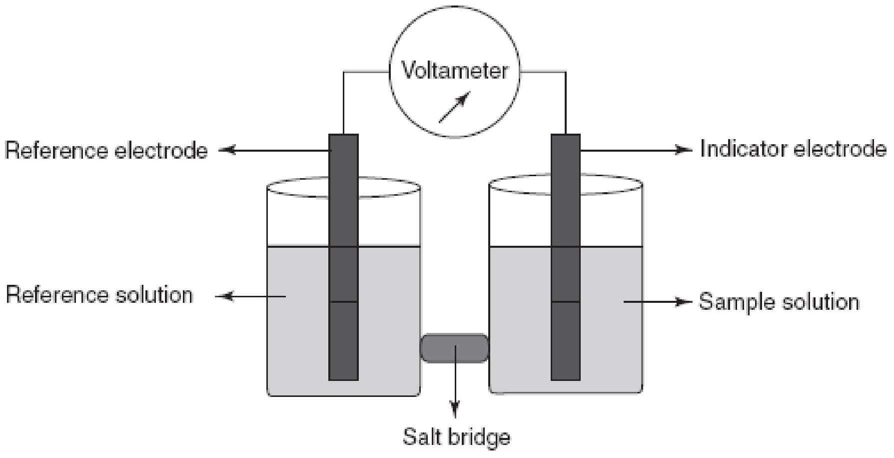
# Summary

- What are biosensors?
- Electrochemical biosensing overview.
  - COVID-19 sensor example.
- Biological recognition elements.
- Transducers
  - Field effect transistors (FETs).
- Example Sensors
- Nanotechnology - nanoparticles
- Immobilization techniques.
  - APTES-GA (3-aminopropyltriethoxysilane) – Enzymes, Ab &proteins

# Electrochemical Detection

## 1) Potentiometry




- A potential difference between two half-cells with negligible current flowing. The cathode is the *indicator* and the anode the *reference electrode.*
- The most prominent potentiometric sensors are *ion-selective electrodes* in which a membrane provides for the ion-selective response.
- In most cases potentiometric sensors are chemosensors; however, when combined with a bioselective separation process they can also be assembled to be full biosensors.
- In FETs the same principle is being applied through the measurement of ions present in the *gate electrode* area of the FET.



Wongkaew N, Simsek M, Griesche C, Baeumner AJ. Functional Nanomaterials and Nanostructures Enhancing Electrochemical Biosensors and Lab-on-a-Chip Performances: Recent Progress, Applications, and Future Perspective. *Chemical Reviews.* 2019;119(1):120-194.

## 2) Voltammetry & Amperometry

- Application of a potential between a working electrode (WE) and reference electrode (RE). A current is flowing and measured between a counter electrode (CE) and the WE as a result of *reduction/oxidation* processes at the surface of the electrodes.
- In its most simple form, a constant potential is applied and current is either measured at a specific time (amperometry) or integrated over a period of time (coulometry).
- Other approaches to improve S/N: differential pulse voltammetry (DPV) and square-wave voltammetry (SWV) or to obtain analytical information on the redox reaction (cyclic voltammetry, CV).



Wongkaew N, Simsek M, Griesche C, Baeumner AJ. Functional Nanomaterials and Nanostructures Enhancing Electrochemical Biosensors and Lab-on-a-Chip Performances: Recent Progress, Applications, and Future Perspective. *Chemical Reviews.* 2019;119(1):120-194.

3) Electrochemical Impedance Spectroscopy (EIS)

- Monitoring the impedance, a frequency dependent resistance, after an electrical stimulation (voltage or current) in the ac mode.

4) Conductometry

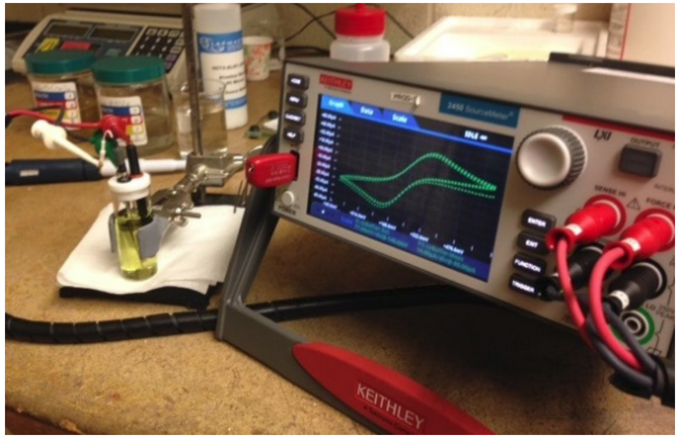
- Conductance is the inverse value of resistance measured in dc mode.
- The resulting sensors are often referred to as chemiresistors and typically serve to measure conductivity changes within the bulk of an electrochemical cell, for gas sensing or enzyme-based strategies.

