American Association of Immunologists
Teacher Summer Research Project and Curriculum Development
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Research Abstract

Exploring the link between regulatory T cells and tumor growth: a lesson in the immune system and cancer.

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Immune response suppression facilitated by regulatory T (T_{reg}) cells mediates immune homeostasis. While T_{reg} cells can suppress the body's immune responses, the role T_{reg} cells play in cancer is unclear. In this study, the murine 4T1 cancer cell line, used to mimic stage four breast cancer, was injected into FoxP3 GFP^{+} mice, and allowed to progress from one to four weeks. T_{reg} cell distribution and frequency in lymphoid and tumor tissue were analyzed via flow cytometry and immunohistochemistry protocols. Preliminary results show that the numbers of T_{reg} cells were markedly higher in draining lymph nodes compared to non-draining lymph nodes. In addition, T_{reg} cell numbers increased over time in tumor-draining lymph nodes, while numbers in the non-draining lymph nodes did not change. These results suggest a correlation between T_{reg} cell numbers and tumor progression in this model system. The knowledge, skills and techniques acquired during this research will be applied to the development and implementation of high school level immunology curriculum. Students will learn about the immune system by analyzing peer-reviewed journal articles about the immune system, cancer, and the role of T_{reg} cells. Applying what they've learned, students will design and present role play activities demonstrating the differences between humoral and cell-mediated immune responses, such as the vital role of T_{reg} cells.

Exploring the link between regulatory T cells and tumor growth: A lesson in the immune system and cancer.

I. Unit Overview: Students will…
   a. Review prior knowledge (see section VIII)
   b. Identify new learning objectives (see section IV)
   c. Conduct literary research into the role of Regulatory T cells and their potential link to cancer and autoimmune disease.
   d. Practice common laboratory techniques: simulation of counting red blood cells in a hemocytometer
   e. Develop and present an immune system role play activity incorporating new knowledge of innate and acquired immunity, including the role of various B & T cells (focus on Tregs)

II. Wisconsin's Model Academic Standards for Science Addressed:
   a. B.12.4 Show how basic research and applied research contribute to new discoveries, inventions, and applications
   b. C.12.6 Present the results of investigations to groups concerned with the issues, explaining the meaning and implications of the results, and answering questions in terms the audience can understand
   c. C.12.7 Evaluate articles and reports in the popular press, in scientific journals, on television, and on the Internet, using criteria related to accuracy, degree of error, sampling, treatment of data, and other standards of experimental design
   d. F.12.2 Understand how cells differentiate and how cells are regulated

III. Science Background

This unit is designed for high school students who have already completed an introductory level biology course. Students should have previously demonstrated mastery of the following concepts: homeostasis, basic cell structure and function including cell cycle, basic knowledge of pathogens, disease transmission and function of the immune system, the role of DNA in genetics and heredity, the role of RNA in protein synthesis, experimental design and use of basic lab equipment, such as a compound light microscope.

IV. Student Outcomes & Learning Objectives

Students will be able to…
   a. Differentiate between and describe innate and acquired immunity
   b. Identify and differentiate between functions of various lymphocytes (subpopulations of B cells and T cells)
   c. Describe and differentiate between the processes of both humoral and cell-mediated immune responses

   d. Distinguish between when and how the immune system identifies cells as self and non-self in terms of autoimmune disorders and cancer
   e. Apply the concept of homeostasis to immune function and describe how lack of immune regulation can be detrimental or deadly

f. Research and explain the specific role of regulatory T cells in immune homeostasis and predict what happens when regulatory T cells react appropriately versus inappropriately.
g. Define cancer, explain several potential causes of cancer, and draw conclusions about how both internal and external factors can lead to tumor growth and metastasis.

