BP 605 T. Pharmaceutical Biotechnology (Theory)

Biosensors - Working and Applications of Biosensors in Pharmaceutical Industries

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Overview

- Definition and Introduction
- Biosensors Working and Applications
- Biosensors in Pharmaceutical Industries

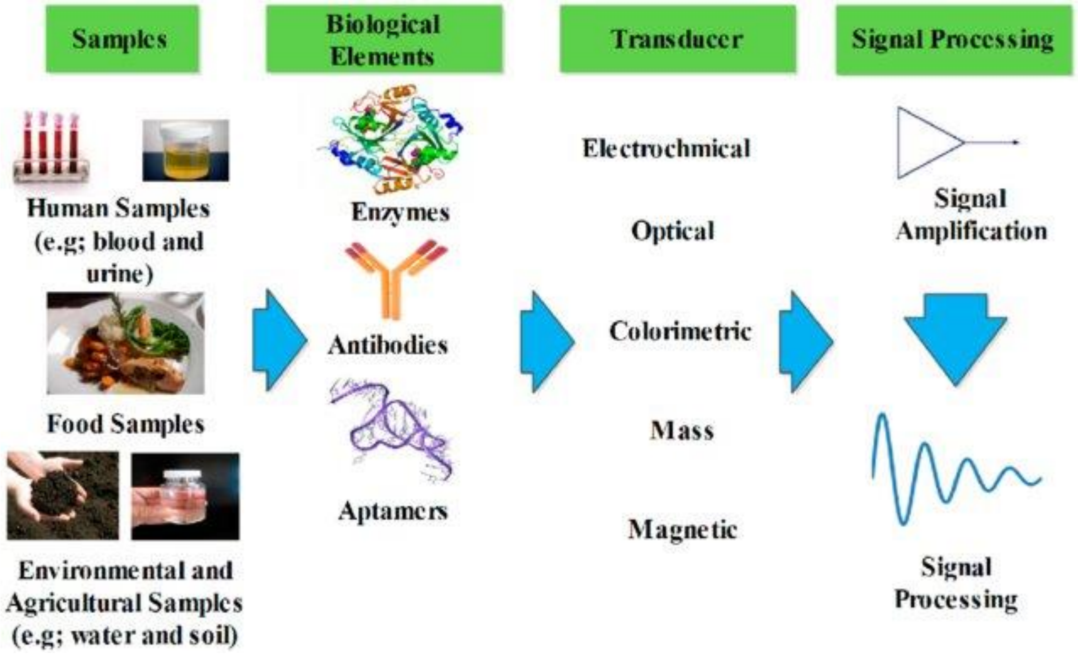
BIOSENSORS

Definition: The term Biosensors is used in the literature in many ways

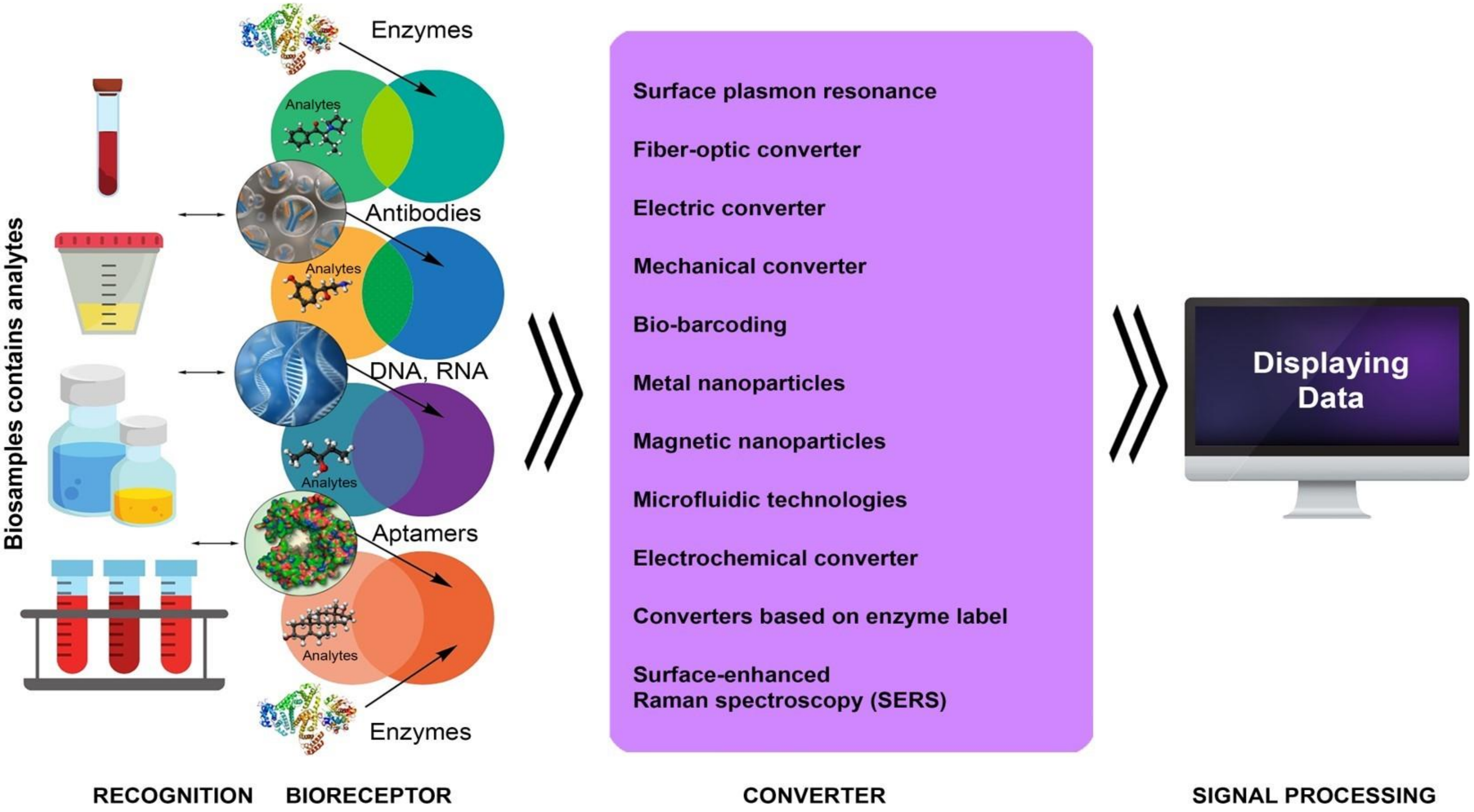
- ✓ A device used to measure biologically-derived signals.
- ✓ A device that "senses" using "biomimetic" (imitative of life) strategies.

  ex.,"artificial nose"
- ✓ A device that detects the presence of biomolecules.
- ✓ Recent IUPAC definition:

"A self-contained integrated device which is capable of providing specific quantitative or semi-quantitative analytical information using a biological recognition element which is in direct spatial contact with a transducer element."