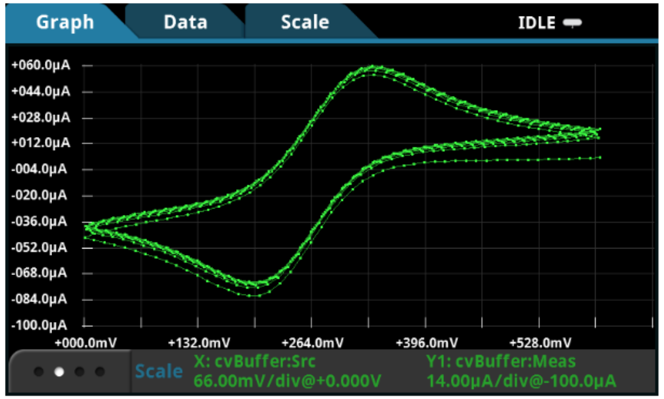


Wongkaew N, Simsek M, Griesche C, Baeumner AJ. Functional Nanomaterials and Nanostructures Enhancing Electrochemical Biosensors and Lab-on-a-Chip Performances: Recent Progress, Applications, and Future Perspective. *Chemical Reviews.* 2019;119(1):120-194.

# Simple Cyclic Voltammetry Setup…

Keithley Model 2450-EC Source Meter (SMU)




Voltammogram from using a cyclic voltammetry *script*. (V vs. I)

