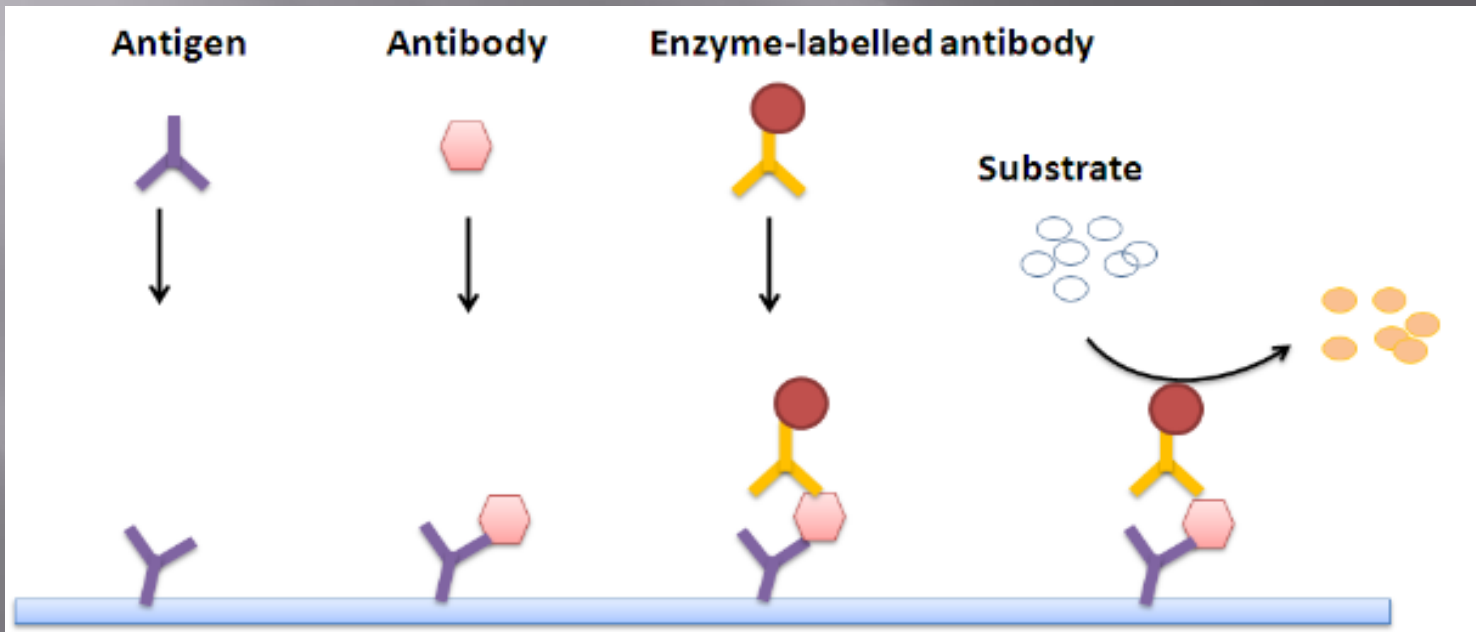


# ELISA

## Target research areas



# ELISA Kits cover a broad range of target research areas

- ▣ Adaptive Immunity System
- ▣ Angiogenic Factors
- ▣ Apoptosis Markers
- ▣ Artherosclerotic
- ▣ Biochemicals and Hormones
- ▣ Blood Proteins
- ▣ Cancer and Oncoproteins
- ▣ Cardiovascular Markers
- ▣ Cell Adhesion
- ▣ Colorimetric Cell Based
- ▣ Cytokines and Cytokine Receptors
- ▣ Developmental Proteins
- ▣ Environmental
- ▣ Growth Factors and Hormones
- ▣ Immunoglobulins
- ▣ Infectious Disease
- ▣ Lipid Metabolism
- ▣ Neuroscience Markers
- ▣ TNF
- ▣ Transcription Factor DNA Binding
- ▣ Other

# What is being tested using ELISA assay?

- ▣ If a protein with multiple epitopes is being detected, a sandwich assay is a good choice. It usually requires two antibodies that react with different epitopes. However, if the molecule has multiple repeating epitopes, it is possible in a sandwich assay to use the same antibody for both capture and detection. Alternatively, if there is a supply of the analyte to be detected in pure form that can absorb effectively to a microwell, then one can set up a competitive assay in which the purified analyte is immobilized and analyte in the sample competes with the immobilized analyte for binding to labeled antibody. In this case it is essential to titrate the antibody so that it is limiting, or else the assay sensitivity will be lowered.

