Medical Microbiology (BI 158)
Spring 2001
MIDTERM EXAM

This is a 3 hour exam covering Basic Principles of Immunology and Virology. This exam is worth 50% of your grade.

MULTIPLE CHOICE FORMAT (1 point each)
Each item or incomplete statement in this section is followed by answers or by completion of the statement. Circle the one lettered answer or completion that is best in each case.

1. You are a physician in the emergency room at Rhode Island Hospital. A young girl, of about 6 years of age, has stepped on a rusty nail. She is not certain that she has ever been vaccinated for protection against tetanus toxin. Based upon what you have learned, which of the following would you do?
   A. Give her IL-5.
   B. Send her home without treatment.
   C. Immunize her with the toxoid and give her passive antibody against tetanus.
   D. Immunize her against influenza.
   E. Immunize her with the toxin only.

2. All of the following are true about CD4 T cells EXCEPT that they
   A. are part of adaptive immunity.
   B. can help B and other T cell responses.
   C. respond to antigenic peptides presented by MHC class II molecules.
   D. are major mediators of cytotoxicity against virus-infected host cells.
   E. can make IFN-γ.

3. Which of the following require the thymus?
   A. Antigen Processing and Presentation of Materials from a Peripheral Infection
   B. B Cell Differentiation
   C. Rearrangement of Genes for the T Cell Antigen Receptor and T Cell Maturation
   D. Rearrangement of Germline Gene Segments to Code for Variable Portions of Antibody Molecules
   E. Differentiation of Granulocytes
4. All of the following are important functions of TNF EXCEPT that it
   (A) is an important growth and proliferation factor for T cells.  
   (B) promotes MHC class 1 expression. 
   (C) contributes to systemic vascular collapse during septic shock conditions. 
   (D) induces resistance to viral infections. 
   (E) is part of innate defense mechanisms. 

5. IL-12 is important for which of the following?
   (A) CD8 Expression 
   (B) IFN-γ Production 
   (C) TNF Production 
   (D) Eosinophil Differentiation 
   (E) CD8 Expression 

6. Dendritic type cells can do all of the following EXCEPT
   (A) secrete IL-12. 
   (B) process and present antigen to CD4 T cells. 
   (C) produce IL-12. 
   (D) express co-stimulatory molecules. 
   (E) travel from the skin to the lymph node. 

7. Upon his return from South America, an otherwise normal 12 year old boy is found to be
   infected with helminth parasites. He is doing well because of an endogenous Th2 type cytokine
   response. Which of the following would you NOT expect to see?
   (A) IgE 
   (B) IL-4 
   (C) IL-5 
   (D) Eosinophilia 
   (E) CD4 T cell IFN-γ Production
8. An individual is genetically deficient for the IFN-α/β receptor. Which of the following would you expect to be blocked?
   A. Immunoglobulin Gene Rearrangement
   B. Cell Surface Expression of A Functional T Cell Receptor for Antigen on Mature T Cells
   C. IgM Antibody Responses
   D. Antiviral Defense
   E. NK Cell Development.

9. All of the following components can be part of innate immune responses EXCEPT
   A. alternative pathway of complement activation.
   B. natural killer cells.
   C. B cells.
   D. MAC.
   E. macrophages.

10. A SPECIAL function of IgA is to
    A. opsonize bacteria.
    B. be secreted into mucus.
    C. activate complement.
    D. cross the placenta.
    E. facilitate ADCC.

11. Which of the following can NOT be found in the spleen?
    A. white pulp
    B. red pulp
    C. marginal zones
    D. cortex
    E. T cells

12. The proteasome is
    A. a component of phagolysosomes.
    B. stored in the lytic granules of cytotoxic CD8 T cells.
    C. associated with the invariant chain (II).
    D. the Cuisinart™ equivalent of antigen processing and presentation to CD8 T cells.
    E. secreted into the blood.
13. The Thoracic duct
   A. was a major aqueduct in ancient Rome.
   B. facilitates transfer of maternal antibody to the fetus.
   C. is part of the air venting system in the BMC.
   D. is the Dendritic cell port of entry for travel from the periphery to the lymph node.
   E. is the lymphocyte port of entry to the blood for the lymphatic system.

