Lecture 10: Fibrous Proteins, Domains, Disulfide Bonds, Immunoglobulins

- Globular versus fibrous proteins.
- How disulfide bonds stabilize proteins
- Quaternary structure of antibodies

Non-globular proteins (Fibrous)
- Often stabilized by similar interactions as globular proteins
- Often play a structural role, e.g. collagen

Domains (Motifs): Segments of proteins that generally fold as independent units. Each domain may have a specific function, e.g. binding to DNA - “DNA binding motif”.

Disulfide Bonds: The formation of a covalent disulfide bond between two cysteine (Cys) residues can contribute to the stability of protein tertiary structure. The "S-S" bond covalently crosslinks two regions of the structure that may be distant in sequence, but nearby in the folded state.

Disulfide bonds are generally found in proteins that function outside of the cell, e.g. extracellular enzymes, antibodies, etc. They stabilize the folded (native) state of the protein.

Overview of the Immune System

- High diversity: ~ $10^6$ different “things” (protein/carbohydrate) can be recognized
- High specificity: Usually a thing is recognized by only one molecule of the immune system.
- Exclusion of self: You do not recognize your own tissues (self-tolerance)

Some Terms:

- **Antigen**: Foreign material that is recognized by the immune system. An antigen is usually a protein, or carbohydrate, but may be a small organic molecule (hapten), or a lipid, or nucleic acid.
- **Epitope**: Region of an antigen to which the antibody binds, e.g. part of a protein.
- **Hapten**: A small chemical that is recognized by an antibody, but cannot generate an immune response by itself.

Cellular Immunity:

- **Key point**: B-cells have an antibody as part of a cell surface receptor. After binding antigen they are activated by T-helper cells (T_h) and the activated B-cell develops into plasma cells that produce soluble antibodies that destroy pathogens.

Antibodies – Y-shaped molecules that have:
- two specific binding sites for antigens (e.g. surface protein on pathogen)
- domains (Fc) that lead to biological effects, such as the destruction of the pathogen.
**Applications of Antibodies.**
- **Immune system:** Antibodies bind to pathogens (bacteria, viruses), leading to their inactivation/destruction.
- **Cancer Treatment:** Antibodies specific to cancer cells can cause the destruction of cancer cells.
- **Protein purification:** Antibodies specific for a protein can be used to purify that protein.
- **Labeling components of a cell:** Antibodies with attached fluorescent groups can be used to localize proteins within a cell.

**Quaternary structure:**
- 2 Light + 2 heavy chains.
- Two binding sites/molecule.
- Chains held together by disulfide bonds and non-covalent forces.
- Light chains are identical on any given Ab. (There are two possible types of light chains, kappa and lambda.)
- Heavy chains are identical on any given Ab. (There are four major classes of heavy chain, IgM, IgG, IgE, IgA, each with a specific biological function, e.g. killing of pathogens.)
- The first 110 residues in each chain are highly variable. The variable region of both heavy and light are responsible for binding antigen. The sequence of these differ from one antibody to the next, resulting in a different specificity.
  - Estimated $\sim 10^3$ different possible light chain $\kappa$, sequences.
  - Estimated $\sim 10^6$ different $\lambda$ sequences
  - Any light chain is can be paired with any heavy chain, giving $\sim 10^8$ different sequences & specificities.

\[
10^3 \times 10^5 = 10^8 \text{ different } \text{Ab}
\]

**Super-Secondary Structure:** Antibodies consists of domains, called Ig folds or Ig motifs, that are linked together by peptide bonds to from the entire chain. These are two beta-sheets held together by disulfide bonds. Light chain = 2 Ig domains ($V_L + C_L$). Heavy chain = 4 Ig domains.
Primary structure.

- V-region, ~110 amino acids, different sequence between antibodies with different specificities.
- Hypervariable regions. 3 per V region, highly different sequences between antibodies with different specificities. The hypervariable loops are also called complementary determining regions (CDRs).
- Constant region - conserved sequence on all antibodies. 110 residues on L-chain, 330 residues on H-chain.

**Antibody Fragments:**

- **Fab Fragment** = light chain + ⅓ of heavy chain. Binds one antigen.
- **Fv Fragment** Variable regions from the heavy and light chain. The Fv domain is the smallest unit that can bind antigen. Useful for cancer therapy due to better penetration into tumor.
- **Fc Fragment** = 2nd part of heavy chains.

**Blood transfusions & Blood Type (In case you wondered):**

- Blood group antigens are carbohydrates on red blood cells (RBC).
- Can have type A, type B, both (AB) or neither (O), depending on genes.
- You have antibodies against other blood types circulating in your blood, but you do not have antibodies against your own blood groups, e.g. type A has anti-B antibodies.
- Antibodies will bind to red blood cells that enter during a transfusion with the wrong blood type, leading to severe problems, such as blocking of capillaries.

**Universal donor** – type O – no antigens on RBC, no antibody binding.

**Universal acceptor** – type AB – no antibodies to bind to the incoming red blood cells.

Try for fun: [http://www.nobelprize.org/educational/medicine/bloodtypinggame/](http://www.nobelprize.org/educational/medicine/bloodtypinggame/)