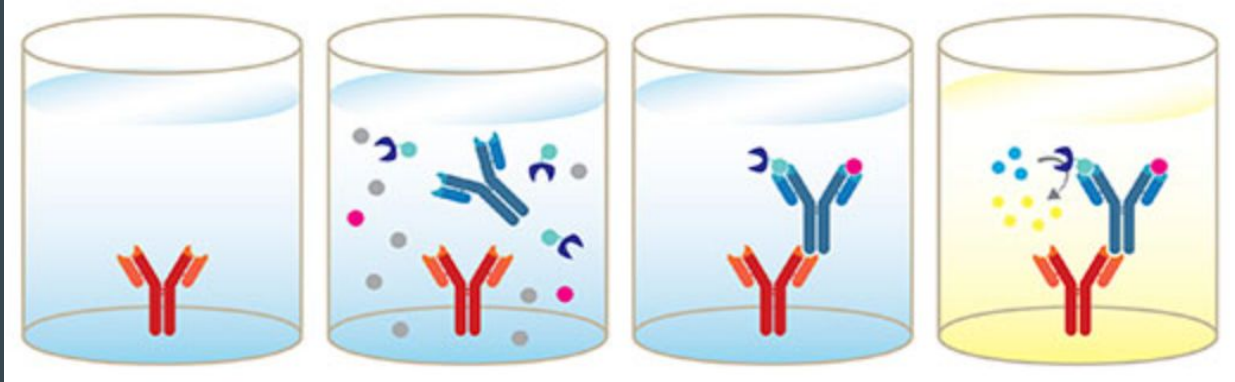
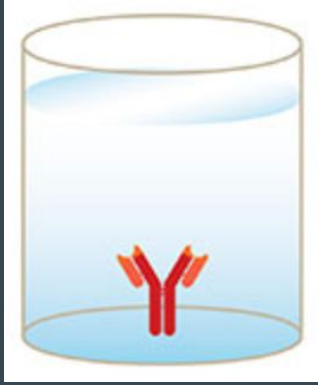


Competitive ELISA



Are other Sandwich &
Direct techniques

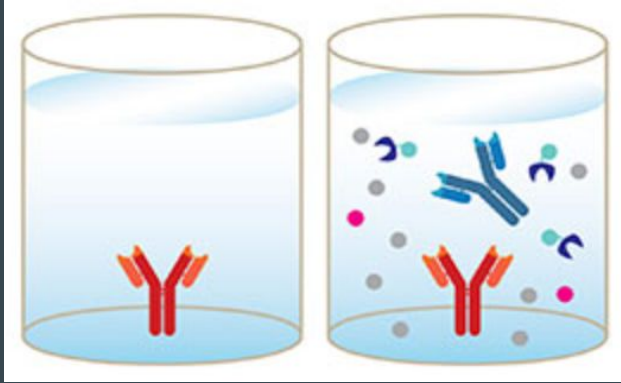
An ELISA example...



Coat wells with a
secondary capture
antibody.

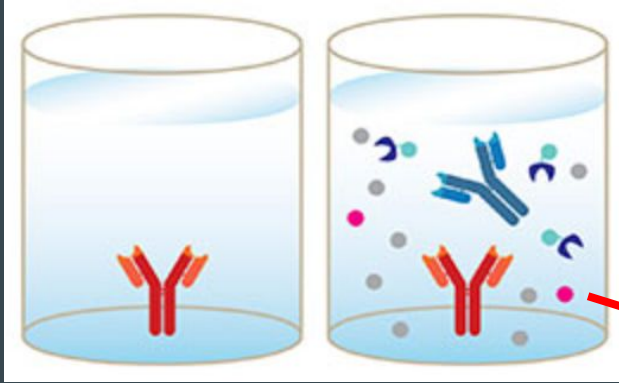
The goal of an ELISA is to quantify the unknown concentration of an analyte in a given sample.

An ELISA example...



Then, roughly at the same time we add...

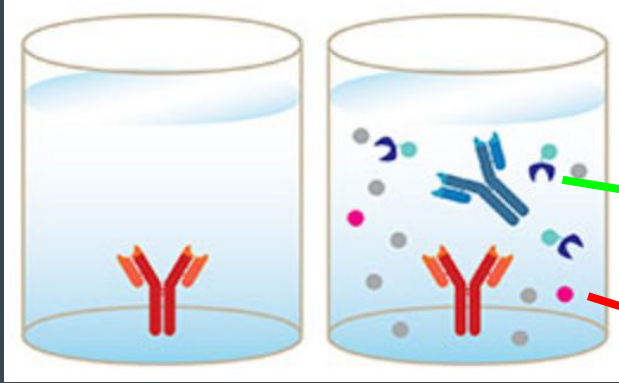
An ELISA example...