14. The signal transducers and activators of transcription (STAT) molecules are important for which of the following?
   A. Cytokine Binding By Cells of the Immune System
   B. Activation of Apoptosis in Response to TNF
   C. Promotion of Gene Expression Following Cell Exposure to Particular Cytokines
   D. Release of Cytotoxic Granules from CTL or NK Cells
   E. Activation of Erythrocytes

15. Essential steps in cytolytic destruction of target cells by either CTL or NK cells include all of the following EXCEPT
   A. delivery of a lethal hit
   B. positive stimulation of cytotoxic cell by target cell
   C. release of IFN-γ from the cytotoxic cells
   D. deposition of perforin on the target cell surfaces
   E. release of granzymes from the cytotoxic cells
SHORT ANSWER FORMAT
(3 points each)

1. In a major heavy weight fight, one boxer (boxer A) bites the ear of his opponent (boxer B) and a lot of blood gets spread all over both players. How would you determine if either boxer was infected with HIV as a result of the incident?

The standard test for HIV is: to test for Ab against HIV using an ELISA (enzyme-linked immunosorbent assay) - where p24 viral Ag is bound to the bottom of the well and then the patient's serum is added to the bottom of the well and then the patient's Ab is added. The 1st Ab will only be absorbed by the Ag which is linked to an enzyme that changes color when a substrate is added. This 2nd Ab will only bind to the patient's anti-HIV Ab (which bound to the Ag on the bottom of the plate) - forming a "sandwich". After a substrate is added, the color change is measured (and can be compared to std curve). This ELISA test will tell you if either of the patients have Ab against HIV - if the test is negative, usually a western blot is done to be certain the patient is HIV positive. The test must be performed right after the incident on both parties and again 2-8 wks later (Ab levels might be too low to detect right away).

2. Explain delayed type hypersensitivity (DTH). How might you expect to induce it? What components of the immune system are required?

DTH is a type 4 hypersensitivity reaction, which is the result of APCs presenting Ag (by MHC II) to a previously sensitized CD4+ T cell. The first time it encounters the Ag, the CD4+ T cells differentiate into Th1 cells that produce IFN-γ and other cytokines, which stimulate macrophages to become more phagocytic and produce more free radicals and therefore damage the surrounding tissue. An example of DTH is the PPD test for TB where if TB is injected into dermis and get DTH reaction. A characteristic feature of DTH is that it takes several days to develop (unlike immediate hypersensitivity where reaction is immediate). A DTH can be induced if Ag that the system has already been sensitized against and if the macrophages (or T cells) that phagocytosed the Ag is presenting Ag on MHC II to CD4+ T cells. The APC has activated CD4+ T cells which causes IL-12 to induce Th1 response. (The APC has costim. T cell which interacts with CD28 on T cell) Both the innate immune system and the adaptive immune system are required (cell-mediated specifically - not humoral immunity).
3. Name three major functions of NK cells:
   1. NK produce IFNγ which stimulates macrophages to be more active, phagocytic, more macrophage ROS production, etc.
   2. NK cells can mediate Ab-dependent cell mediated cytotoxicity (ADCC) when their Fc receptors bind IgG directed to Ag, causing them to degranulate and “kill” the source of the Ag (i.e., bacteria)
   3. NK kill virus-infected cells by secreting perforin and granzyme, secreting TNFα and have Fas ligand, which induce infected cell to die via apoptosis. They have MHC I receptors that inhibits them from killing hosts own cells via an ITAM domain that prevents all from degranulating and they also have receptors with ITAM domains that cause cell to degranulate.

4. Which immune system components are most important for protection during secondary exposure to influenza virus?
   During a second exposure to influenza, the most important part of the immune system needed is IgA, which is in the mucus secretions of nasal passages (influenza is transmitted through resp. path). Ig A can neutralize the viruses in the mucus and prevent them from causing infection. After the 1st infection it is important to make B & T memory cells that can help create a quick, specific, and increased immune response. The B memory cells will produce lots of Ab in response to Ag (IgG & IgA, etc) that are very specific for that strain of influenza (Ab has undergone affinity maturation already). The memory T cells are needed to stimulate/help B cells. Th2 cells will make cytokines (IL-2, IL-4, IL-5, etc.) and provide co-stimulation (by increasing co-receptor which binds to CD40 on B cell) → stimulating B cells to proliferate, differentiate, AND type switch (for example IL-2 causes IgA) and make lots of Ab.
5. An individual has a mutation severely inhibiting the natural function of the transporter associated with antigen processing (TAP). Which antigen processing pathway would be rendered inefficient? What classes of agents would the individual have a problem attacking? Why?