BIOSENSORS


Enzymes
Analytes
Antibodies
Analytes
DNA, RNA
Analytes
Aptamers
Analytes
Enzymes
Biosamples contains analytes
Surface plasmon resonance
Fiber-optic converter
Electric converter
Mechanical converter
Bio-barcoding
Metal nanoparticles
Magnetic nanoparticles
Microfluidic technologies
Electrochemical converter
Converters based on enzyme label
Surface-enhanced
Raman spectroscopy (SERS)
Displaying Data
RECOGNITION
BIORECEPTOR
CONVERTER
SIGNAL PROCESSING

BIOSENSORS

Uses of biosensors

- Quality assurance in agriculture, food and pharmaceutical industries. ex. *E. Coli, Salmonella*
- Monitoring environmental pollutants & biological warfare agents. ex., *Bacillus anthracis* (anthrax) spores
- Medical diagnostics. ex., glucose
- Biological assays. ex., DNA microarrays

CLASSES OF BIOSENSORS

A) Catalytic biosensors: kinetic devices that measure steady-state concentration of a transducer-detectable species formed/lost due to a bio-catalytic reaction

Monitored quantities:

i) rate of product formation

ii) disappearance of a reactant

iii) inhibition of a reaction

Biocatalysts used:

i) Enzymes

ii) Microorganisms

iii) Organelles

iv) Tissue

v) Samples

CLASSES OF BIOSENSORS

B) Affinity biosensors: devices in which receptor molecules bind analyte molecules "irreversibly", causing a physicochemical change that is detected by a transducer

Receptor molecules:

i) Antibodies

ii) nucleic acids

iii) hormone receptors

- ✓ Biosensors are most often used to detect molecules of biological origin, based on specific interactions.

COMPONENTS OF BIOSENSORS

Analyte: chemical/biological target

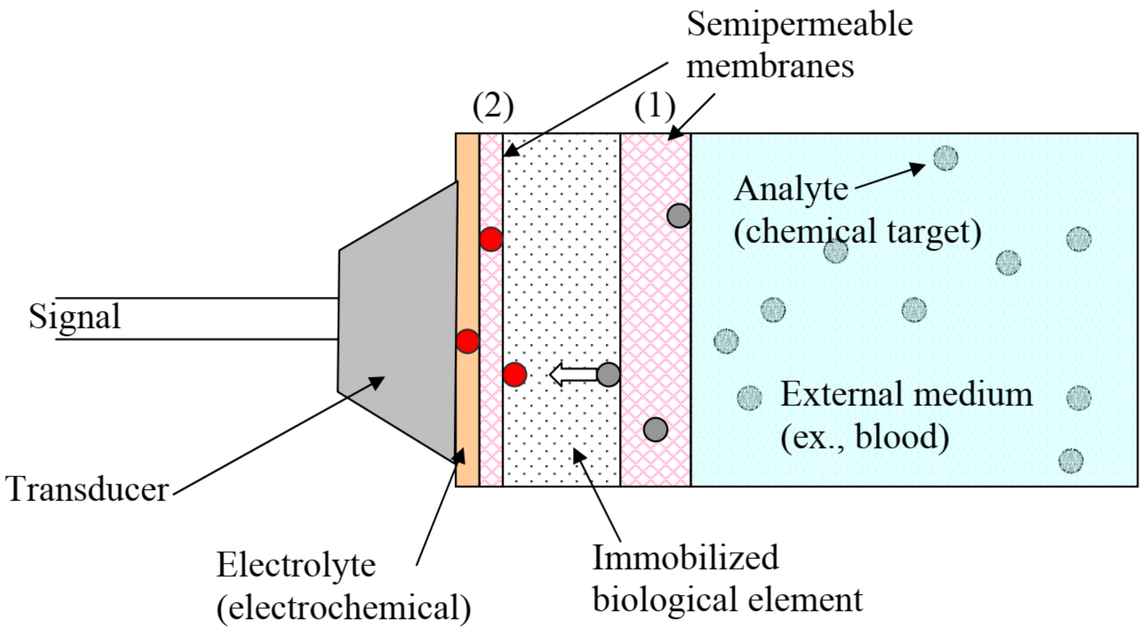
Semipermeable Membrane (1): allows preferential passage of analyte (limits fouling).

Detection Element (Biological): provides specific recognition/detection of analyte.

Semipermeable Membrane (2): (some designs) preferential passage of by product of recognition event

Electrolyte: (electrochemical-based) ion conduction medium between electrodes

Transducer: converts detection event into a measurable signal



BIOSENSORS

A successful biosensor must possess at least some of the following beneficial features:

- The biocatalyst must be highly specific for the purpose of the analyses, be stable under normal storage conditions and, except in the case of colorimetric enzyme strips
- The reaction should be as independent of such physical parameters as stirring, pH and temperature as is manageable. This would allow the analysis of samples with minimal pre-treatment.
- The response should be accurate, precise, reproducible and linear over the useful analytical range, without dilution or concentration. It should also be free from electrical noise.

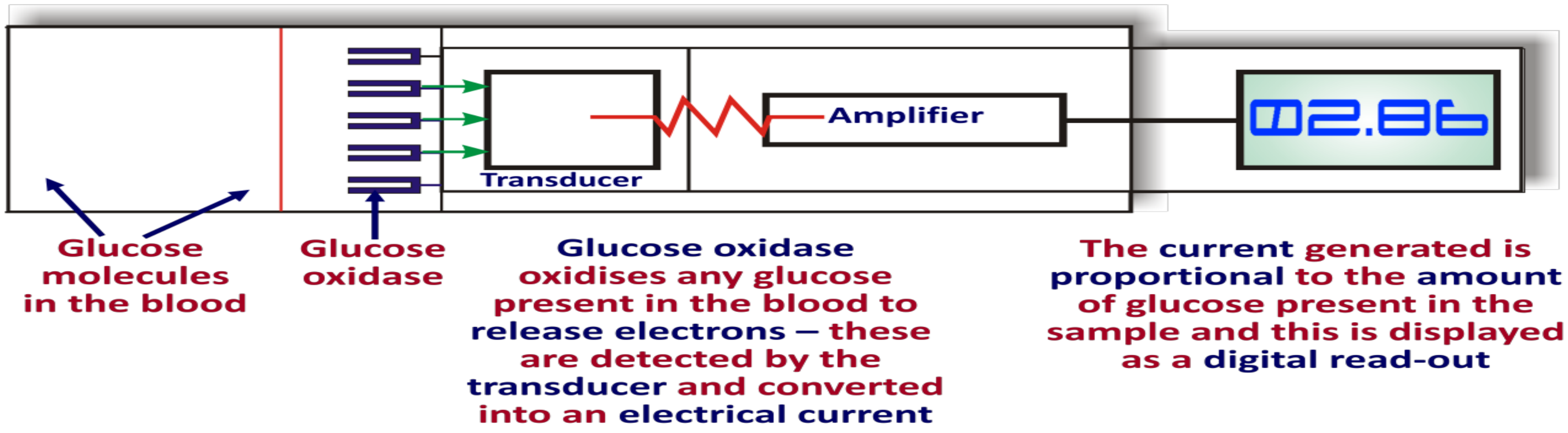
BIOSENSORS

- If the biosensor is to be used for invasive monitoring in clinical situations, the probe must be tiny and biocompatible, having no toxic or antigenic effects. If it is to be used in fermenters it should be serializable. This is preferably performed by autoclaving but no biosensor enzymes can presently withstand such drastic wet-heat treatment. In either case, the biosensor should not be prone to fouling or proteolysis.
- The complete biosensor should be cheap, small, portable and capable of being used by semi-skilled operators.

