This exam consists of 14 questions on 5 pages for a total of 100 points. Use the space provided or the back of the previous page, allot 1 min/2 pts. On questions with choices all of your answers will be graded and you will be given the highest score. *Well labeled* drawings are acceptable answers.

1. (6 pts) There are five principal differences between the innate and acquired immune system: i) speed, ii) presence, iii) specificity, iv) memory, v) adaptiveness. Select any **two** of these and provide examples that illustrate the difference between the two systems.

**Speed** – innate is fast, e.g. inflammation occurs over several hours. Acquired is slow, it takes days to generate a response.

**Presence** – innate system is always there, e.g. complement proteins, macrophages. Acquired has to be induced, e.g. antibody production will not occur until B-cells are activated.

**Specificity** – innate system has low specificity – e.g. recognizing PAMPs such as dsRNA, LPS. Acquired system has high specificity, e.g. antibodies, BCR, TCR.

**Memory** – innate has none, e.g. response is the same regardless of the history. Acquired has memory due to the production of B and T memory cells.

**Adaptiveness** – innate has minor adaptation, e.g. can recruit neutrophils or activate protein production in the liver if necessary. Acquired is capable of modifying the immune response by altering the nature of the antibody, detecting different peptides on MHC, etc.

2. (2 pts) Give one example where activation of the acquired system enhances the innate response.

- Antibodies can activate complement
- Activated T-helper cells can activated macrophages using TNFα
- Antibodies can aid in NK activity via ADCC

3. (5 pts) Briefly discuss either one physical or one chemical barriers associated with the innate system. Your answer should clearly indicate how the barrier protects the host.

**Physical** – skin blocks pathogens from entering, mucus traps pathogens.

**Chemical** – low pH prevents pathogen growth, defensins disrupt the membrane of pathogens, lysozyme degrades bacterial cell wall.

4. (8 pts) Neutrophils, macrophages, dendritic cells, and B-cells all destroy pathogens by essentially the same mechanism. Please answer both parts of this question:

i) Give one example of how receptors enhance the ability of these cells to acquire pathogens.

ii) Briefly describe how the pathogens are killed.

iii) What additional step after killing of the pathogen is important for the acquired system? Are all cells capable of performing this step?

i) Complement receptors on neutrophils, macrophages, dendritic cells. B-cell receptor on B-cells (the complement receptor on B-cells aids in activation, not so much is acquiring the pathogen).

Fc receptors on neutrophils/macrophages/dendritic cells.

ii) Phagosome fuses with primary and then with secondary granules and then the lysosome. The primary granules contain destructive enzymes. The secondary granules produce reactive oxygen species that will kill pathogens. The lysosome has digestive enzymes to complete the fragmentation of the pathogen.

iii) Peptides obtained from the pathogen are presents to T-helper cells on class II MHC. Possible with professional antigen present cells – macrophages, dendritic cells, B-cells.
5. (8 pts) Please answer all of the following parts:
   i) What are the three complement pathways?
   ii) How do they differ in activation?
   iii) In what ways are they similar?

   i) & ii) Alternative – activated by spontaneous conversion of C3 to iC3.
   Lectin – activated by binding of MBL to mannose on surface of pathogens
   Classical – activated by C-reactive protein or antibodies.

   iii)
   All three produce a C3 convertase and then a C5 convertase followed by formation of the MAC complex.

6. (6 pts) Select two of the following proteins and briefly discuss how the absence of this protein (e.g. a genetic deficiency) would affect the complement pathway and the usefulness of the complement system to fight pathogens. Specifically comment on what complexes could not be generated and the roll of those complexes in fighting pathogens.

   C3, D, B, C2, C4, C5, C7, MBL, C1s

   C3 – cannot make C3 convertase of the alternative pathway, not possible to make opson C3b or anaphylatoxin C3a, or C5 converase or MAC complex.
   D – this is used to convert factor B to Bb to make C3 convertase of the alternative pathway. The alternative pathway would not function, but the other two pathways would operate normally.
   B – this is part of the C3 (and C5) convertase of the alternative pathway. The alternative pathway would not function, but the other two pathways would operate normally.
   C2 – neither the lectin or the classical pathway can operate since C2 is part of the C3 convertase.
   C4 – same as C2
   C5 – no C5a would be produced, interfering with inflammation (C5a causes mast cells to degranulate). No MAC complex formed, so it would not be possible to disrupt bacterial membranes.
   C7 – only formation of the MAC complex would be affected. it would not be possible to disrupt bacterial membranes.
   MBL – lectin pathway would not operate, but the other two would operate normally.
   C1s – Classical pathway would not operate since C1s cleaves C2 and C4 after activation.