MHC-I/Ag processing pathway would be inefficient because TAP is used to allow intracellular proteins that have been ubiquitinated and then degraded by proteasome to enter into the ER. These proteins then combine with the newly synthesized MHC-I peptide and β2 microglobulin to form a stable molecule which is delivered to cell membrane (Golgi). MHC-I on APC is used to present to CD8+ T cells.

This individual could therefore not attack intracellular organisms, such as intracellular bacteria and intracellular parasites, not viruses, because the cells infected would not be able to present these Ag to CD8+ T cells causing the infected cell to die due to the organism (at the same time). However, viruses could be fought off via MHC-II presentation to CD4+ T cells; NK cells; B cells not T cells against the actual virus Ag. The MHC-I pathway could present products made by virus.

6. Describe the two classes of interferons and how they mediate anti-microbial effects?

IFN type I (α/β) are made by virus infected cells and macrophages, they are produced in response to a virus, RNA in the infected cell and induce an anti-viral state in uninfected cells by causing them to decrease protein synthesis (the virus cannot replicate in these cells). IFN also activate NK cells to kill virus infected cells. IFN-I can also increase MHC-I expression allowing for more Ag presentation to CD8+ T which can differentiate into CTL's and kill infected cells.

IFN type II (γ) is known as IFN-γ, which is made by NK cells, CD4+ T - CD8+ T cells. IFN-γ increases MHC-I and II expression so more Ag can be presented to CD8+ T cells. IFN-γ increases MHC-I II expression so more Ag can be presented to CD8+ T cells. IFN-γ causes CD4+ T to differentiate into TH1 which then make more IFN-γ and can activate macrophages to increase phagocytosis and production of enzymes that free radicals to kill bacteria. IFN-γ also cause B cells to proliferate/differentiate and let type switch to IgG. IgG can help opsonize, activate complement, and neutralize the Ag.
7. Describe the steps for immunoglobulin heavy chain class switching from IgM to IgG. Are the events antigen dependent? Do they always result in functional products? What are the functional consequences of successful completion to the host?

In order to class switch from IgM to IgG the constant part of the heavy chain must be changed, but the VDJ region (variable/Ag specific part) are not changed (the light chain, too, is left alone). IgM is changed to IgG under appropriate cytokine stimulation (for example IFN-γ).

- The DNA is cleaved in the introns of the constant part (as explained in diagram), and internal sequence is lost so cannot go from IgG-γ to these events are Ag dependent, because the Ag causes different cytokines to be made in response which then cause isotype switching.
- They always result in functional products (unless there are some weird mutations of Cμ).
- Consequences of IgG formation allows better neutralization of Ag, it can cross the placenta (if pregnant to give fetal passive immunity), IgG forms smaller complexes than IgM (Ig 6 is a monomer vs. Ig M is pentamer) and IgG can access different effector functions (ex: ADCC), allowing for better attack/kill ability of certain organisms.
1. (2 pts) Define CONGENITAL INFECTION and name TWO viruses that are transmitted by this route.

A congenital infection is an infection that can cross the placenta and be transmitted from mother to fetus so the fetus is born with the infection (or dies from it).

- HIV-1 (lentiviruses, retroviridae)
- Rubella (rubiviruses, Togaviridae)

2. (1 pt) Which of the following classes of antibody would best be associated with protection from a congenitally acquired infection (CIRCLE ONLY ONE)

- IgG
- IgM
- IgD
- IgE
- IgA

3. (2 pts) Define NEONATAL INFECTION and name TWO viruses that are transmitted by this route.

A neonatal infection is one that is transmitted from the mother to the fetus during the birth or right after birth (i.e. infects newborns)

- HSV-2 (genital herpes) → α Herpesviridae
- RSV (pneumoviruses - Paramyxoviridae)
4. (2 pts) Define active versus passive immunization and give two examples of a situation involving virus infections where both types of immunization would be administered to the patient.

