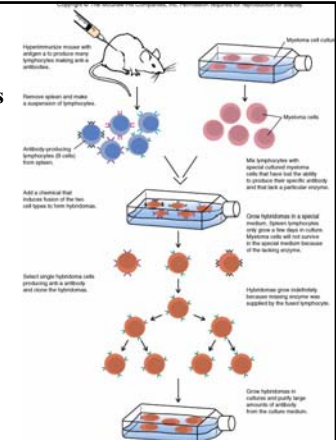


Immunological testing

- Monoclonal antibodies
- Serology
- Quantifying antigen – antibody reactions

17-1

Perspective 17.1 Monoclonal Antibodies



17-2

Serology

- Antibodies
- Antibodies detect and identify antigens

17-3

Quantifying antigen – antibody reactions

- Seroconversion or rise in titer
- Serial dilutions

17-4

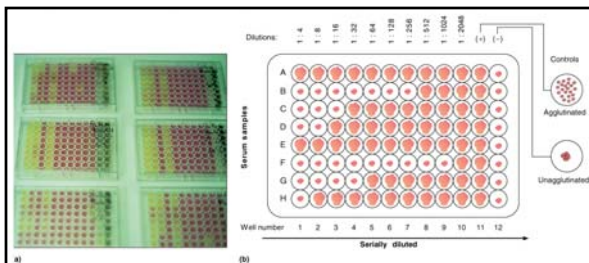


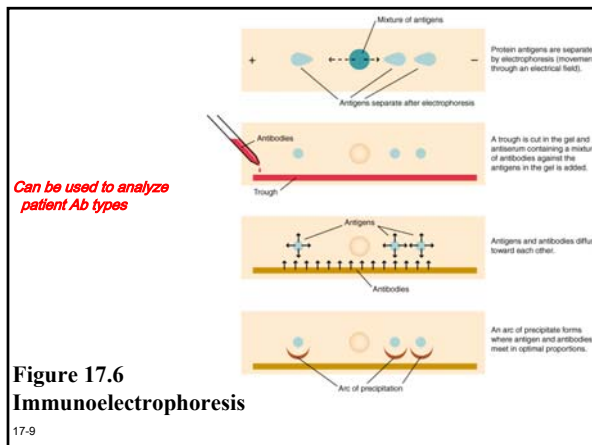
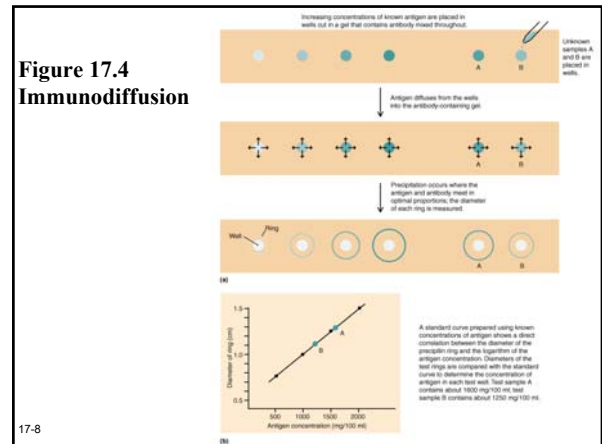
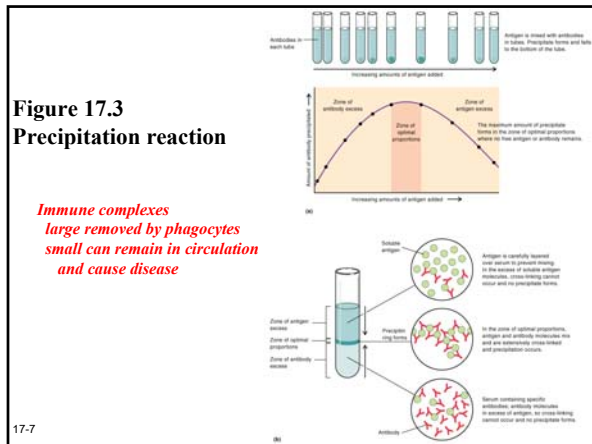
Figure 17.2 - Quantitation of immunologic tests