Then, roughly at the same
time we add...

A solution with antigen

An ELISA example...

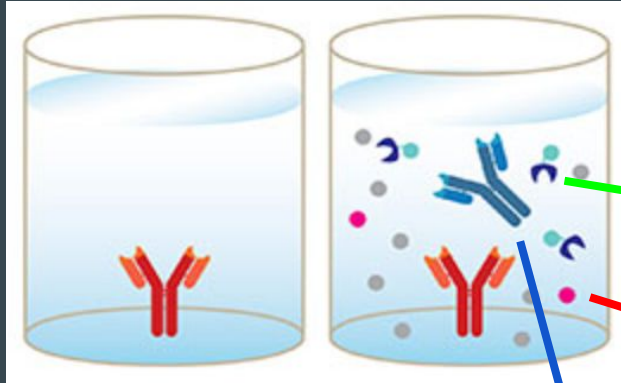


Then, roughly at the same time we add...

A solution with enzyme-conjugated antigen

A solution with antigen

An ELISA example...



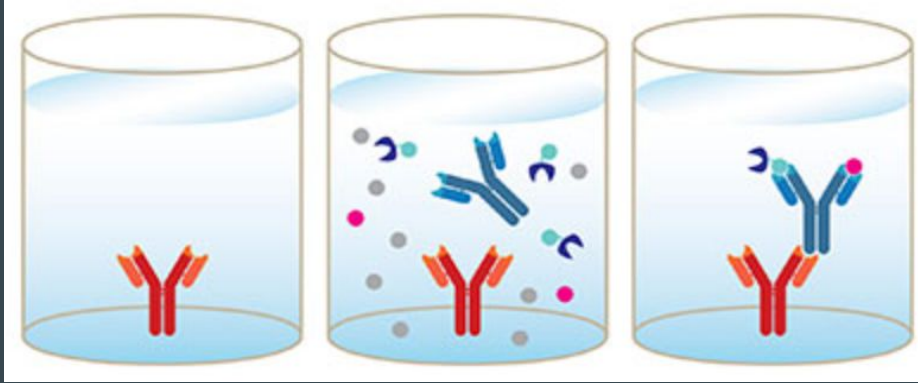
Then, roughly at the same time we add...

A solution with enzyme-conjugated antigen

A solution with antigen

A solution with primary detection antibody

An ELISA example...



The primary antibody
binds with...

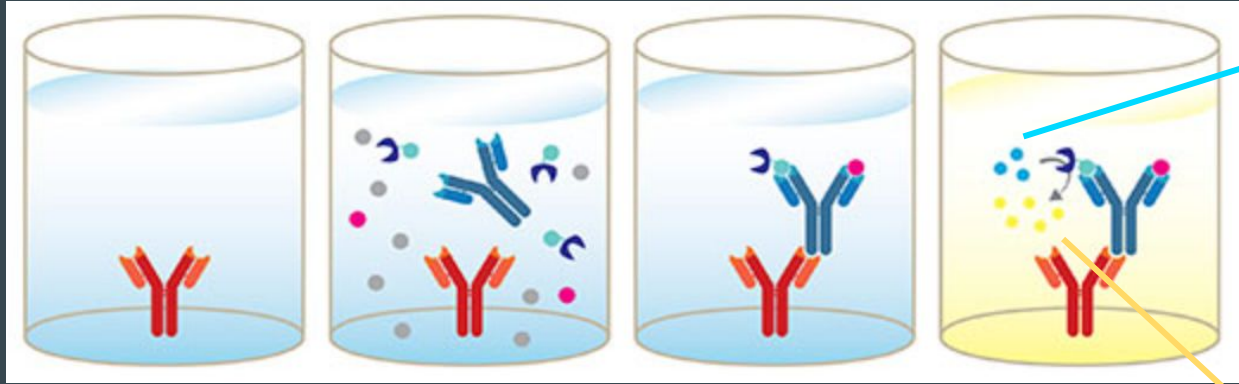
the sample antigen

the conjugated antigen

& secondary antibody
coated to the plate

We then wash away any
excess antigen and
enzyme-conjugated
antigen from the well.

An ELISA example...