7. (6 pts) Select any one of the following three membrane bound proteins and describe its role in the complement pathway:
   a) MCP (membrane cofactor), b) DAF (decay accelerating factor), c) CD59.

   What might happen to an individual who is missing these factors?
   All of these are host regulatory factors to protect the host cell from complement activity.
   MCP – prevents factor B (or Bb) from binding to C3b, this will prevent C3 convertase from forming.
   DAF – enhances the decay of C3 convertase of the alternative pathway because it catalyzes the release of Bb.
   CD59 – interferes with the formation of the MAC complex.

   Loss of function for these factors will make the host cell susceptible to opsonization and formation of the MAC complex – causing the release of cell contents.
8. (4 pts) Please do one of the following choices:
   
   **Choice A:** How is the inflammatory response initiated by macrophages. How does the macrophage detect the presence of pathogen and what cytokines does it secrete?
   
   **Choice B:** How do cells detect that they are infected by dsRNA viruses? What cytokines do they secrete?

   **Choice A:** Bacterial lipopolysaccharides are recognized by toll like receptor 4 on the surface of the macrophage, the activated macrophage will secrete IL-1, IL-6, IL-8, TNFα.

   **Choice B:** The dsRNA will activate TLR3 in the endosome, causing the release of INFα/INFβ.

9. (12 pts) Please do all of the following choices:
   
   i) What aspects of the inflammatory response are useful in fighting pathogens (6 pts)
   
   ii) State one additional mechanism (besides macrophages) that initiates the inflammatory response (1 pt)
   
   iii) Select one of the following molecules and state which cell produces it and how a deficiency in that molecule will affect the local inflammatory response: IL-8, IL-1, TNFα, Histamine, MIP-1β (3 pts)
   
   iv) Under what conditions is the inflammatory response detrimental to the host? (2 pts)

   **Bonus:** How might you treat the condition that is described in part iv of the above question (2 pts).

   i) Complement proteins and neutrophils are brought to the site of the infection to enhance complement and to provide effective killing of pathogens by neutrophils. If the acquired system has engaged, antibodies and Tcells will also enter the site.

   ii) a) Binding of C3a and C5a to mast cells causes them to degranulate and release histamine.
       
       b) Tissue damage activated bradykinin.

   iii) IL-8 – if absent it cannot be induced on the endothelium surface, so neutrophils will have a hard time slowing down since binding to the IL-8 enhances the affinity of the neutrophils to adhesion molecules. Will also not have chemotaxis of the neutrophil to the site of the infection from IL-8 from macrophages.

   IL-1 – produced by macrophages, local effect is that endothelium cells will not be activated to produce IL-8, difficulty recruiting neutrophils. Also involve in acute inflammation, so there will be reduced synthesis of complement proteins form the liver.

   TNFα – produced by macrophages. Also will reduce the expression of adhesion molecules on the surface of the endothelium, making it more difficult for neutrophils to enter.

   Histamine – produced by mast cells. reduction in permeability will reduce fluid flow into the inflamed area, reducing the influx of proteins and cells.

   MIP-1β - produced by neutrophils, recruits additional macrophages to the area.

   iv) If large numbers of the pathogen becomes systemic, a large number of macrophages in the spleen and liver will produce large amounts of TNFα, raising permeability in all vessels, leading to the loss of a large amount of fluid from the blood, leading to organ failure and death.

   **Bonus:** Somehow inactivate TNFα – perhaps using an antibody against it or an antagonist against its receptor
10. (8 pts) Both NK cells and T_{CTL} are responsible for killing either virally infected cells or cancer cells by inducing apoptosis. Please do one of the following choices:

- **Choice A:** In what way are NK cells and T_{CTL} cells similar in their recognition of the target cells, in what way are they different?
- **Choice B:** Describe one method by which NK and T_{CTL} cells kill infected/cancerous cells.
- **Choice C:** Give two reasons why death by apoptosis beneficial from an immunological point of view.

**Choice A:** Both recognize MHC I on the surface of the cell. NK cells respond to just the MHC I, irrespective of the bound peptide. Low levels of MHC I will cause the NK cell to kill. Killing in the presence of normal levels of MHC require high levels of an activation protein on the surface of infected cells. T_{CTL} recognize the MHC I-peptide complex with high specificity.