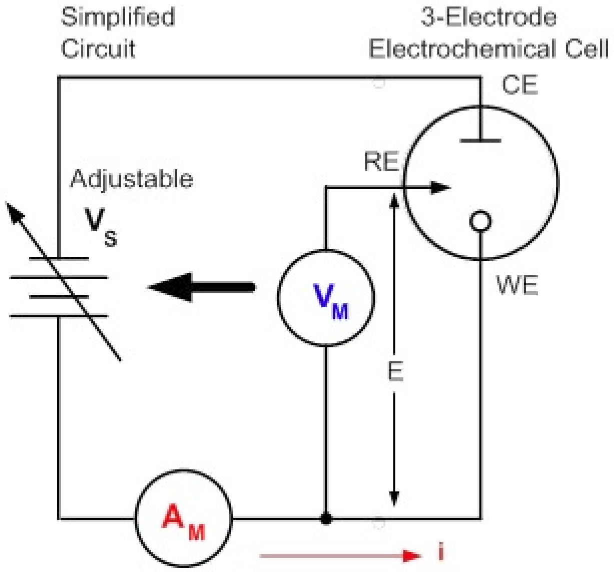



*Figure 1. Simplified measurement circuit for performing cyclic voltammetry*

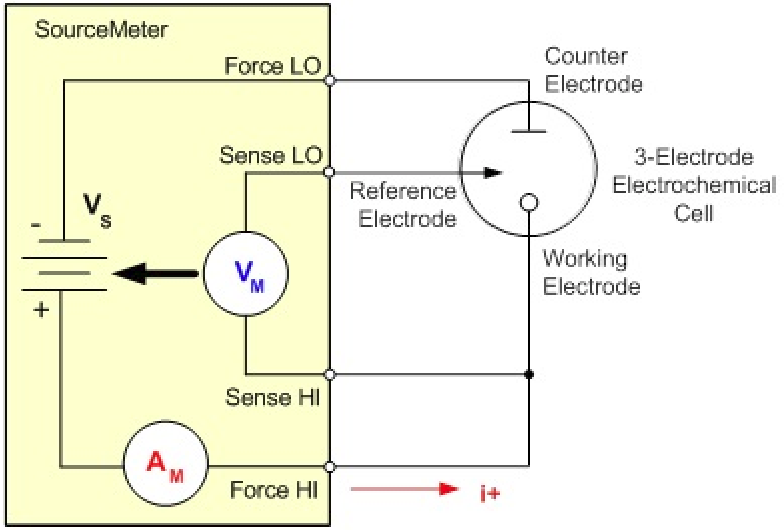


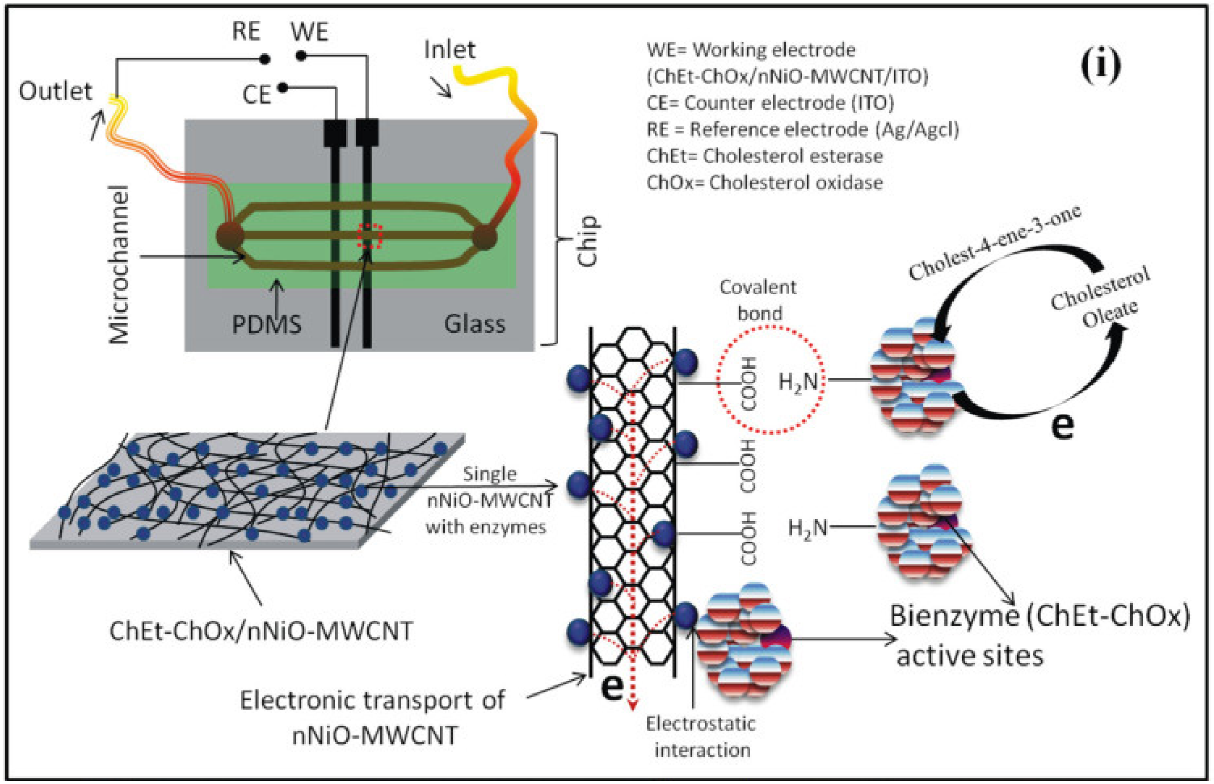

*Figure 2. Connecting a Source Meter to an electrochemical cell for cyclic voltammetry*

When an SMU is programmed to source voltage in the remote sense (4-wire) configuration, internal sensing provides a feedback voltage that is measured and compared to the programmed level. The voltage source is adjusted until the feedback voltage equals the programmed voltage level. Remote sensing compensates for the voltage drop in the test leads and analyte, ensuring that the programmed voltage level is delivered to the working electrode.

