WHAT IS AUTOIMMUNITY?

Understanding Autoimmune Diseases (NIH)

Autoimmunity occurs when the immune system loses the ability to distinguish “self” from “non-self” and mistakenly attacks one’s own cells or tissues.

There are over 80 autoimmune diseases currently identified. Some are organ-specific whereas others are systemic (occurring throughout the body).

http://autoimmune.pathology.jhmi.edu/whatis_spectrum.cfm
MOLECULAR MIMICRY HYPOTHESIS

*Molecular mimicry* occurs when the structure of a foreign antigen is very similar to antigen present on our own cells or tissues:

- Upon presentation of the foreign antigen, lymphocytes are activated to target this antigen
- These lymphocytes could potentially also react to self-antigen
- This leads to the destruction of one’s own cells or tissues

*Molecular Mimicry as a Mechanism for Autoimmune Disease*  
Another potential issue that contributes to the breakdown of tolerance seen in autoimmunity is an imbalance between Th17 and Treg cells.

The following autoimmune diseases are known to be related to Th17/IL-17 production:

- Rheumatoid Arthritis
- Inflammatory Bowel Disease
- Multiple Sclerosis
- Psoriasis
- Ankylosing Spondylitis
- Sjögren’s Syndrome
Rheumatoid arthritis (RA) is a chronic inflammatory condition that impacts the lining of the joints. Many of our joints are surrounded by a synovial membrane – and when this area becomes inflamed, it leads to cartilage thinning and bone loss.
RHEUMATOID ARTHRITIS: ANIMAL MODEL

- Adjuvant-induced Arthritis in Lewis Rats
ONSET OF DISEASE & ADMINISTERING TREATMENT
DEVELOPMENT OF DISEASE: CONTROL
DEVELOPMENT OF DISEASE: TREATMENT
DIAGNOSING RHEUMATOID ARTHRITIS

As seen in the active simulation, one of the most important cytokines released by Th17 cells is IL-17, which promotes bone erosion through osteoclasts. However, there are many IL-17/Th17-mediated autoimmune diseases.

Other potential markers for rheumatoid arthritis include:

• Rheumatoid Factor (RF)
  • Occurs in ~80% of people with RA, but also present in other inflammatory diseases
• Anti-CCP antibodies
  • Occurs primarily in people with RA (stronger indication)

These markers can be used alongside other tests to diagnose someone with rheumatoid arthritis.

• How are these markers found?

https://www.practicalpainmanagement.com/sites/default/files/imagecache/lighbox-large/images/2015/11/19/Rheumatoid%20Arthritis%20Diagnosis.jpg
ENZYME-LINKED IMMUNOSORBENT ASSAY

The enzyme-linked immunosorbent assay (ELISA) is used to detect the presence of an antibody or antigen in a particular blood sample.

• Presence of antibody can be used to identify allergies or serious infections
• Presence of antigen can be used for drug testing, pregnancy tests (hCG hormone)

In the case of rheumatoid arthritis, the ELISA technique can be used to look at many markers, including IL-17, rheumatoid factor, and anti-CCP antibodies.
TYPES OF ELISA

• Direct ELISA
• Indirect ELISA
• Sandwich ELISA

To compare Direct and Indirect ELISA: watch this animation and take the quiz.

Sandwich ELISAs are considered to be very effective because they have high sensitivity and specificity.

• Two antibodies (capture antibody and detection antibody) must interact with different epitopes of the antigen
• Possible experiment using rats as an animal model:
  • Coat plate with antibody to rat IL-17
  • Put healthy and diseased rat serum in wells
  • Put secondary antibody for IL-17, conjugated with enzyme
  • Put in substrate to produce color change
  • Determine if concentration of IL-17 is higher in diseased rats compared to healthy rats

ELISA ANALYSIS

The reaction of the enzyme binding to the substrate produces a color change which can be analyzed **qualitatively or quantitatively**.

Qualitatively, it is possible to see large differences in color production to determine the presence or absence of the target antigen/antibody.

Quantitatively, a special machine can be used to determine the optical density of each sample. Researchers use a standard curve to relate the optical density to antibody titer.
DIAGNOSING RA: ANTI-CCP ELISA

We will perform an indirect ELISA to test patient’s serum samples for anti-CCP.

Our simulated reagents for this lab activity include:

- Purified CCP (cyclic citrullinated peptide)
- Serum samples from patients (potentially containing anti-CCP antibodies)
- Anti-human immunoglobulin antibodies w/conjugated HRP enzyme
- TMB substrate
- Serum containing anti-CCP antibodies
- Serum from an unaffected person

http://www.sinobiologicalcdn.com/styles/default/images/pdyimg/ELISA/how_to_select_a_high_quality_elisa_kit_/u24.png