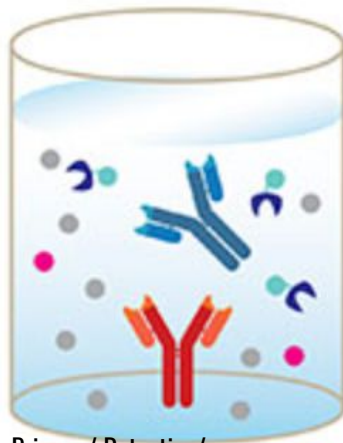
Then we add substrate solution. It reacts with the enzyme attached to the conjugated antigen.

The reaction produces a yellow product when stopped with an acid solution.

Competitive ELISA



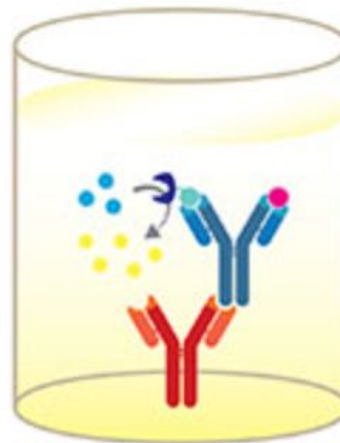
Wells are pre-coated with secondary capture antibody



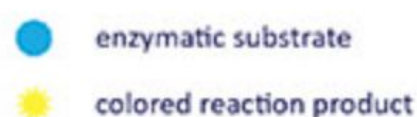
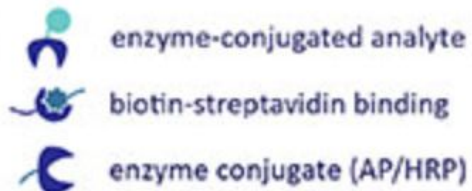
Primary/ Detection/
Capture antibody, conjugate and sample are added simultaneously



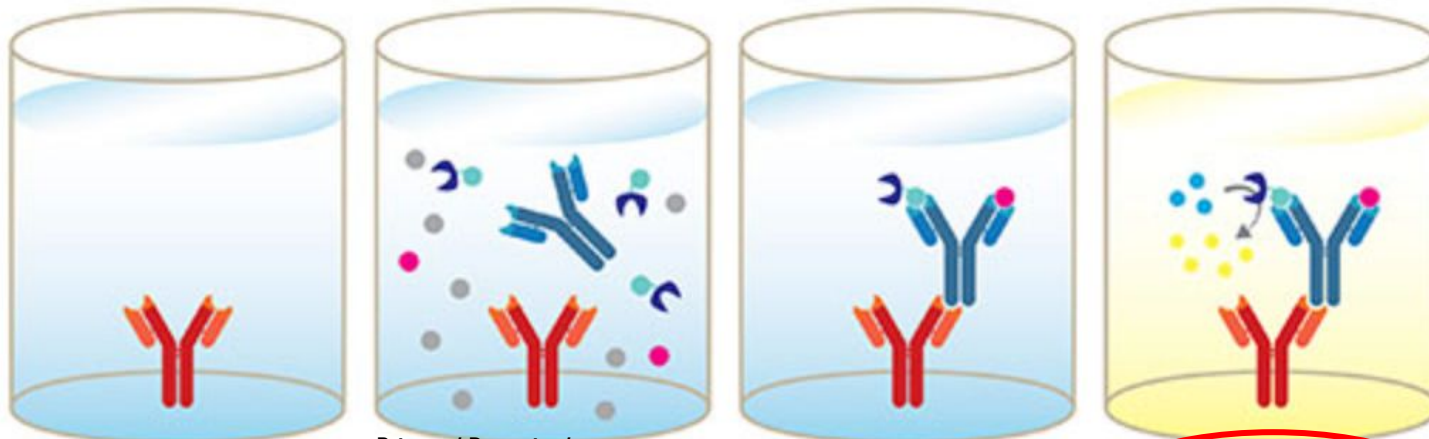
Analyte and conjugate compete for binding