GLUCOMETER

- Current glucometers use test strips containing glucose oxidase, an enzyme that reacts to glucose in the blood droplet,
- When the strip is inserted into the meter, the flux of the glucose reaction generates an electrical signal
- The glucometer is calibrated so the number appearing in its digital readout corresponds to the strength of the electrical current

A biosensor has been developed for detecting glucose in the blood of diabetics



BIO-ELEMENT

- It is a typically complex chemical system usually extracted or derived directly from a biological organism.
- **Types**
- Enzymes
- Oxidase
- Polysaccharide
- Antibiotics
- Tissue
- Nucleic acid

Conti…

- **Function**
- To interact specifically with a target compound i.e compound to be detected.
- It must be capable of detecting the presence of a target compound in the test solution.
- The ability of a bio-element to interact specifically with the target compound (specifically) is the basis for biosensor.

TRANDUCER

- **Function**
- To convert biological response in to an electrical signal.
- **Types**
- Electrochemical
- Optical
- Piezoelectric

RESPONSE FROM BIO-ELEMENT

- Heat absorbed (or liberated) during the interaction.
- Movement of electrons produced in a redox reaction.
- Light absorbed (or liberated) during the interaction.
- Effect due to mass of reactants or products.

TYPES OF BIOSENSORS

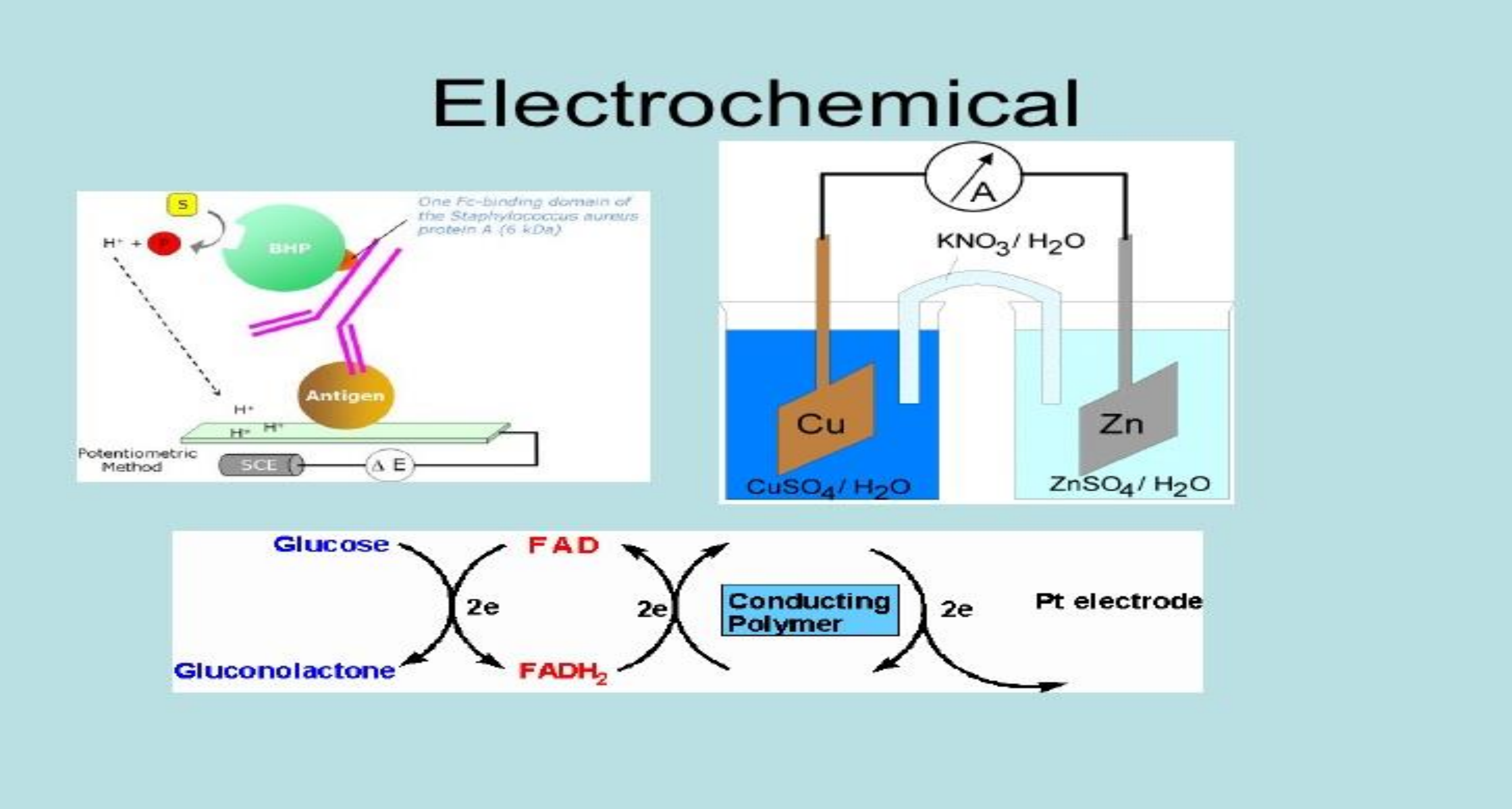
- Electrochemical biosensor
- Optical biosensor
- Thermal biosensor
- Resonant biosensor
- Ion-sensitive biosensor

ELECTROCHEMICAL BIOSENSOR

- **Principle:**
- Many chemical reactions produce or consume ions or electrons which in turn cause some change in the electrical properties of the solution which can be sensed out and used as measuring parameter.
- **Classification**:

1. Amperometric Biosensors
2. Conductimetric Biosensors
3. Potentiometric Biosensors

Electrochemical biosensor



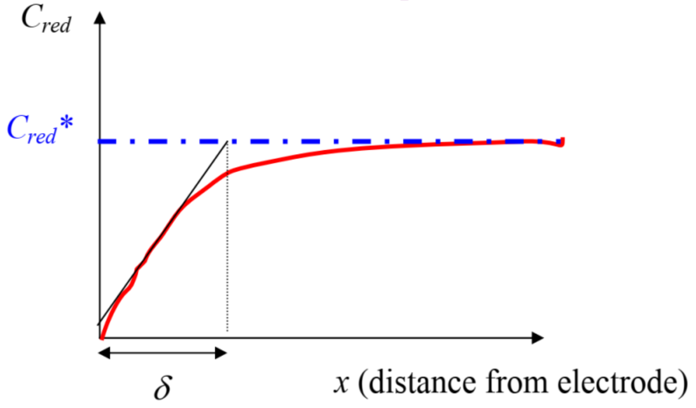
AMPEROMETRIC BIOSENSORS

- The high sensitivity biosensor can detect electro-active species present in biological test samples.
- Since the biological test samples may not be intrinsically electro-active, enzymes are needed to catalyze the production of radio-active species.
- In this case, the measured parameters is current.
- Translate a chemical event to an electrical event by measuring current passed (amperometric = most common), potential change between electrodes, etc. Oxidation reaction of the reduced chemical species

$$C_{red}: C_{red} \rightarrow C_{ox} + ne^{-}$$

Amperometric Devices

Measured current is mass transport limited



$$i = i_{\lim} = -n\Im AJ$$

where J is the flux:

$$J = -D\frac{dC_{red}}{dx} \approx -D\frac{C^*_{red} - 0}{\delta} \Rightarrow i \approx \frac{n\Im ADC^*_{red}}{\delta}$$

$\Im$ = 96,487 coulombs (Faraday const.)

