APPLICATIONS OF ELISA (ENZYME-LINKED IMMUNOSORBENT ASSAY) IN DIAGNOSTIC MEDICINE

SEMINAR PRESENTION

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SUMMARY

An enzyme-linked immunosorbent assay (ELISA) is typically performed to detect the presence and/ or amount of a target protein of interest within an experimental sample. Detection of the target protein is made possible by antibodies which make the ELISA an immunoassay. Through a series of incubation and washing steps, these antibodies/ reacts with a substrate to give a colour change when it detects protein on the microtitre plate, hence indicating the presence of the protein of interest in the sample.

SUMMARY CONT'D

The result can either be qualitative, semi-quantitative or quantitative. The type includes direct, indirect, sandwich and competitive ELISA and plays important roles in toxicology (drug screening), antigen or antibody detection in sample (HIV virus, hepatitis b and c etc), hormone detection (HCG, LH, TSH), in food industries (for detection of food allergens) and the detection of various diseases such as syphilis, Chlamydia, etc.

Introduction

Enzyme-linked immunosorbent assay (ELISA) also known as enzyme immune assay (EIA) is a biochemical technique used mainly in immunology to detect the presence of proteins (antibody or antigen), hormones etc in a sample.

History

The origin of ELISA was the idea of finding an alternative method to radio immune assay (RIA) which uses radioactive label of which some scientists came up with idea of using enzyme labels in immune assay.

In 1966, Perlmann and Schuurs invented the method to prove the using of enzyme-linked immunoassay was feasible.

In 1971, peter Perlman and Eva Engvall at Stockholm University in Sweden published papers introducing EIA/ ELISA methods.

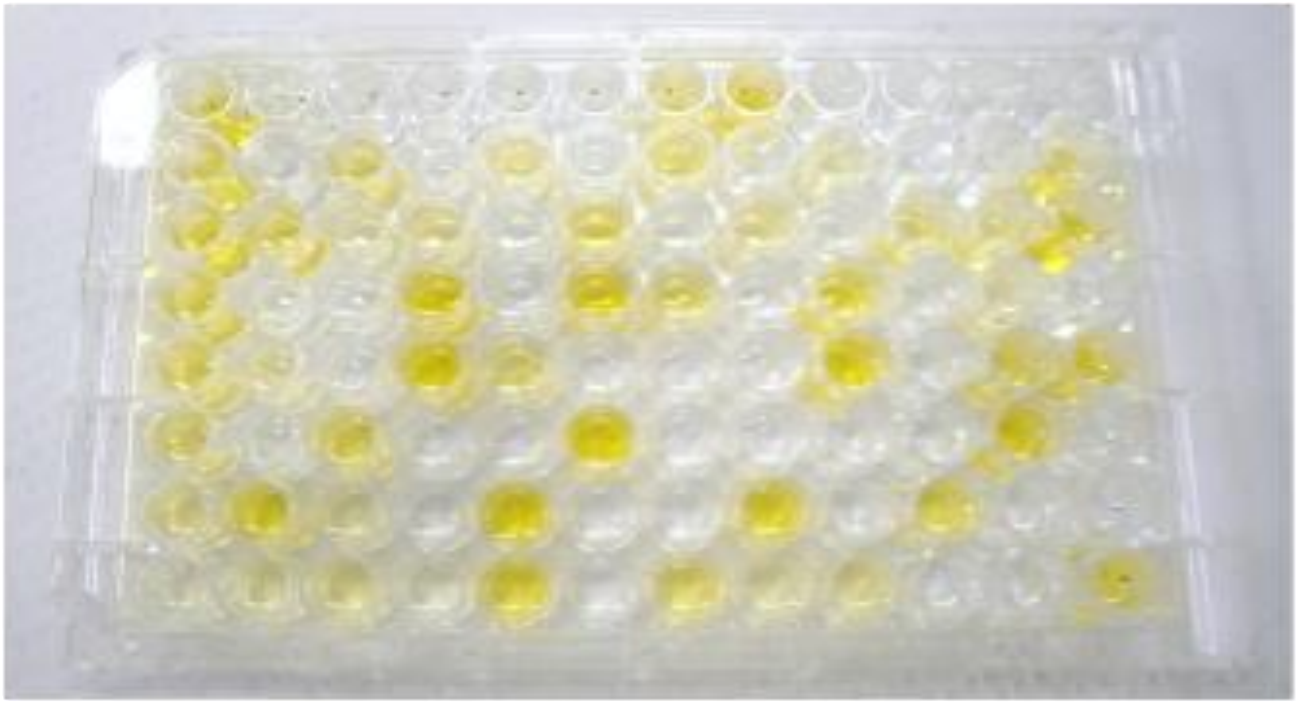
Principles

- Enzyme- linked immunosorbent assay (ELISA) is a method of target antigen (or antibody) capture in samples using a specific antibody (or antigen) and of target molecule detection/ quantitation using an enzyme reaction with its substrate which produces colour change.