**Choice B:**
- Perforin/granzymes – perforin makes a pore in the membrane, allowing granzymes to enter. These are proteases that activate caspases, causing the cell to enter apoptosis.
- FasL/Fas. FasL is a trimer on the surface of NK/T_{CTL} cells. It induces Fas on the target cell to trimerize. This activated the DEAD domain inducing apoptosis.

**Choice C:**
- Prevents virus from spreading
- Keeps intracellular self-antigens hidden from the immune system, reducing the risk of generating an autoimmune response.
- Membrane blebs can be taken up by macrophages, and peptides from them can be presented on class II MHC.

11. (9 pts) Briefly describe the life of a B-cell, from its birth to death. Your answer should include the following: Where is it born, what are the important checkpoints in its development, where does the checkpoint happen, what is the fate of B-cells that are not activated, what is the fate of a B-cell that is activated.

Stem cell in bone marrow, check for self-tolerance, lymph node, 2^{nd} check for self tolerance, lymph node (or other secondary organ), binds to pathogen, activated by T-helper cells, proliferates, generates Bmemory and antibody producing plasma cells.

If not activated by T-helper cell, dies in a few days.

12. (12 pts) Describe the activation process of either B-cells or T_{C} cells. Your answer should include:

i) the role of the lymphatic circulatory system in enhancing the immune response.

ii) the role of secondary lymphoid organs in enhancing the immune system.

iii) any additional cells, and cell surface proteins involved in the process.

i) lymphatic circulatory system will ensure a B or Tc cells will see antigens in different parts of the body, enhancing the probability of encountering an antigen (peptide – MHC) that they can recognize.

ii) Both B and Tc cells require assistance from T-helper cells. In the case of B-cells – direct recognition of MHC II on B-cell, in the case of Tc cells, a T-helper has to either license a dendritic cell by interaction with MHC II or become activated by a proAPC presenting on class II. The likelihood of this occurring is enhanced by having a high concentration of B and T-helper cells in the secondary organs.

iii) B-cell activation – foreign peptides presented on class II are recognized by TCR on T-helper cells, leading to activation of the B-cell. TCR is specific for class II due to CD4 on the surface of the T-helper cell.

Tc- activation – must recognize MHC I-foreign peptide using its TCR in a highly specific manner. Class MHC I recognized by CD8 on the surface of the Tc cell. Secondary activation signal required – either IL-2 from nearby activated T-helper cell, or b7-CD28 interaction on a licensed dendritic cell.
13. (6 pts) Compare and contrast the structure of class I and class II MHCs and their interaction with T-cells.

Both structures are similar – membrane bound with a peptide binding domain composed on a pair of alpha helices.
Peptide – Class I MHC are recognized by TCR on Tc cells, specific for class I due to CD8 on Tc cell.
Peptide – Class II MHC are recognized by TCR on T-helper cells, specific for class II due to CD4 on T-helper cell.

The recognition by the TCR is specific for a particular peptide on a particular MHC.

14. (8 pts) Please do **one** of the following choices:

**Choice A:** Describe the peptide-binding properties of MHCs and why these properties are beneficial to the immune response.

**Choice B:** Describe the level of diversity of MHC molecules on cells in an organism and the relationship between diversity and the ability to fight pathogens. Given an example.

**Choice A:**
There is relatively low specificity, allowing one MHC to present a number of different peptides.

**Choice B:**
A number of different MHC molecules are present on each cell and each cell would be identical. Each MHC will present different peptides. This would enhance the probability that the antigen presenting cell would find a T cell with the appropriate TCR to recognize that MHC and activate the presenting cell.

A more diverse MHC (heterozygous alleles) enhances the ability to fight viral infections.

**Bonus (2 pts).** Individuals who are treated with large doses of antibiotics can develop a serious intestinal infection by Clostridium difficile (C. diff) that can be come life threatening in some cases. C. diff is normally found in the human intestinal system. Why do you think this occurs after antibiotic treatment and can you suggest a possible treatment that would enhance recovery?

The C.diff is competing with the normal microbial flora. These are killed by antibiotic treatment, allowing C. diff to grow unchecked.

This can be treated by re-introduction of commensal bacteria, e.g. as in food with active cultures. Fecal transplants have also been used with greater success.