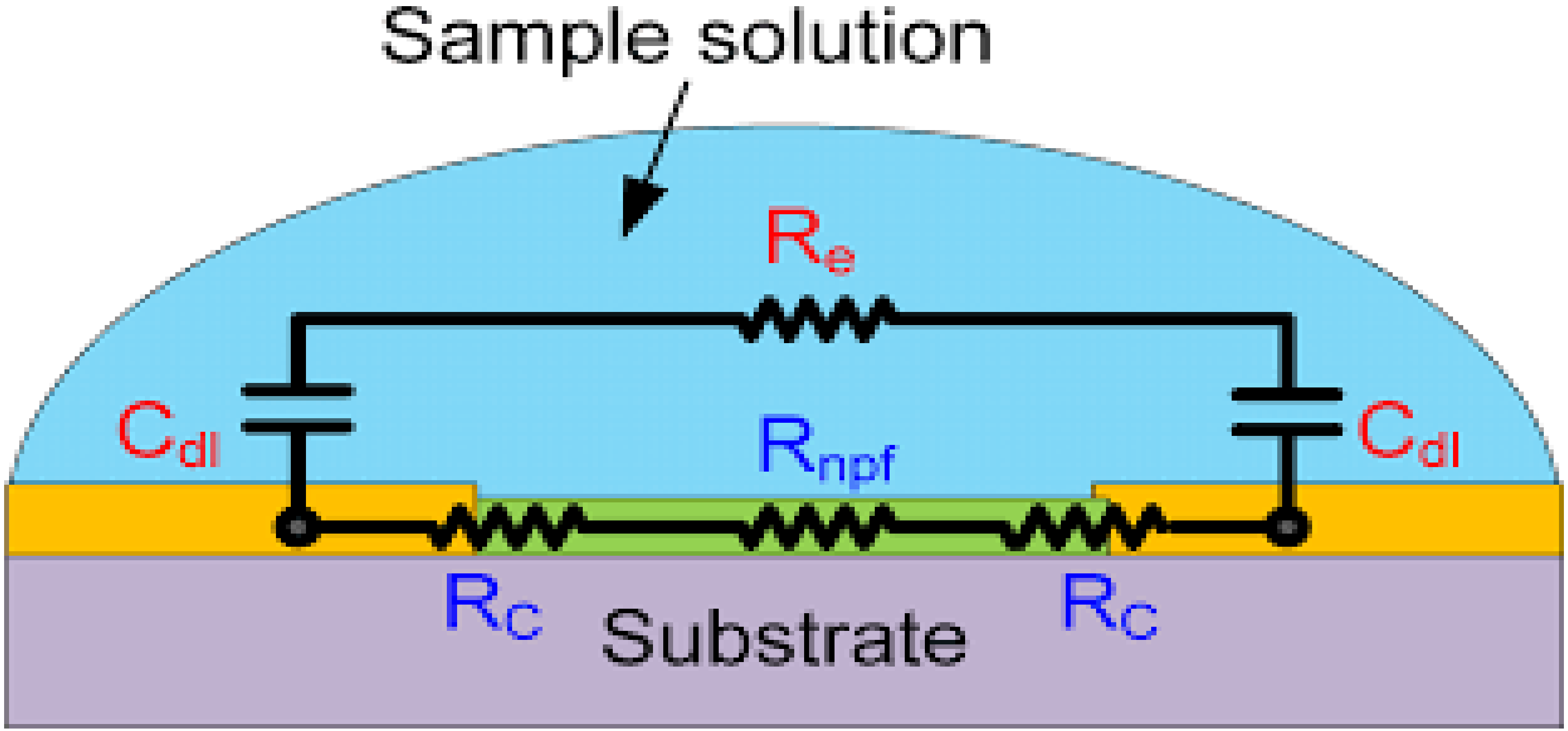
A = electrode area

δ = boundary layer width

CONDUCTIMETRIC BIOSENSORS

- The measured parameter is the electrical conductance resistance of the solution.
- When electrochemical reactions produce ions or electrons, the overall conductivity or resistivity of the solution changes. This change is measured and calibrated to a proper scale (Conductance measurements have relatively low sensitivity).
- The electric field is generated using a sinusoidal voltage (AC) which in minimizing undesirable effects such as Faradaic process, double layer charging and concentration polarization.

CONDUCTIMETRIC BIOSENSOR



POTENTIOMETRIC BIOSENSORS

- In this type of sensor the measured parameter is oxidation or reduction potential of an electrochemical reaction.
- The working principle relies on the fact that where a ramp voltage is applied to an electrode in solution, a current flow occurs because of electrochemical reactions.
- The voltage at which these reaction occurs indicate a particular reaction and particular species.

