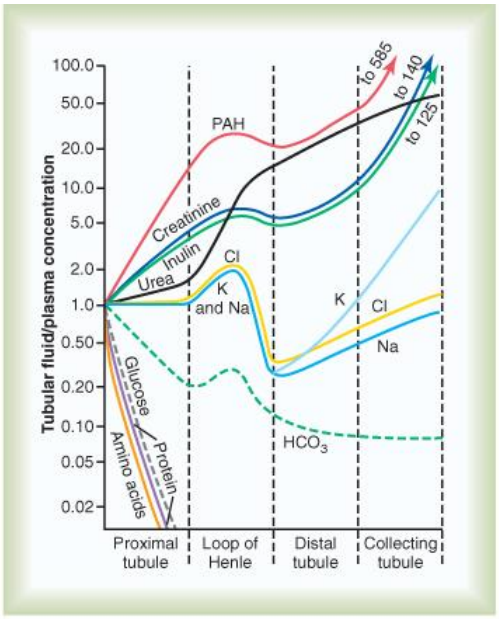
Exam KEY

NROSCI/BIOSC 1070 and MSNBIO 2070
Exam # 3
November 17, 2017

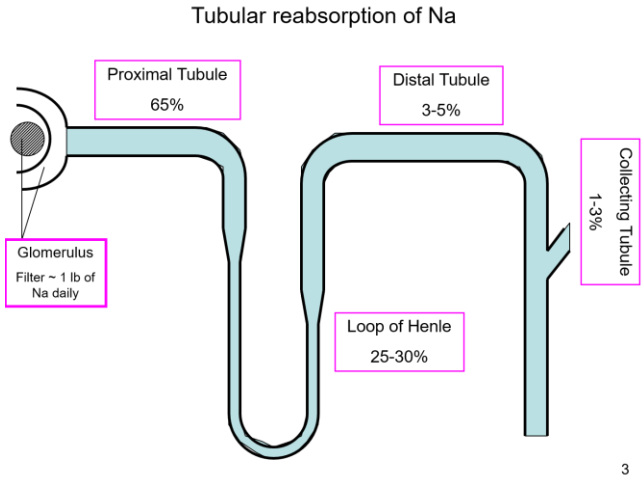
Total POINTS: 100 20% of grade in class

1) Draw a rough sketch of a nephron ***(2 points)***.

Then, for each major segment of the nephron, note the Na^+ concentration and the approximate percent of the filtered load of Na^+ that remains in that segment ***(8 points)***.



© Elsevier. Guyton & Hall: Textbook of Medical Physiology 11e - www.studentconsult.com



NOTE: they need to be clear that in the collecting tubule the concentration of Na+ can vary tremendously based on ADH

2) Drugs the block aldosterone receptors in the nephron and drugs that block the Na^+-K^+-$2Cl^-$ transporter in the nephron are diuretics.

a) Explain how each of these drugs acts to produce diuresis. ***(5 points)***.

Aldosterone promotes Na+ reabsorption in the collecting tubule (via upregulation of Na+/k+ pumps and ENaC) (1 point) and blocking that reduces Na+ reabsorption and the excretion of Na+ pulls water along with it (1.5 points).

Loop diuretics inhibit active reabsorption of solutes (especially sodium) in the ascending limb of the loop of Henle (1 points). This eliminates the osmotic gradient for water reabsorption in the deep renal medulla therefore increasing Na+ and water excretion and impairing the kidney's ability to concentration urine (1.5 points).

b) Which of the two drugs would provide a larger diuresis? Why? ***(5 points).***

Loop diuretics would provide a larger diuresis (2 points). Loop diuretics prevent sodium absorption in the ascending limb, while aldosterone inhibitors only effect the collecting tubule (1 point). Thus loop diuretics result in great sodium excretion, and therefore greater water loss (2 points).

3) A mutation in nephrin was found to markedly increase filtration fraction.

a) Explain the mechanism through which a nephrin mutation could increase filtration fraction. ***(5 points).***

Nephrin occurs in the slit diaphragm in between podocyte foot processes (2 points). A mutation could increase the filtration coefficient (Kf) by increasing the permeability of the slit diaphragm (2 points), therefore increasing GFR without changing renal plasma flow which increases filtration fraction (1 point).

b) What tests could be run in a renal clinic to confirm your predicted mechanism? ***(5 points).***

Filtration fraction = (GFR) / (Renal plasma flow) (2 points)

GFR can be calculated by measuring clearance of inulin or creatinine (1.5 points)

Renal plasma flow can be calculated by measuring clearance of PAH (1.5 points)

4) ***a)*** What is nephrogenic diabetes insipidus? ***(2 points).***

A lack of renal response to ADH.

b) If a patient with diabetes insipidus were deprived of water for a day, what would happen to each of the following and how would that differ from an individual with normal renal function? ***(2 points each/8 points total).***

Urine volume:

Normal renal function: Low urine volume (obligate water loss is 0.5 L/day)

Diabetes insipidus: Urine volume remains high (20 L/day)

Urine osmolarity:

Normal renal function: High urine osmolarity (1200 mOsm/L)

Diabetes insipidus: Low urine osmolarity (50 mOsm/L)

Plasma volume:

Normal function: Low, but patient is still alive (2.5 L)

Diabetes insipidus: This person is dead, they have little to no plasma volume.