# What is being tested using ELISA assay?

- ▣ Polystyrene will bind a wide variety of proteins in an increasing amount depending on their concentration in the coating solution. The specific and optimal amount needs to be determined for each protein, but some general observations have been made for antibodies. Medium to low binding plates bind typically up to 100-200 ng of IgG/cm<sup>2</sup> while high binding plates typically can bind up to 400-500 ng of IgG/cm<sup>2</sup>. In addition to proteins, polystyrene plates will absorb peptides generally of 15-20 amino acids in length. In order to achieve strong binding, a peptide will need both hydrophobic and hydrophilic interactions. Typically a drawback to adsorbing peptides directly is that they tend to have few epitopes, and if these are involved in interaction with the plastic, it will be difficult for an antibody to bind to them. One alternative is to attach the peptide to a larger protein through a spacer arm that provides some distance between the peptide and the protein, allowing the antibody to interact with the peptide.

# What is being tested using ELISA assay?

- ▣ An organism such as bacterial or viral assays that detect whole organisms can also use sandwich assays with the same antibody for both capture and detection. If the target molecule is small or consists of a single epitope, a modification of the formats described above is needed. Small molecules by themselves either do not adsorb well to a solid phase, or may be masked by the blocking protein added. However, small molecules can often be attached to larger proteins which provide a means to attach the desired epitope to a solid phase in a configuration that allows the epitope to be bound by an antibody.



# Advantages and disadvantages of ELISA

## ADVANTAGES

- ▣ High sensitivity and specificity: it is common for ELISAs to detect antigens at the picogram level in a very specific manner due to the use of antibodies.
- ▣ High throughput: commercial ELISA kits are normally available in a 96-well plate format. But the assay can be easily adapted to 384-well plates.
- ▣ Easy to perform: protocols are easy to follow and involve little hands-on time.
- ▣ Quantitative: it can determine the concentration of antigen in a sample.
- ▣ Possibility to test various sample types: serum, plasma, cellular and tissue extracts, urine, and saliva among others.

## DISADVANTAGES

- ▣ Temporary readouts: detection is based on enzyme/substrate reactions and therefore readout must be obtained in a short time span.
- ▣ Limited antigen information: information limited to amount or presence of the antigen in the sample.

# ELISA Advantages

1. **High Sensitivity** The high sensitivity of ELISA, comes from the enzyme as a reporting group. As is known to all, the enzyme is an organic catalyst, a small amount of which could induce a large span of catalytic reactions to produce observable chromogenic reaction phenomenon. Therefore, this system is often taken as the amplification system of enzyme. By ELISA, a tracer of the antigen or antibody is achieved in the cell or subcellular level, also, antigen or antibody quantification can be done in the microgram or even nanogram levels.
2. **Strong Specificity** Specificity of ELISA is because of the selectivity of the antibody or antigen. Actually, the binding of antigen or antibody only occurs in the epitope of an antigen or antigen-binding site of an antibody. Since, there is a complementary relationship between epitope and antigen-binding site both in chemical structure and spatial configuration, the reaction between antigen and antibody shows a strong specificity.

# ELISA Advantages

- ▣ Compared to other immunoassay methods, there are many advantages of ELISA. ELISA tests are more accurate. They are considered highly sensitive, specific and compare favorably with other methods used to detect substances in the body, such as radioimmune assay (RIA) tests. ELISA possesses the added advantages of not needing radioisotopes (radioactive substances) or a costly radiation counter (a radiation-counting apparatus).



# References

- ▣ <https://www.abcam.com>
- ▣ <https://www.sinobiological.com>
- ▣ <https://www.bio-rad>
- ▣ <https://www.thermofisher.com>