Active immunization is when a less virulent form of the antigen is given so that the patient develops "memory" and can fight off the infection when it reappears. Passive immunization is when given antibodies against the virus to help fight off the infection—this does not cause "memory" to be induced so would be able to fight off the disease at a later point.

1. If infected with rabies and suspected to have been bitten by a rabid animal, get both rabies Ig and the vaccine.
2. If infected with Hepatitis A can get Ig & against the virus and also a vaccine is available if known to have been exposed.

5. (4 pts) During your pediatric clinical clerkship you encounter the following situation.

A mother and her infant daughter (SARAH) have just come into your office for the purpose of getting a polio vaccine. The mother wants an explanation of the LIVE-ATTENUATED POLIOVIRUS vaccine. Explain the essential properties of the vaccine that you feel the needs to know.

Live attenuated polio (Sabin) vaccine is given orally and is less expensive than the Salk vaccine. It will help her daughter not to get polio which is very bad—can cause her to be crippled or even cause her brain to die. This vaccine should not harm Sarah in any way unless she is immunocompromised or has a family member who is. Sometimes the cells used to grow the virus and attenuate it can have other viruses in them and therefore, if she is immunocompromised she could get this other virus. The vaccine will also let her gain gut immunity so if fecal oral transmission she can immunize her little friends in the neighborhood.

After your explanation she informs you that her older son has AIDS and has recently moved back into the family home for care and support. Does this concern you, and if so, how will you respond.

Yes—as explained above—this could cause her brother to become infected with another virus carried in the attenuated cells used to attenuate the virus and this would be bad because his immune system cannot fight it off. He also might react to the the attenuated virus and get sick.

Therefore, Sarah should not get the Sabin vaccine (she could get the Salk one, though).
6. (2 pts) List or describe the main feature(s) that distinguish a capsid from an envelope.

The envelope comes from the host lipid membrane (cell, nuclear, endosomal membranes) and the only part that is encoded by viral genome in the envelope are the glycoprotein "spikes." A capsid on the other hand is a protein shell surrounding the viral genome which is coded for by the virus.

7. (1 pt each) The initial goal of all viruses is to produce mRNA.

Describe how each of the following viruses accomplish this goal (be sure to include relevant enzymes when appropriate)

**Hepatitis A**

Hep A is **+**ssRNA so already mRNA and just needs to be translated.

**Rotavirus**

- **+**ssRNA cannot use **+** strand already has
- **-**ssRNA
  - RNA polymerase
  - pick **+** strand
  - make **+** strand (mRNA)

**Respiratory Syncitial Virus**

- **+**ssRNA
  - RNA polymerase
  - make ssRNA (mRNA)

**HTLV-I**

- **+**ssRNA
  - reverse transcriptase
  - make ds DNA intermediate
  - integrate
  - DNA polymerase
  - mRNA
8. (1 point each) The first step in the life cycle of any virus is attachment to specific receptors on the surface of susceptible cells. Name the relevant receptors and co-receptors when appropriate for each of the following viruses.

HIV-1 (T-cell tropic strains)  
- CD4
- CXCR4 co-rec.

HIV-1 (macrophage tropic strains)  
- CD4
- CCR5 co-rec.

Influenza A  
- HA receptor (Sialic acid)

Poliovirus  
- AChR at NMJ

Rhinovirus  
- ICAM-1

Rotavirus  
- Receptors on M cells of GI tract
9. (1 point each) Herpes viruses establish classic latent infections in specific cells and tissues. For each of the HERPES viruses listed name the cell or tissue in which latency is established.

EBV - B cells

HSV-2 - neurons (DRG)

VZV - neurons (DRG)

CMV - monocytes

10. (2 pts) What do each of the following assays measure?

- Hemagglutination assay - measures presence of virus

- Hemagglutination inhibition assay - measures presence of Ab (against the virus)

11. (1 pt) Name one virus that can be titrated using either of the above assays.