ELISA MACHINE

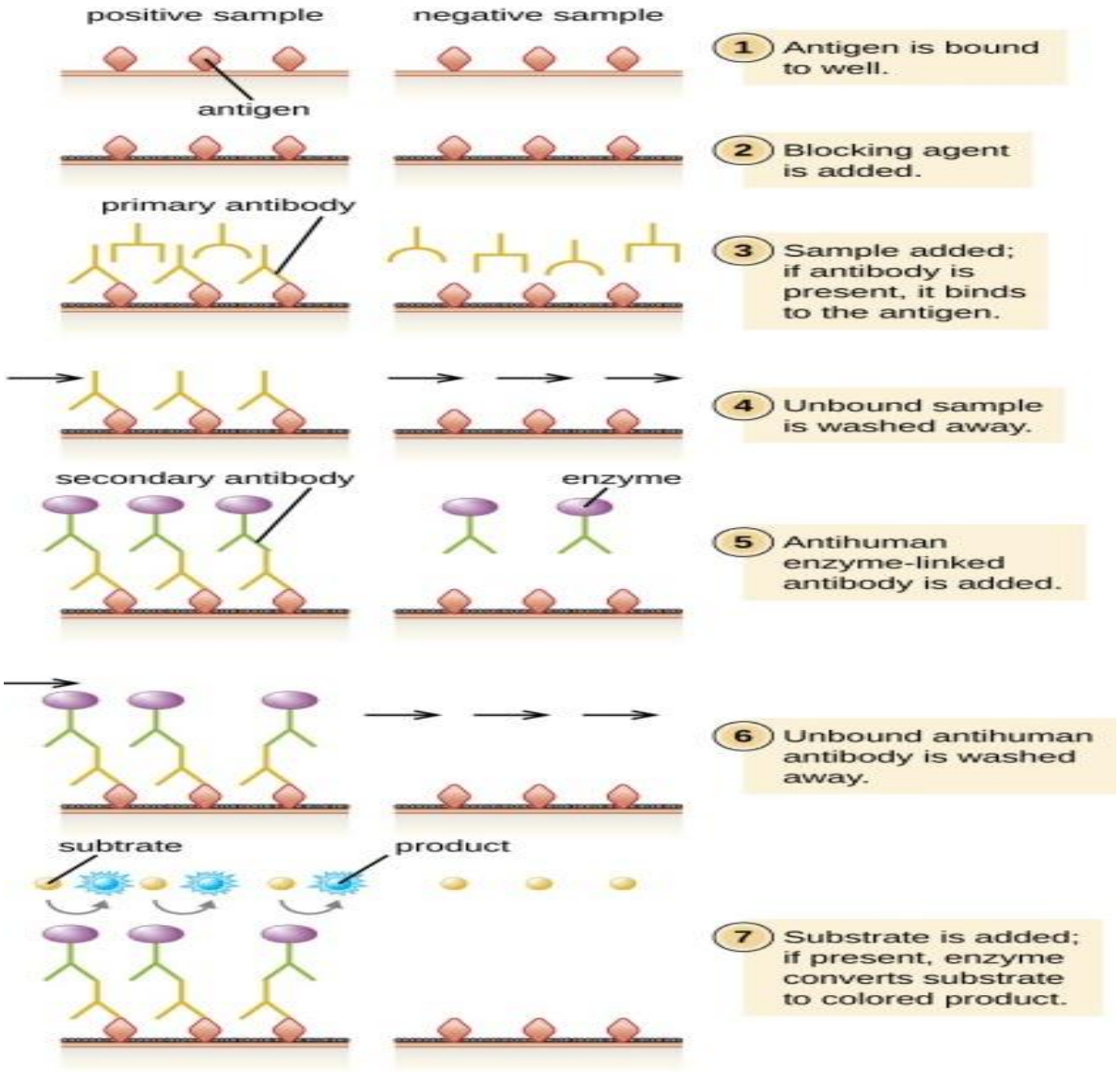
MICRO TITRE PLATE

ELISA: An example of an assay using a 96-well plate.



The yellow color indicates that the target protein is present. The higher degree of the color, the higher concentration of the target protein.

MECHANISM


positive sample
negative sample
antigen
primary antibody
secondary antibody
enzyme
subtrate
product
1 Antigen is bound to well.
2 Blocking agent is added.
3 Sample added; if antibody is present, it binds to the antigen.
4 Unbound sample is washed away.
5 Antihuman enzyme-linked antibody is added.
6 Unbound antihuman antibody is washed away.
7 Substrate is added; if present, enzyme converts substrate to colored product.

TYPES OF ELISA

- Direct ELISA
- Indirect ELISA
- Sandwich ELISA
- Competitive ELISA

APPLICATIONS

- Screening donated blood for evidence of viral contamination by
  - HIV- 1 and HIV- 2 (presence of anti- HIV antibodies)
  - Hepatitis C (presence of antibodies)
  - Hepatitis B (for both antibodies and viral antigen)

- Measuring hormone levels
  - HCG (as a test for pregnancy)
  - LH (determining the time of ovulation)
  - TSH, T3 and T4 (for thyroid function)

APPLICATIONS CONT'D

- Detecting infections
  - sexually- transmitted agents like HIV, syphilis and Chlamydia
  - hepatitis B and C virus

- Detecting allergens in food
- Measuring Rheumatoid factors and other antibody in autoimmune diseases like lupus erythematosus
- Detecting illicit drugs e.g. cocaine, opiates

REFERENCES

- Elgert, K,.D. (2009). Immunology: Understanding the immune system. John Wiley and sons .149-50
- Ferretti, F., Porcelli, B., terzuoli, l. (2014). Assessments of a test for the Screening and Diagnosis of Celiac Disease. *Journal of Clinical laboratory analysis;* 30(1): 65- 70.
- Griffin, J., Spittle, E., Rodgers, c. (2005). Immunoglobulin GI Enzyme-linked Immunosorbent Assay. *Journal of Clinical and vaccine immunology;* 12: 1401- 1409.
- Msagati, A.T. (2017). Food forensics and Toxicology. *The American Journal of Gastroenterology;* 95(5): 1253-1257.,
- Yalow, Rosalyn S., Berson, Solomon A. (2009). Immune assay of endogenous plasma iinsulin in man. The journal of clinical investigation ; 39. 1157 – 1175.