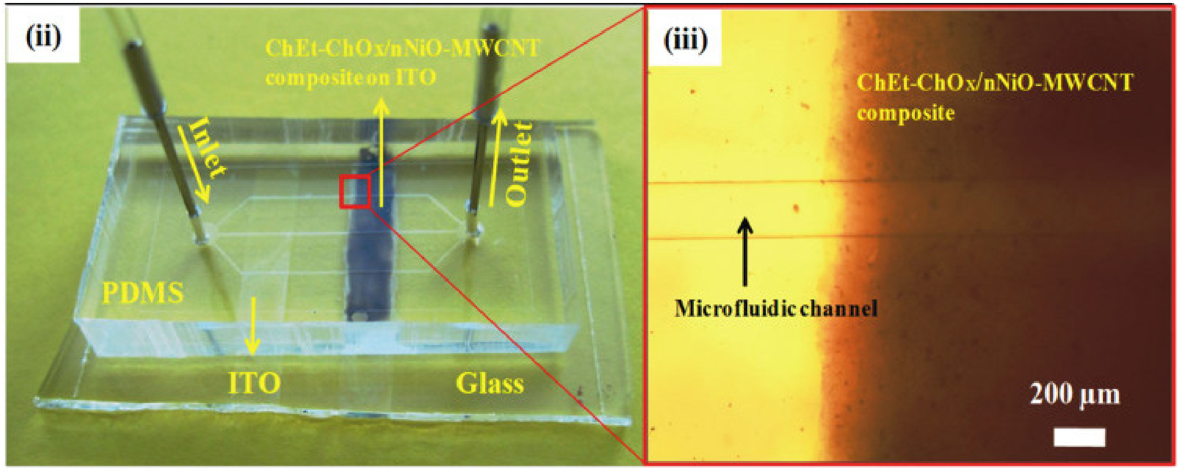


Tupta, MA. *Keithley Product Line at Tektronix,* https://tw.tek.com/blog/performing-cyclic-voltammetry

# Example of Voltammetry Use…




Nickel oxide nanoparticles (nNiO) and multiwalled carbon nanotubes (MWCNT). ITO – Indium Tin Oxide WE and RE.