Plasma osmolarity:

Normal function: Slightly elevated, but near normal

Diabetes insipidus: Extremely elevated, the patient would be dead.

5) Compare and contrast the renal handling of glucose in a person with normal blood glucose levels, a person with a blood glucose concentration about twice the normal level, and a person with a blood glucose concentration about 5X the normal level. ***(8 points*)**.

Normal plasma glucose concentration is ~100 mg/100 mL. Normal GFR is 125 mL/min. Glucose is entirely reabsorbed up until the transport maximum of around 375 mg/min (2 points). In a person with normal blood glucose, they are secreting 125 mg/min of glucose, and reabsorbing all of it (2 points). A person with 2X normal blood glucose is secreting 250 mg/min, and reabsorbing all of it (2 points). A person with 5X normal blood glucose is secreting 625 mg/min, reabsorbing 375 mg/min, and excreting 250 mg/min (2 points). Specific numbers are not important, just the general idea.

c) Some drugs used in the treatment of diabetes mellitus partially interfere with glucose transport in the proximal tubule. How would such a drug impact your answer in part A? ***(2 points*)**.

These drugs would decrease the transport maximum of glucose, thus hindering reabsorption and increasing excretion (2 points). For example, the person with 5X normal blood glucose would excrete even more glucose, while the person with 2x normal glucose might begin to excrete glucose rather than reabsorbing all of it. The person with normal blood glucose would likely remain unaffected.

6) Briefly (1-2 sentences) define each of the following immunological terms. (**2 points each).**

a) Recombination activating gene 1 (RAG1):

RAG proteins are enzymes that recognize and bind to recombination signal sequences (RSS) in the genes encoding the variable domain of the TCR and BCR. Once bound, RAG mediates double stranded breaks and works with other proteins to carry out somatic recombination in the TCR and BCR variable regions. Without it you fail to generate adaptive antigen receptors or generate B and T cells.

b) Anergy***:***

T cell anergy is a peripheral tolerance mechanism where T cells, who recognize a peptide/MHC in the absence of co-stimulation, become resistant to future stimulation.

c) Alarmin:

Endogenous molecules that are released from damaged human cells and are recognized by and activate the innate immune system. They are also called Danger associated molecular patterns (DAMPs). Many trigger TLR4.

d) Defensin:

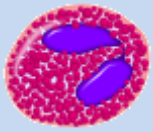
Defensins are small cationic peptides that function as natural antibiotics. They disrupt bacterial cell membranes. They are found on the skin and mucosal surfaces, but also produce by innate immune cells, particularly neutrophils.

Exam KEY

7) The following table is partially completed, showing the sources and effects of three cytokines. Complete the table by filling-in the 5 unshaded boxes ***(1 point each; 5 points total).***

		Effect on:			
Cytokine	**Sources**	**T cells**	**APC**	**Stromal Cells**	**Deficiency Results in**
IL-10	Th2 cells and Treg	Inhibits T cell responses	Limits their stimulatory capacity	X	Autoimmunity
Interferon-γ (IFN γ)	Th1 CTL	Inhibits Th2	Increased CD80-CD86; MHCI and II	Increased MHC I; Antiviral functions	Infections with intracellular pathogens; poor tumor clearance
IL-4, -5, -13	Th2 cells	Inhibits Th1 or Type 1 T helper responses	X	Mucus production; tissue repair	Poor parasite clearance; lack of allergy

8) The following table is partially completed. Complete the table by filling-in the 5 unshaded boxes ***(1 points each; 6 points total).***

Cell Name	**Schematic Representation**	**% of PBMC**	**Pathogen Targeted**	**Granules Phenotype after H + E staining**	**Name *Two* effector mechanisms used**
Neutrophil or Polymorphonuclear leukocytes (PMNs)		60%	Bacteria Fungi/yeasts	Some fine, faintly pink granules	1. NETosis 2. Anti-microbial cytotoxic enzymes including myeloperoxidase (MPO), lysozyme, elastase 3. Defensins 4. Generation of reactive oxygen and nitrogen 5. Phagocytosis
Eosinophil		2%	Parasites	Full of large pink-orange granules	1. Degranulation against large targets that cannot be phagocytosed, especially parasities. 2. Use granules containing Major Basic Protein, Peroxidase and Cationic Protein 3. Phagocytosis

9) Three signals are needed to activate a $CD4^+$ T cell to carry out its effector functions against extracellular bacteria.

a) List these three signals. ***(6 points).***

Signal 1:
Signal 1 is TCR/CD3 signaling induced in T cell by peptide/MHC on APC.