Influenza A (Orthomyxoviridae)
12. (4 pts each) For each of the viruses listed describe:

1. Mode of transmission, 
2. Disease association 
3. Target tissues associated with disease 
4. Methods of control/prevention/or treatment

Hepatitis A

1. Mode of transmission, 
   - fecal/oral and blood

2. Disease association 
   - acute hepatitis

3. Target tissues associated with disease 
   - liver

4. Methods of control/prevention/or treatment
   - control:
     - washing hands to prevent fecal/oral transmission
     - do not eat raw seafood or contaminated food
   - prevention: vaccine if go to endemic area
   - tx: pooled IgG against HVS within 2wks of infection
Poliovirus
1. Mode of transmission, **faecal/oral**
2. Disease association, *polio causes spinal and bulbar encephalopathy (damages CNS)*
3. Target tissues associated with disease CNS (motor neurons in spinal cord and medulla)
4. Methods of control/prevention/or treatment
   - **vaccination**
   - If not vaccinated - wash hands well & don’t share drink with infected person

HSV-2
1. Mode of transmission - sexual / vertical / contact
2. Disease association genital herpes
3. Target tissues associated with disease skin (epidermis) of genital area and can be latent in DRG.
4. Methods of control/prevention/or treatment
   - **tx**: acyclovir
   - prevent/control - do not touch/have sex w/ infected person
RSV

1. Mode of transmission, respiratory

2. Disease association: pneumonia in < 6 months (lower resp tract infection) in infants

3. Target tissues associated with disease: lungs

4. Methods of control/prevention/or treatment:
   - Rx: Ribavirin
     Control: Wash hands good between taking care of patients in ped. ward and isolate kids who have it so don’t give it to infant next to him.

Influenza A

1. Mode of transmission, respiratory/contact

2. Disease association: flu (fever/malaise/sore throat/headache, diarrhea, runny nose)

3. Target tissues associated with disease: respiratory tract (oronasal epithelium)

4. Methods of control/prevention/or treatment:
   - Washing hands well
   - Avoid being around people who have it
   - Tx with Amantadine prophylactically
     + Vaccinate
1. Mode of transmission, via a tick bite

2. Disease association - fever + encephalitis
   (Colorado tick fever)

3. Target tissues associated with disease - brain

4. Methods of control/prevention/or treatment:
   - avoid tick bites - stay away from squirrels
     who have the ticks (at west)
   - wear protective clothing
   - no tx once get it.

Rabies virus

1. Mode of transmission, via bite from infected animal

2. Disease association - coma, death
   - paralysis (focuo) + furious - bite, violent
   - thirsty
   - hydrophobic

3. Target tissues associated with disease - neurons/CNS
   salivary glands

4. Methods of control/prevention/or treatment:
   - avoid getting bit (stay away from wolves, bats, dogs, etc.)
   - rabies vaccine + IgG if do get bit
13. (1 pt each) Describe the mechanism of action of the following drugs and name the virus for which they are indicated.

Acyclovir - For HSV-2 (herpesviridae)
Acyclovir only is active after phosphorylated by thymidine kinase (herpes enzyme) and then phosphorylated 2x more by other kinases than it is active and incorporated as a nucleotide by DNA polymerase and causes chain termination = inhibiting DNA synthesis or severe CMV (cytomegalovirus, herpesviridae). Same mechanism as acyclovir except is more toxic. = Stops DNA synthesis in viral infected cell (causes chain termination)

Zidovudine (AZT) (lentiviridae)
AZT is a NRTI. It gets incorporated into the growing DNA strand by RT and causes chain termination. (blocks replication)

Nevirapine
For HIV 1/2 - It is a NNRT - inhibits RT so it is not functioning and cannot reverse transcript DNA to RNA and blocks DNA synthesis = blocks viral replication

Saquinavir - A protease inhibitor (for HIV 1/2)
Inhibits the viral protease dimer (aspartyl protease) from cleaving gag & pol = so make non infectious viruses

Amantadine-HCL
To treat influenza A
It blocks/inhibits the M2 (H+ channel) so virus cannot uncoat

BONUS QUESTIONS:
What virus is responsible for the recent outbreak of foot and mouth disease in the UK (1 point)?
Picornaviridae - (Ovinevirus - Sp.)
What virus was responsible for the death of legendary screen actor, Rock Hudson (1 point)?
HIV-1 or 2 (he died of AIDS)