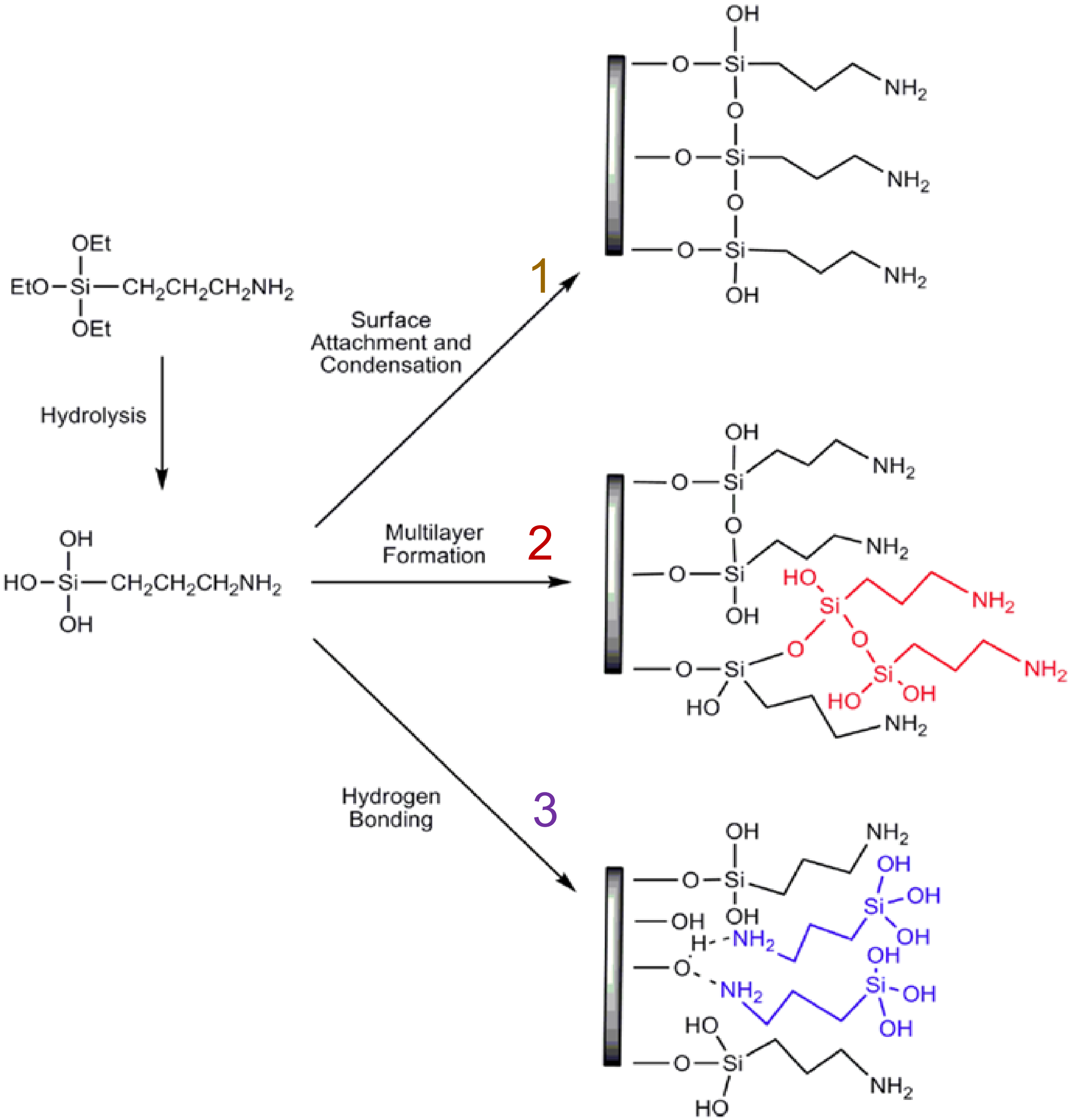

Bi-enzyme (cholesterol oxidase (ChOx) and cholesterol esterase (ChEt) functionalized nanocomposite microfluidic-based biosensor developed for the detection of cholesterol .



Ali, M.A.; Srivastava, S.; Solanki, P.R.; Reddy, V.; Agrawal, V.V.; Kim, C.; John, R.; Malhotra, B.D. Highly efficient bienzyme functionalized nanocomposite-based microfluidics biosensor platform for biomedical application. Sci. Rep. 2013, 3, 2661.

## Three Bonding Modes for APTES on a Substrate Surface:

1. Surface attachment and condensation lead to horizontal polymerization when a surface-bound APTES molecule forms siloxane with its neighboring surface-bound APTES.
2. In vertical polymerization, the surface-bound APTES reacts with a nearby APTES in solution.
3. Where the substrate possesses surface hydroxyl groups, the amine of APTES forms a hydrogen bond with the metal surface or becomes protonated by abstracting protons from the surface.




Vashist SK, Lam E, Hrapovic S, Male KB, Luong JHT. Immobilization of Antibodies and Enzymes on 3-Aminopropyltriethoxysilane-Functionalized Bioanalytical Platforms for Biosensors and Diagnostics. *Chemical Reviews.* 2014;114(21):11083-11130.

# Pre-Treatment with KOH and $0_2$ Plasma…

Silanization occurs on any substrate, normally with chemically active hydroxyl groups for silane grafting.

Some substrates (e.g., silica, agarose, etc.) already have hydroxyl groups, while others require a pretreatment step using KOH/NaOH, acid, piranha solution, or plasma treatment to introduce hydroxyl groups.