POTENTIOMETRIC BIOSENSOR

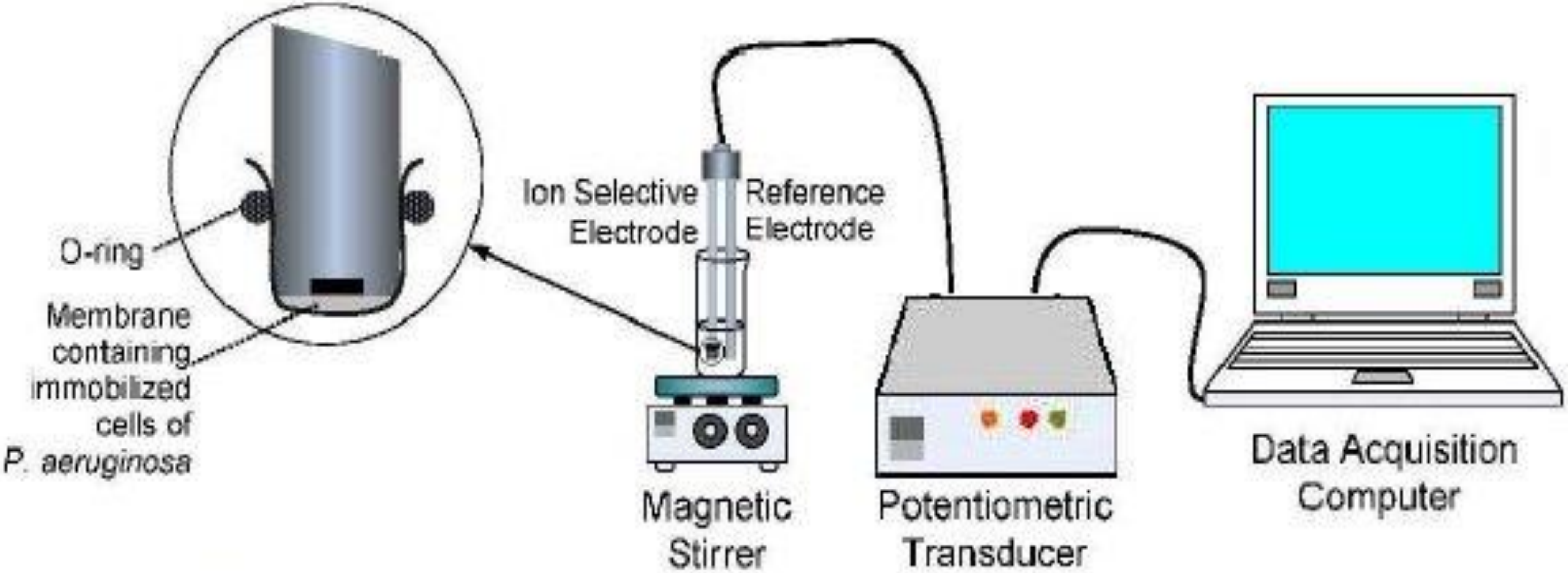
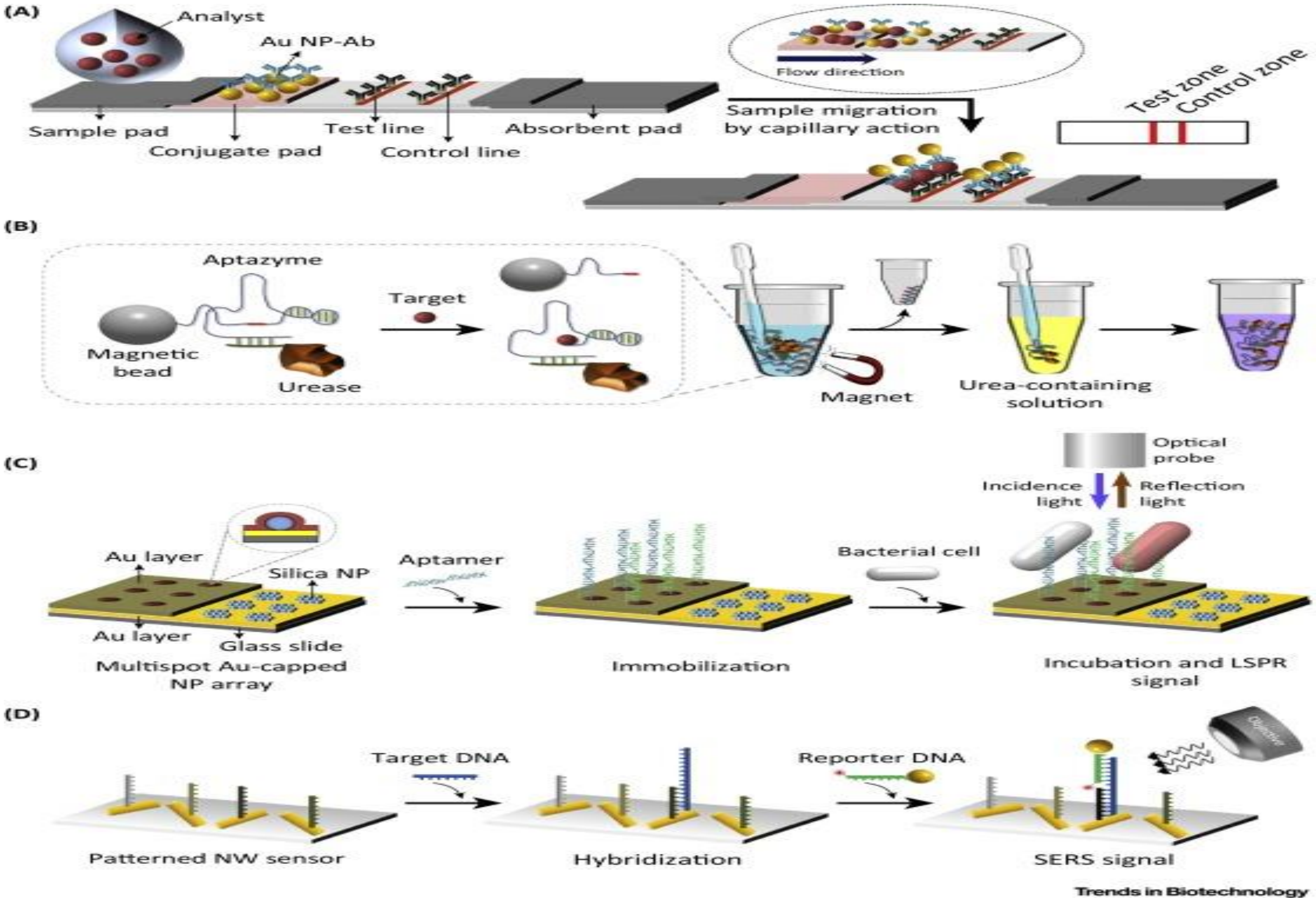


Figure 2. Experimental setup for potentiometric biosensor assays.

OPTICAL DETECTION BIOSENSOR

- The output transduced signal that is measured is light for this type of biosensor.
- The biosensor can be made based on optical diffraction. In optical diffraction based devices, a silicon wafer is coated with a protein via covalent bonds. The wafer is exposed to UV light through a photo-mask and the antibodies becomeinactive in the exposed regions. When the diced wafer chips are incubated in an analyte, antigen-antibody bindings are formed in the active regions , thus creating a diffraction grating. This grating produces a diffraction signal when illuminated with a light source such as laser. The resulting signal can be measured.