Signal 2:
Signal 2 is CD28 signaling in T cell triggered by co-stimulatory molecules (CD80, CD86, CD40) on APC

Signal 3:
Signal 3 is cytokine signaling in T cell triggered by cytokines secreted by local cells, including APC

b) Briefly explain how antigen-presenting cells (APC) are critical for the delivery of these three signals. ***(3 points).***

Signal 1 can typically be found on APC and other cells of the body. However, only APC that have been activated via their PRRs after they recognize non-self can deliver co-stimulation. Activated APC are also crucial sources of certain cytokines, such as IL-12 and IL-15.

10) Pathogens have evolved mechanisms to avoid detection by the adaptive immune system.

a) Epstein Barr virus (EBV) infects B cells and encodes a protein (EBNA1) that prevents the proteasome from breaking down cytosolic peptides. Why would this limit detection of virally infected B cells by cytotoxic T lymphocytes? ***(4 points)***

The proteasome breaks down proteins into peptides that can be loaded onto MHC class I for presentation on the cell surface. CTL assess the intracellular state of the cell by examining peptides loaded onto MHCI. In the absence of proteasome function, viral peptides would not get degraded and could not be displayed on the cell surface. This would prevent CTLs (CD8+ T cells) from detecting an EBV infected cell.

b) NK cells are another other group of lymphocytes that could aid in the detection of EBV-infected cells if cytotoxic T lymphocyte recognition was hindered. What set of innate receptors would NK cells use to recognize virally infected cells, and what effector molecules would be utilized? ***(4 points).***

Killer inhibitory receptors (KIRs) and Killer activating receptors (KARs): KIRs are expressed by natural killer (NK) cells and regulate the killing function of these cells by interacting with major histocompatibility (MHC) class I molecules. In the absence of MHCI on a virally infected cells, NK cell activity would be released. Virally infected cells could also upregulate MICA and MICB which would stimulate NK cell activity. NK cells could secrete cytokines such as IFN. They could also release perforin, which would make holes in the target cell membrane, and granzyme, which would degrade cell proteins and initiate apoptotic cell death of the target cell.

11) You receive a grant from the World Health Organization to generate a vaccine against the Zika virus (ZIKV). This virus is thought to cause birth defects in children exposed to ZIKV in utero.

a) An effective vaccine will need to generate potent a humoral response consisting of antibodies able to both neutralize ZIKV, and also be able to transported across the placenta. What antibody isotype meets these requirements? ***(1 point).***

IgG

b) You make a vaccine that consists of inactivated ZIKV and alum as a preservative. You give your first group of test subjects two injections separated by 30 days and then measure circulating antibodies in the blood thirty days later. The blood from every patient shows high levels of ZIKV-specific IgG, IgE and IgA. Based on this information, did your vaccine induce a Th1 or Th2 response? ***(1 point).***

The vaccine induced a Type 2 or Th2 response.

c) What cytokines would the responding subset of T cells make to support the generation of IgE and IgA? ***(2 points).***

This would be associated with Th2 cell production of IL-4 and IL-5.

d) At what physical location would you expect to significant levels of IgA to be present? ***(2 points).***

Epithelial barrier tissues / Mucosal barrier tissues

12) Transplant recipients require life-long immunosuppression, which blocks the function of $CD8^+$ T cells. Considering the typical functions of $CD8^+$ T cells in the body, explain why transplant recipients are at higher risk for developing cancer than the general population. ***(8 points).***

CD8+ T cells or CTLs are crucial to immune surveillance where they recognize and remove self cells that are expressing tumor antigens in their MHCI. They are also a potent source of IFN-gamma that increases MHCI on the tumor cells which often have a low level of MHCI expression. This way they can further their recognition by other CTLs. CTL can repeatedly use perforin and granzyme to kill the numerous cells making up a tumor. Given the non-specific nature of immunosuppression, these functions of CD8+ T cells will be blocked along cancer to develop. Furthermore, some viruses express oncogenes that lead to cancer, thus the failure to remove virally infected cells effectively also increases the risk of cancer development.