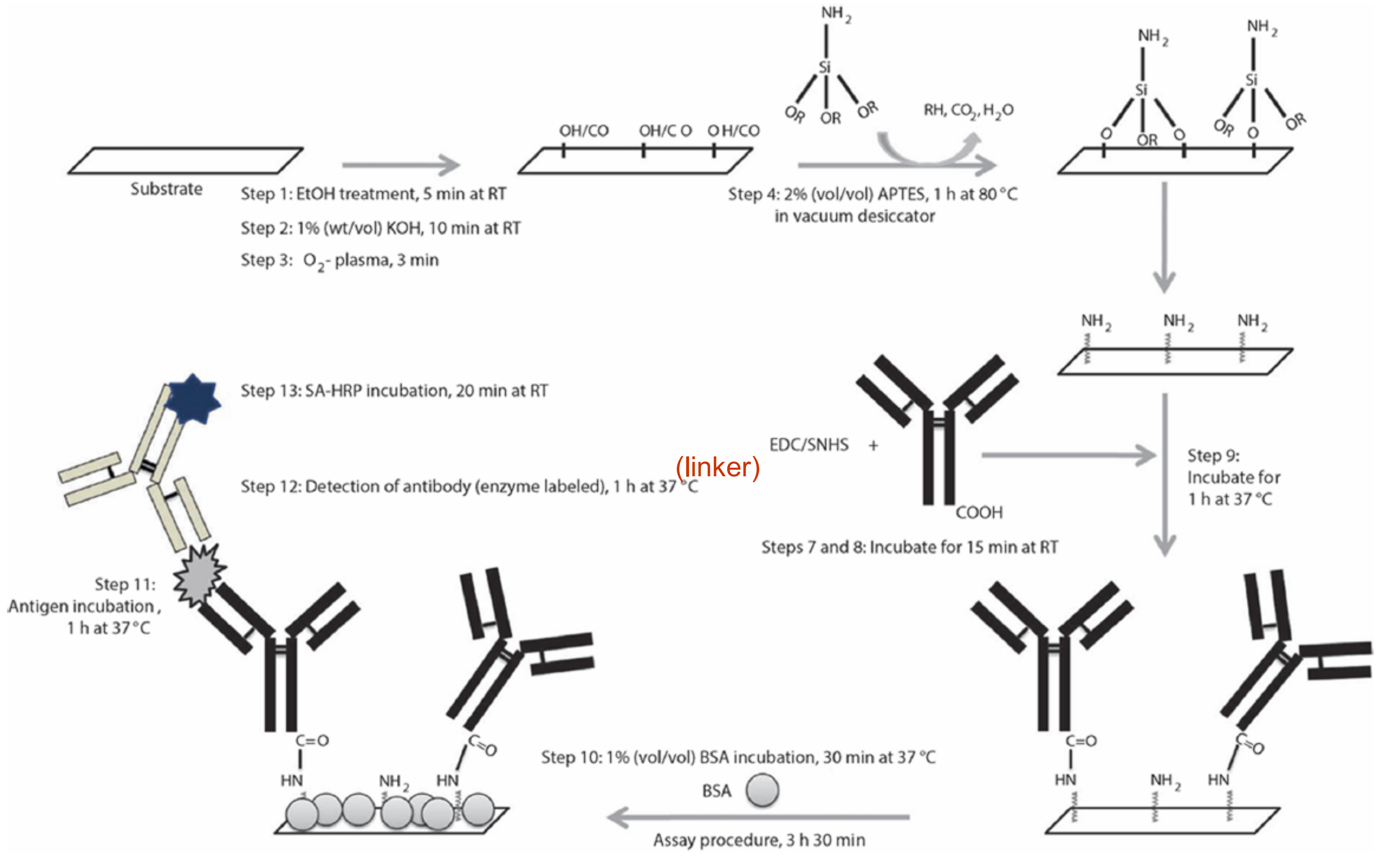



Figure 1. Treatment of a substrate with KOH and $O_2$ plasma prior to APTES functionalization as part of an APTES-based sandwich ELISA procedure. Reproduced with permission from ref 60. Copyright 2011 Nature America Inc.



Dixit, C. K.; Vashist, S. K.; MacCraith, B. D.; O'Kennedy, R. Nat. Protoc. 2011, 6, 439.