17-5

Precipitation reactions

- Immunodiffusion
- Immunoelectrophoresis

17-6



Agglutination reactions

- Direct agglutination
- Indirect agglutination
- Hemagglutination

17-10

Direct agglutination

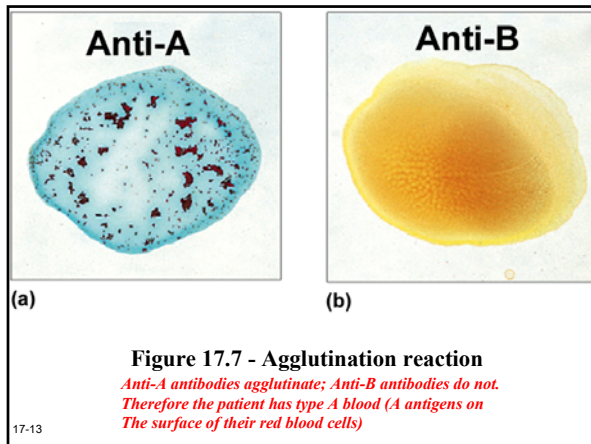
- Cross – linking and lattice formation
- Antibodies react with particulate antigens (red blood cells, bacteria, fungi)
- Visible clumps
- Estimate amount of antibody

17-11

Indirect agglutination

- Soluble antigen is coated onto particles (red blood cells, latex beads)
- Allow for visible clumps (agglutination)

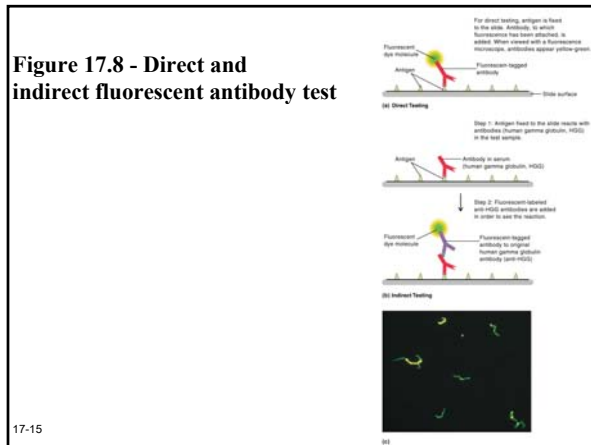
17-12



Immunofluorescence tests

- Direct fluorescent antibody test
- Indirect fluorescent antibody test

17-14



Antigen – antibody assays

- Radioimmunoassay (RIA)
- Enzyme – linked immunosorbent assay (ELISA)
- Western blot

17-16

Radioimmunoassay (RIA)

- Competitive inhibition assay
- Measure antigen or antibody
- Ex. Measure small amounts of hormones or drugs in a clinical sample
- Ex. Measure small amounts of IgE antibody (radioallergosorbent test)

*Unlabeled Ab is used to coat well
 Labeled specific Ag is added with sample
 Ability of unlabeled Ag in sample to compete with labeled Ag binding to Ab is measured
 Reduced binding indicates competition by unlabeled Ag in sample
 Amount of competition a measure of unlabeled Ag levels*

17-17

Enzyme – linked immunosorbent assay (ELISA)

*Widely used; very sensitive; small volumes; little reagent; lots of samples
 Used for HIV testing of blood before it is used for transfusion*

17-18

Neutralization test

- Antibody bind to specific antigen (virus, toxin)
- Antibody – antigen complex prevents antigen from binding (neutralization)
- *Viral or toxin activity is diminished in tests*

17-25

Cellular immunology test

- Identification of subsets of lymphocytes (*using FACS*)

17-26

Cellular immunology test

- Identification of subsets of lymphocytes Lymphocyte response to mitogens
- Cytotoxic T – cell function
- Cell – mediated immunity to infectious agents
Ag used instead of mitogen to stimulate lymphocytes

17-27