OPTICAL DETECTION BIOSENSOR



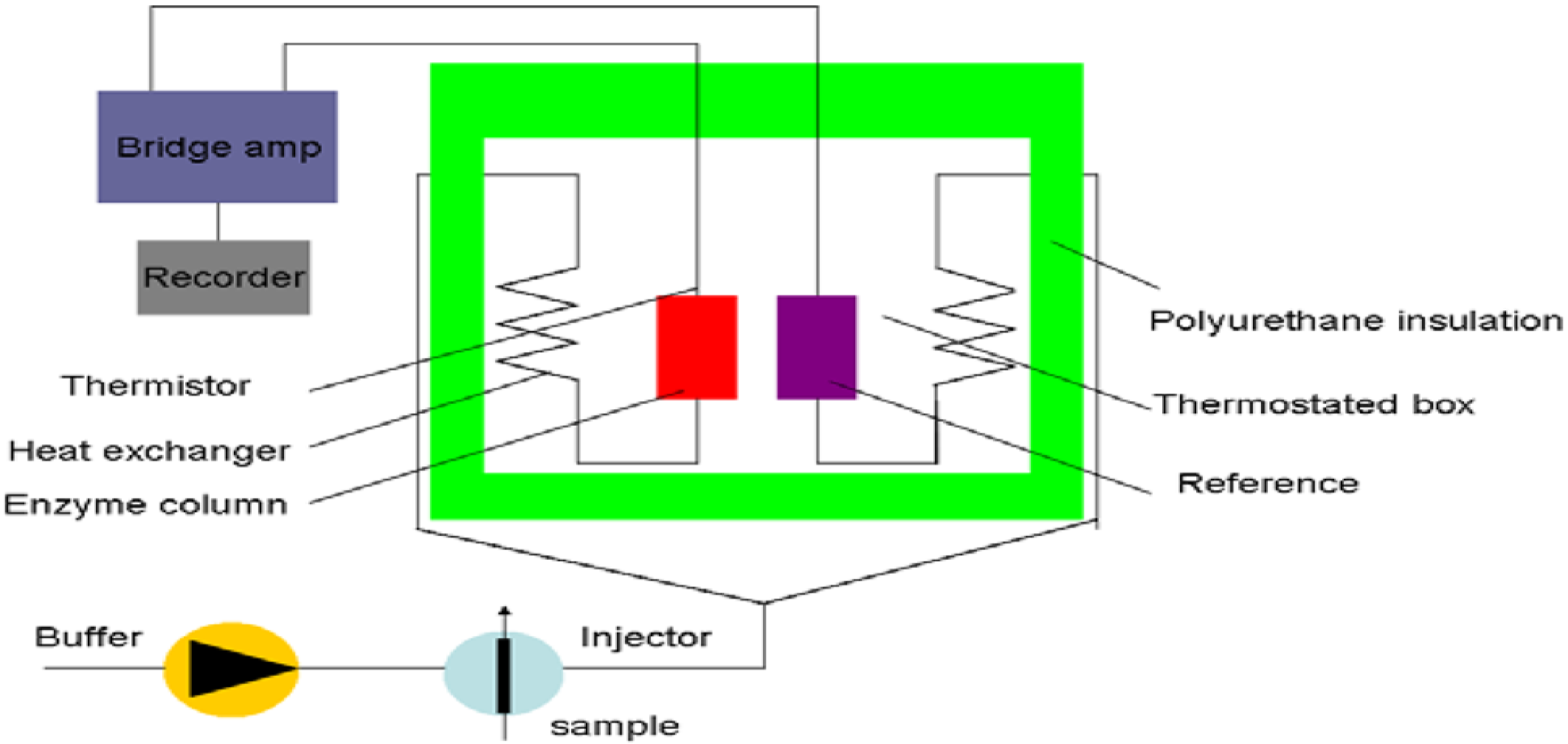
THERMAL DETECTION BIOSENSORS

- This type of biosensor work on the fundamental properties of biological reactions, namely absorption or production of heat , which in turn changes the temperature of the medium in which the reaction takes place.
- They are constructed by combining immobilized enzymes molecules with the temperature sensors. When the analyte comes in contact with the enzyme is measured and is calibrated against the analyte concentration.
- The total heat produced or absorbed is proportional to the molar enthalpy and the total number of molecules in the reaction.

Conti…

- The measurement of the temperature is typically accomplished via a thermistor, and such devices are known as enzyme thermistors. Their high sensitivity to thermal changes makes thermistor ideal for such applications.
- Unlike other transducers, thermal biosensors do not need frequent recalibration and are insensitive to the optical and electrochemical properties of the sample.
- Common applications of this type of biosensors includes the detection of pesticides and pathogenic bacteria.

Thermal detection biosensors



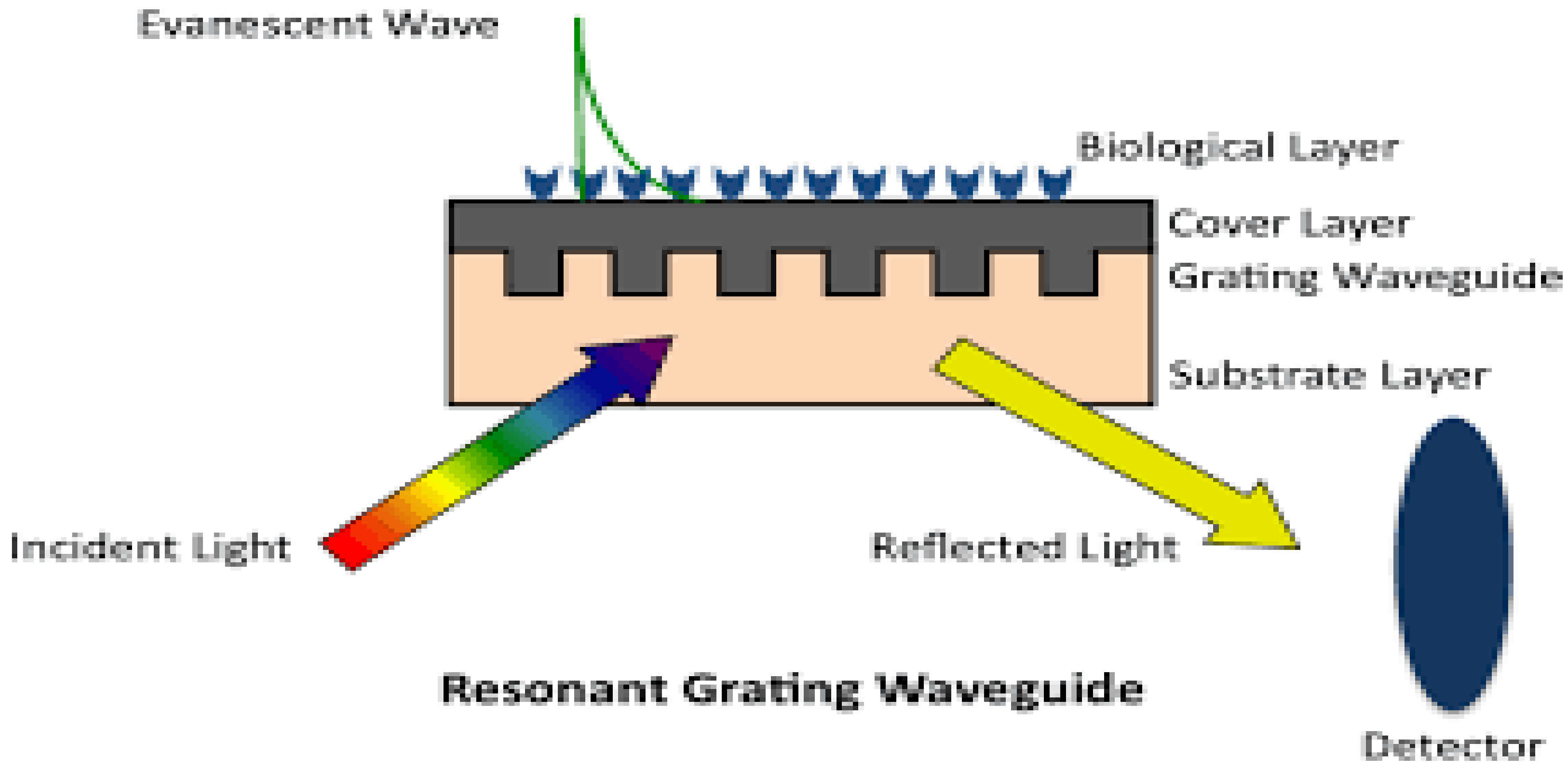
RESONANT BIOSENSORS

- It utilize crystal which undergo an electric deformation when an electrical potential is applied to them. (Alternating potential (A.C) produces a standing wave in the crystal at a characteristic frequency)
- In this type of biosensor, an acoustic wave transducer is coupled with an antibody (bio-element).

Conti…

- When the analyte molecule (or antigen) gets attached to the membrane, the mass of the membrane, the mass of the membrane changes. The resulting change in the mass subsequently changes the resonant frequency of the resonant frequency of the transducer. This frequency change is then measured.