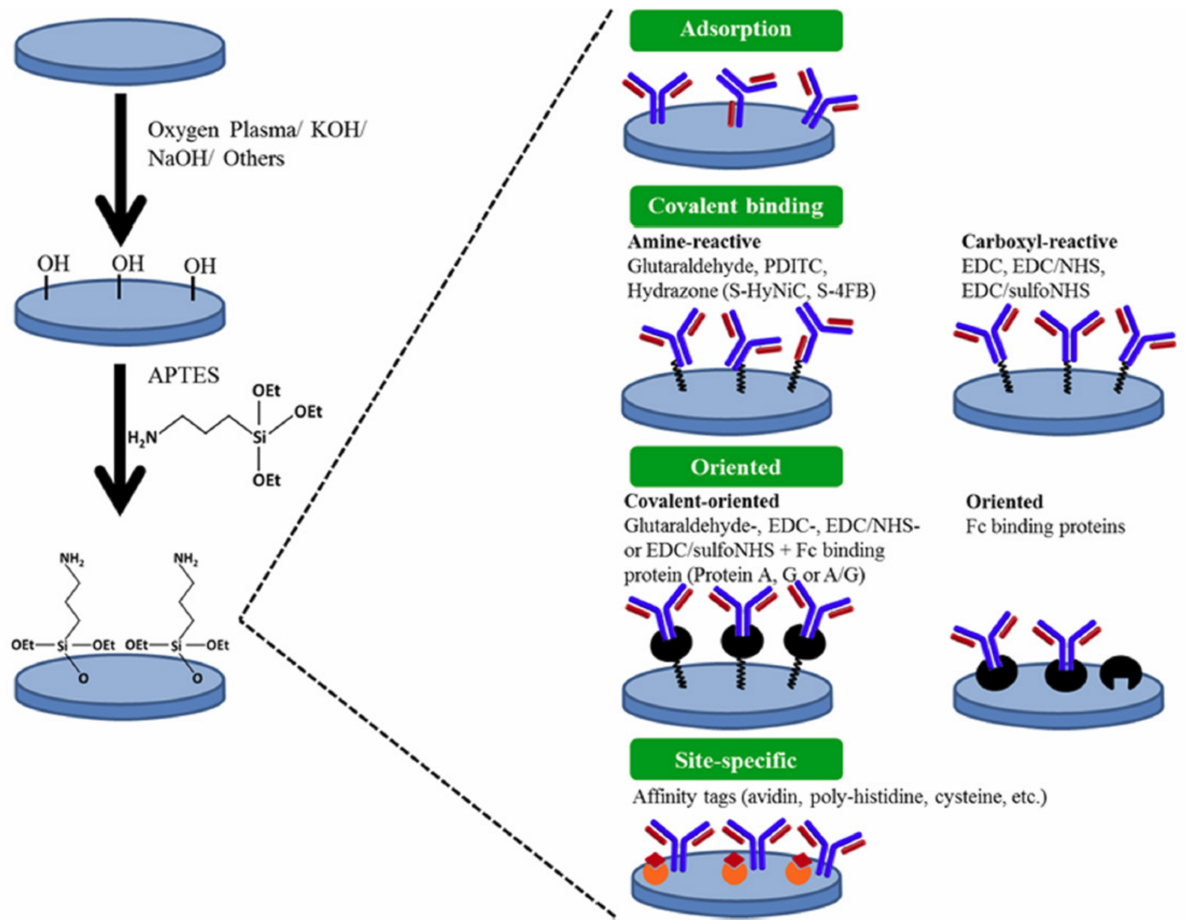
# Antibody Immobilization…

- The amine group of APTES enables the covalent bonding of biomolecules based on the use of a functional linker. GLD (glutaraldehyde), a *homo-bifunctional agent* (same reactive groups on each end of the crosslinker), cross-links the amino groups of APTES-functionalized surfaces to the biomolecule amino groups.

- Aldehyde groups form imine bonds with amine groups of lysine common to almost every protein or enzyme to form reversible Schiff bases.

- The most widely used *hetero-bifunctional* cross-linker combination is EDC and N-hydroxysuccinimide (NHS)/N-hydroxysulfosuccinimide (SNHS), where EDC of the EDC-NHS/SNHS complex first binds to the carboxyl-terminal of the Abs.

Scheme 2. Schematic of Most Commonly Used Ab Immobilization Strategies on APTES-Functionalized Bioanalytical Platforms



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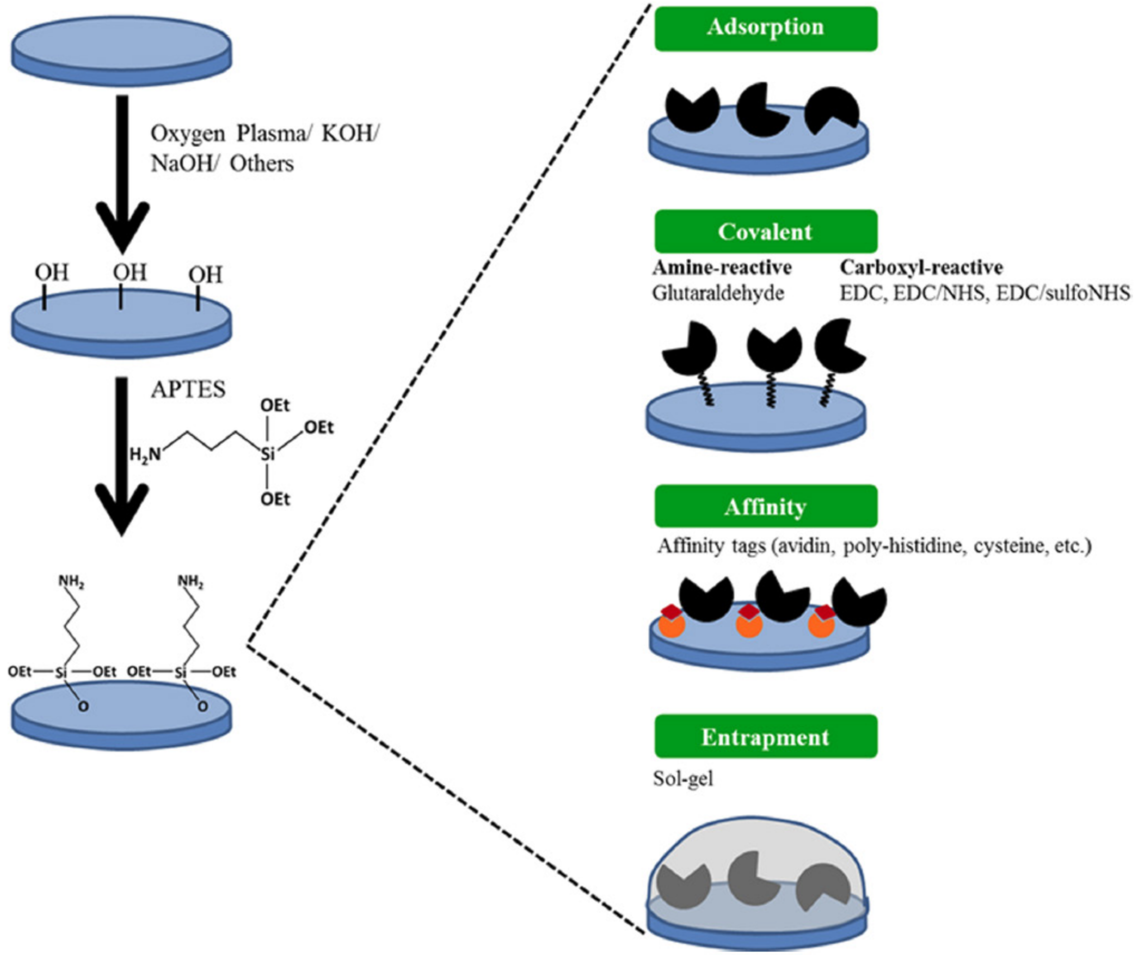
# Example: Enzyme Immobilization…