Enzymatic color reaction proportional to bound conjugate



Competitive ELISA

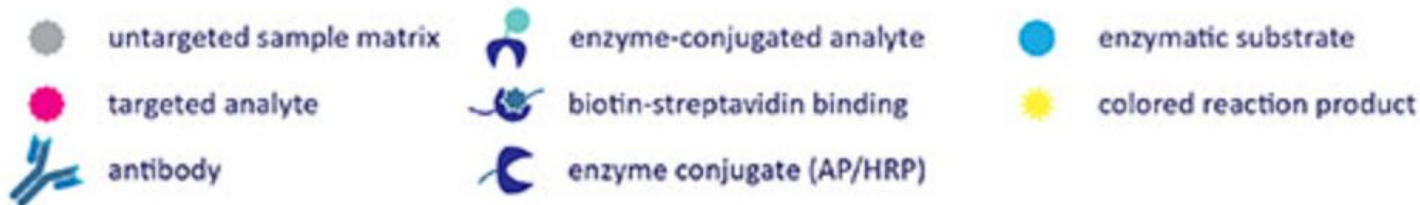


Wells are pre-coated with secondary capture antibody

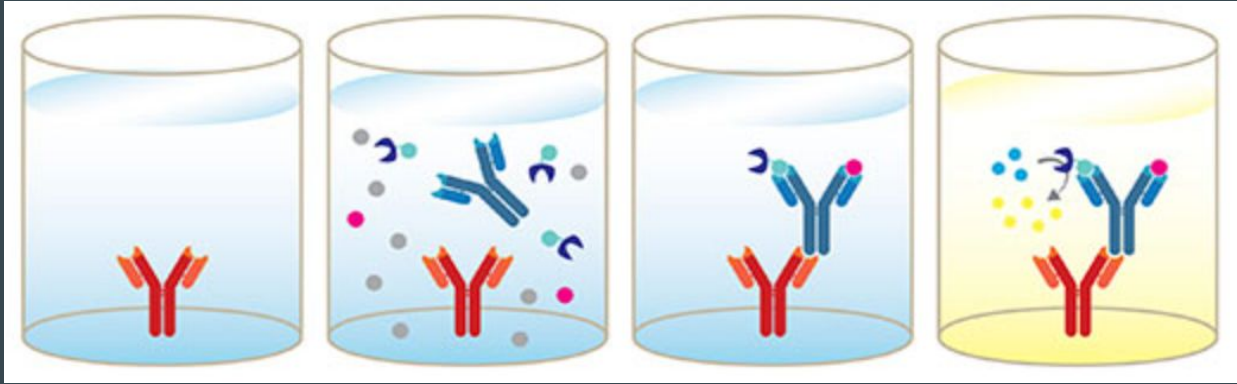
Primary/ Detection/
Capture antibody, conjugate and sample are added simultaneously

Analyte and conjugate compete for binding

Enzymatic color reaction proportional to bound conjugate



Competitive ELISA

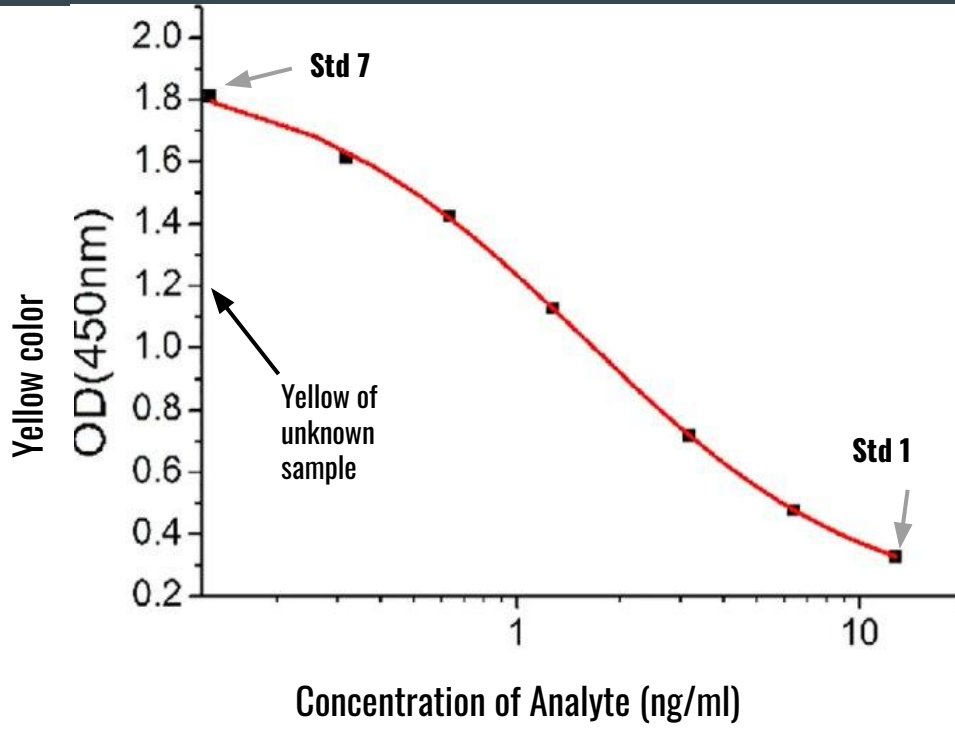


How does the color reaction help us figure out the unknown concentration of the analyte?