RESONANT BIOSENSORS



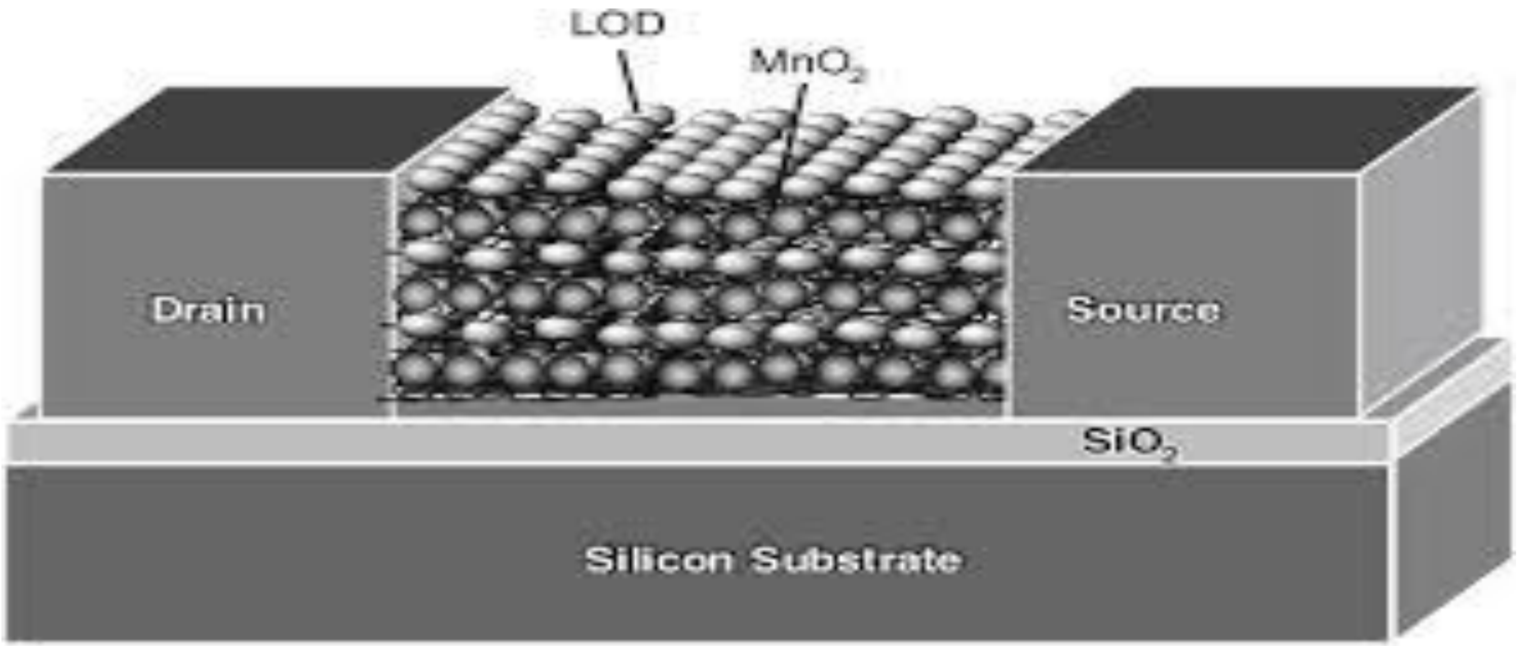
Resonant Grating Waveguide

Ion sensitive biosensor

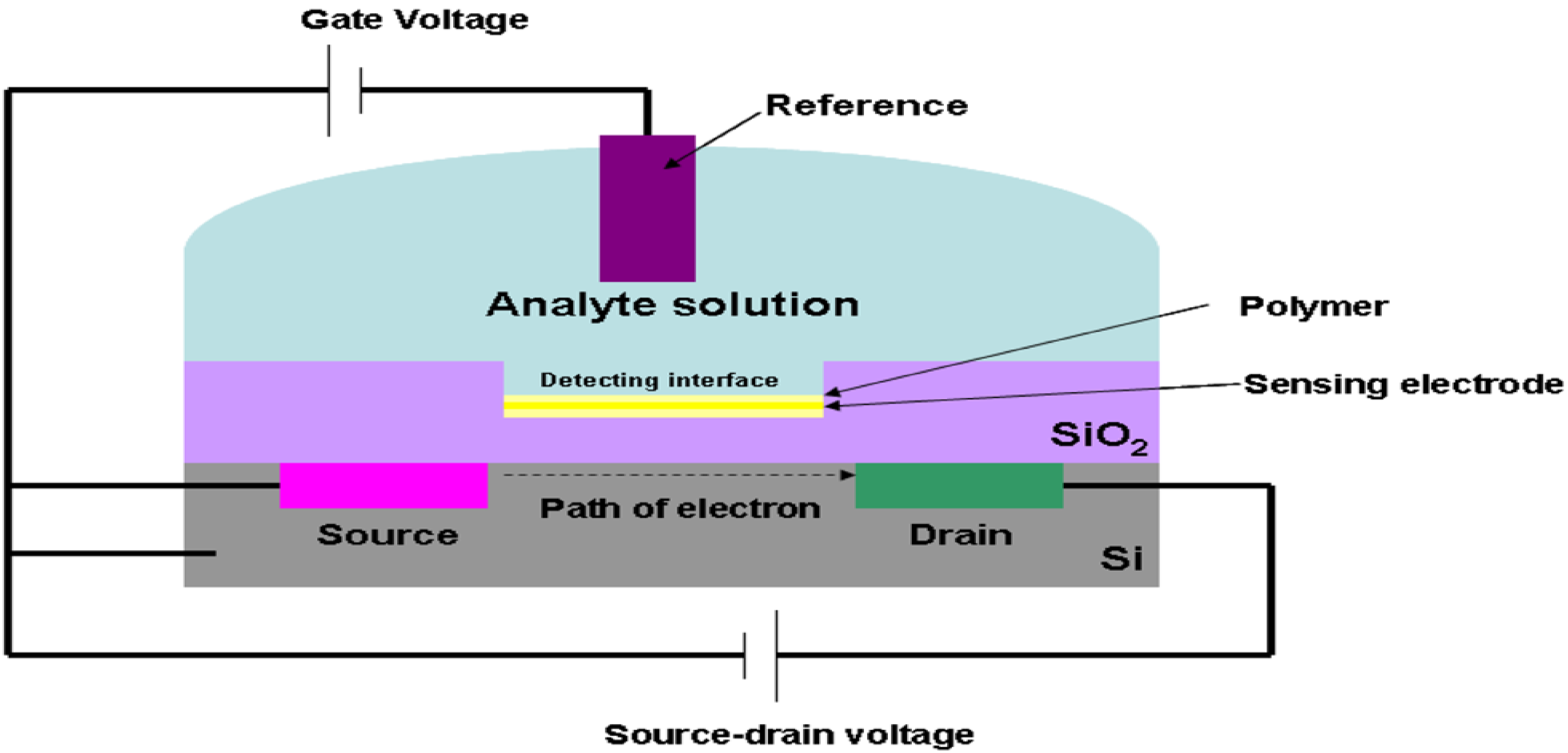
- These are semiconductor FETs having an ion-sensitive surface.
- The surface electrical potential changes when the ions and semiconductor interact. (This change in the potential can be subsequently measured).
- The Ion sensitive Fielf Effect Transistor (ISFET) can be constructed by covering the sensor electrode with a polymer layer. This polymer layer is selectively permeable to analyte ions. The ions diffuse through the polymer layer and in return cause a change in the FET surface potential.

Conti…

- This type of biosensor is also called an ENFET (Enzyme Field Effect Transistor) and is primarily used for pH detection.