Most commonly used enzyme immobilization strategies on APTEs - functionalized bioanalytical platforms.

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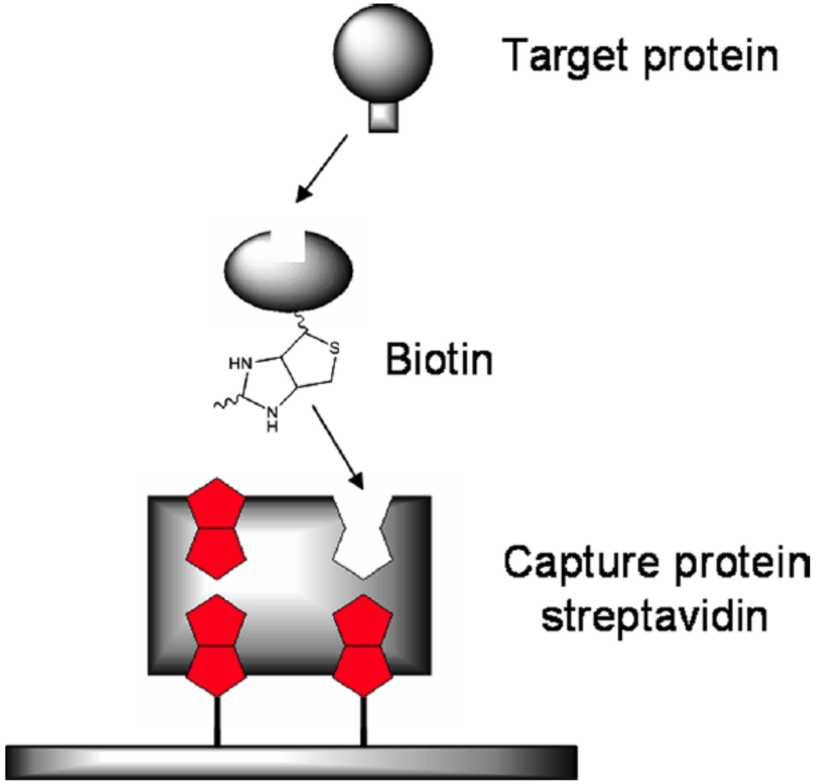
# Protein Immobilization with Biotin/Avidin…

- The specific molecular interaction between the avidin and biotin pair is a well-known phenomenon and has been extensively advocated for biomolecule immobilization as the cubic shape avidin possesses four biotin binding sites.
- Conceptually, avidin is covalently attached to APTES-surfaces by either GLD-mediated or EDC-mediated reactions as previously described.
- Avidin then acts as a biocompatible linker between APTES-surfaces and biotinylated biomolecules.

**Scheme 5. Immobilization of a Protein by Affinity through a Streptavidin/Biotin Interaction**

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