The Standard Curve



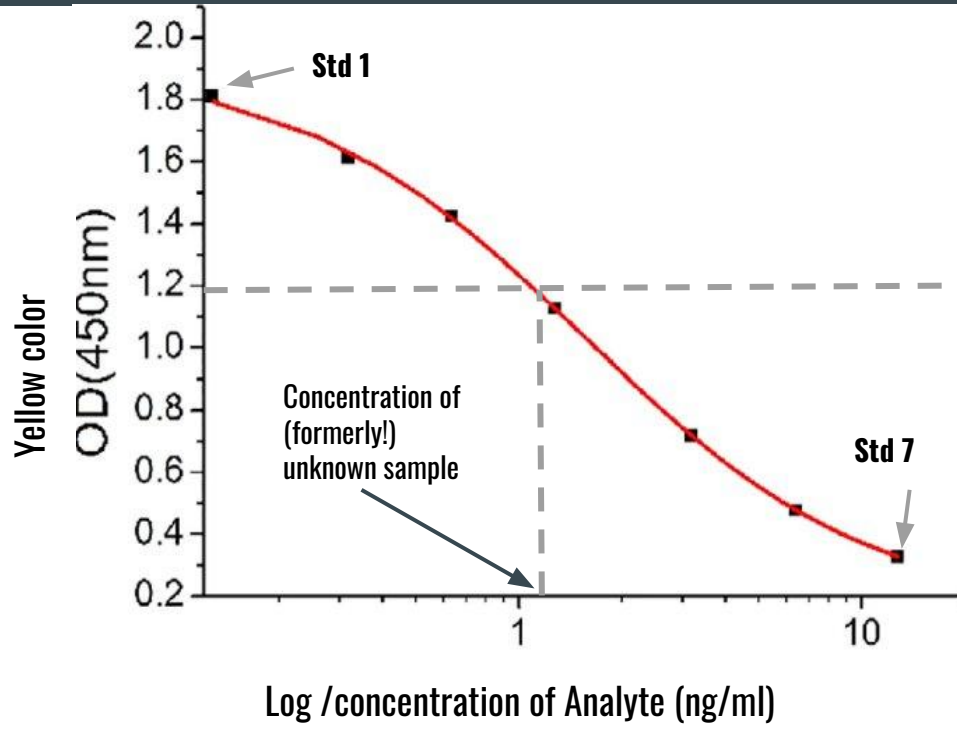
After a plate reader quantifies the yellow color (OD) for each well in the plate...

We use samples of KNOWN concentration to show the relationship of OD's (y-axis) with concentration.

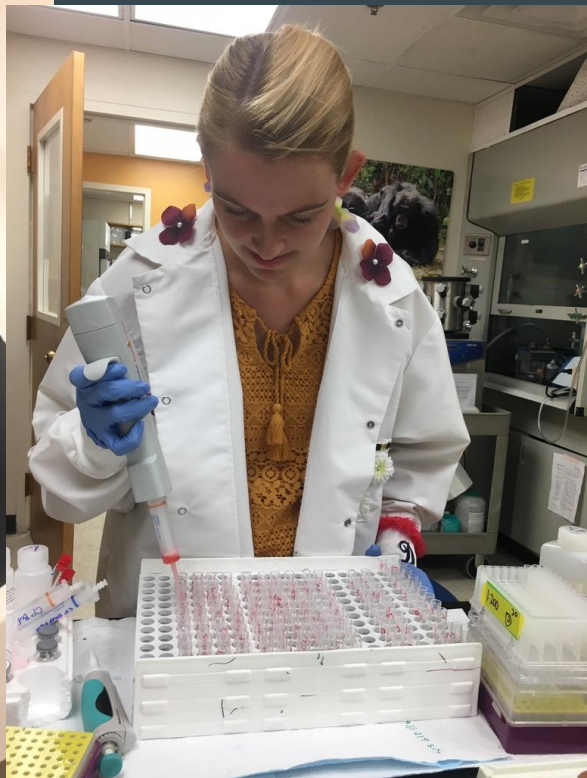
Then when we have an UNKNOWN concentration, we take its OD...



The Standard Curve



... and infer its corresponding concentration from the curve.



RIA: radio
immunoassay