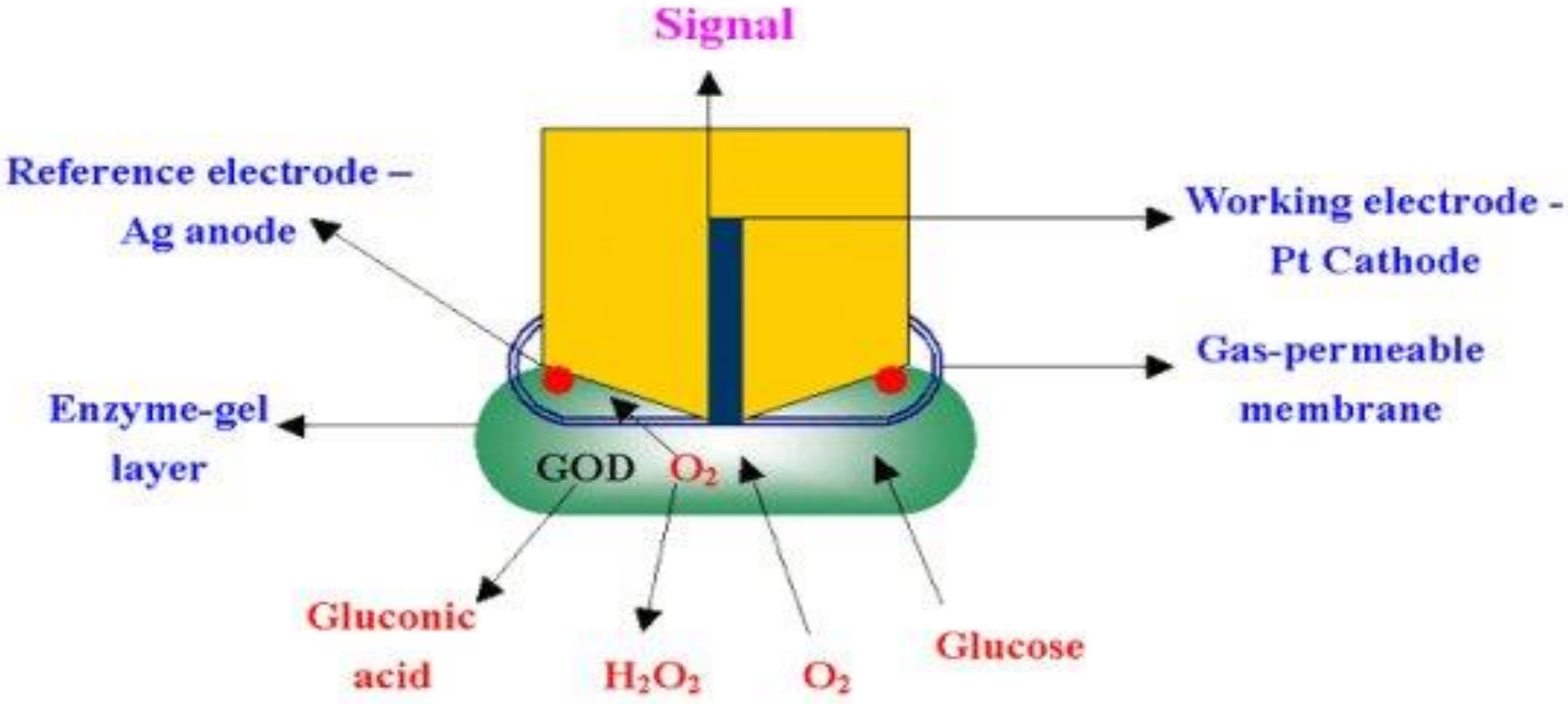
ION SENSITIVE BIOSENSORS



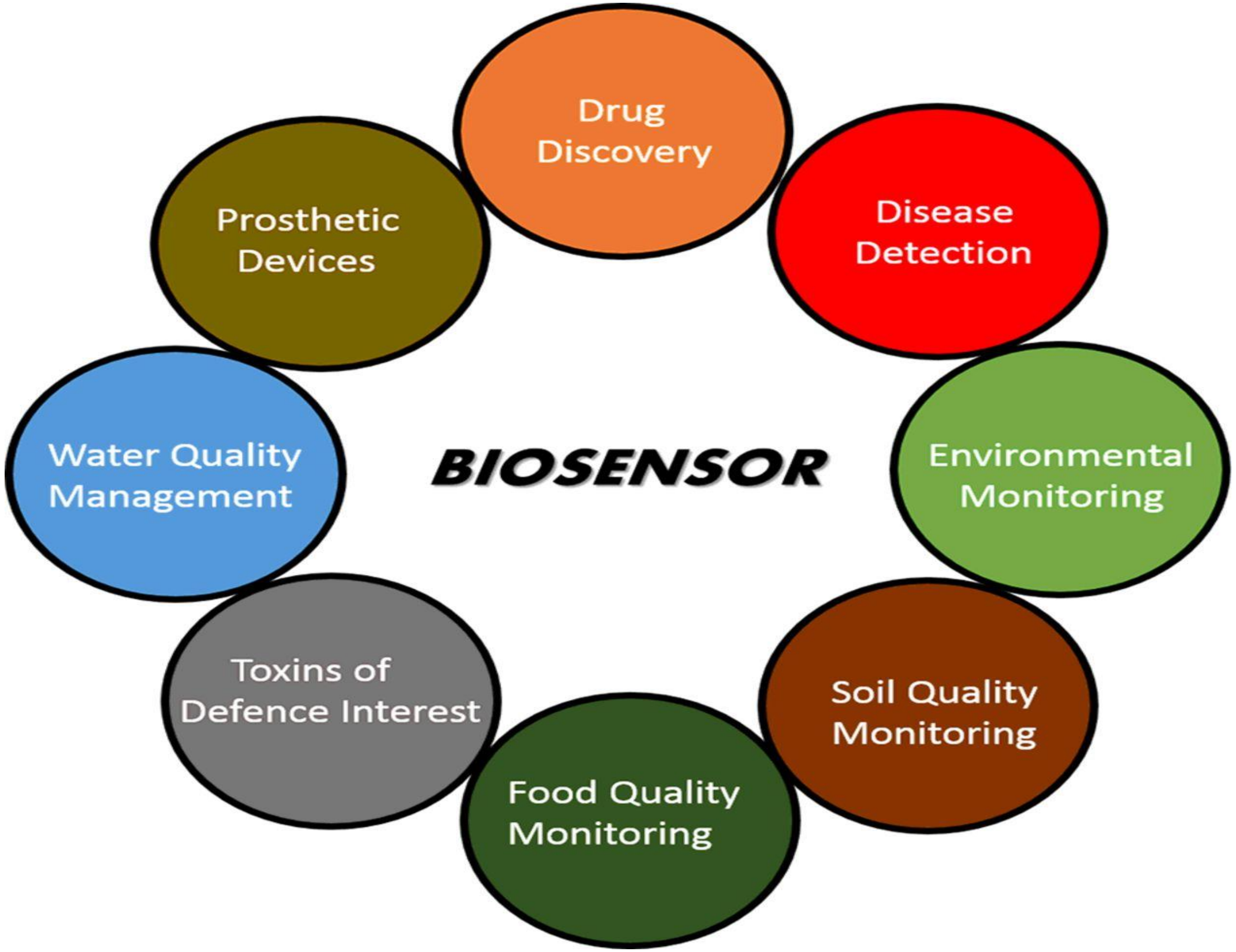
GLUCOSE BIOSENSORS

- Glucose reacts with glucose oxidase to form gluconic acid. Two electrons and two protons are also produced.
- Glucose mediator reacts with surrounding oxygen to form H2O2 and glucose oxidase.
- Now this glucose oxidase react with more glucose.
- Higher the glucose content, the higher the oxygen consumption.
- Glucose content can be detected by Pt-electrode.

Glucose biosensors



Applications



CONCLUSION

- As the potential threat to bioterrorism increase, there is great need for a tool that can quickly, reliably and accurately detect contaminating bio-agents in the atmosphere.
- Biosensors can essentially serve as low-cost and highly efficient devices for this purpose in addition to being used in other day-to-day application.
- Biosensors are known as immuno-sensors,optrodes, chemical, canaries, resonant mirrors, glucometers biochips bio-computers and so on.

Pharmaceutical Biotechnology

Concepts and Applications

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