V. Time Requirements
a. 9 class periods (50 minutes each)
    i. Review, notes & lecture discussions: 2 class periods
    ii. Research Article and Questions: 1 class period, and time outside of class.
    iii. Counting Simulated Red Blood Cells: 1 class period
    iv. Role play discussion and preparation: 2 class periods
    v. Role play presentations: 1 class period
    vi. Summary and review, lab and role play analysis: 1 day
    vii. Summative assessment: 1 class period

VI. Advance Preparation
a. Prepare copies of the Gallimore article and scaffolding questions, 1 for each student.
b. Prepare materials for Counting Blood Cells lab:
    i. mix yeast, safranin O and DI water according to teacher notes included in lab kit.
c. Prepare role play figures/pictures and rubric: 1 figure for each student, groups of 7-8 students.
d. Prepare powerpoint or other lecture/discussion notes on the Immune System. (Ch. 43 in Campbell Biology 7th Edition)

VII. Materials and Equipment
a. Campbell Biology 7th edition text, or comparable
b. Projection system for lecture notes (Smartboard or the like)
c. Review articles:
    i. Gallimore, Awen and Andrew Godkin. "Regulatory T cells and tumour immunity – observations in mice and men." 2007. Blackwell Publishing. *Immunology*, 123, 157-163.
    http://onlinelibrary.wiley.com/doi/10.1111/j.1365-2567.2007.02748.x/abstract;jsessionid=B23FA5125ED1B629A217AD8E9555F4D9.d04t01
d. Scaffolding questions to go with Article.
e. Lab Kit: "Counting Simulated Red Blood Cells Using a Hemocytometer." 2011. Flinn Scientific. Includes Safranin O (1g), Yeast (active), disposable Hemocytometers (10), and needle tip disposable pipets (12)
Flinn Scientific, Catalog No. FB1863 - http://www.flinnsci.com/store/Scripts/prodView.asp?idproduct=21834
f. Lab equipment: Compound microscope, 2L or larger flask or beaker, Balance, 2L DI water.
g. Role Play "action figures" – pictures of various types of cells and components in the immune system. Ex: B cells, Cytotoxic T cells, Helper T cells, T reg cells, Antibodies, Antigens.
h. Guidelines and rubric for role play activity. 1 for each student.

VIII. Student Prior Knowledge
a. What is a pathogen?
b. What system in our bodies protects us from pathogens?
c. How are infectious diseases transmitted?
d. How do antibiotics fight infection?
e. What are our bodily defenses against pathogens?

f. What is the function of the immune system?
g. What is cancer?

IX. Student Expectations and Anticipated Results
a. (Answer keys/Discussion points)

X. For Classroom Discussion:
a. Chapter 43 in Campbell Biology, 7th edition
b. Powerpoint lecture notes
c. Text book guided reading questions

XI. Assessments
a. Scaffolding questions for Gallimore review article
b. Lab questions for Hemocytometry: Counting Blood Cells lab
c. Unit summative test (separate attachment)

Exploring the link between regulatory T cells and tumor growth: A lesson in the immune system and cancer.

Unit Background and Rationale:

Like any other system in our bodies, the immune system requires strict regulation in order to maintain homeostasis and prevent inappropriate and even harmful immune responses. Suppressing the immune response is a critical component in maintaining proper health. However, lack of immune suppression has been implicated in the onset and progression of various disorders, like autoimmune disease and cancer. Immune suppression is facilitated, in part, by regulatory T (Treg) cells.

This unit is designed to clarify the roles of various populations of B and T cells in acquired immune responses and to explore emerging research on the link between regulatory T cell malfunction and cancer. You will be reading a peer-reviewed article that shares the background research that links regulatory T cells and tumor immunity. In addition, you will be conducting a common laboratory technique used in immunology, called Hemocytometry. Hemocytometry is a technique used to count and estimate the number of blood cells in suspension. Finally, using what you've learned, you will be designing and presenting a group role-play, demonstrating your understanding of the mechanisms of humoral and cell mediated immunity, including the vital role of Treg cells and their link to cancer.

Materials:

1. Textbook: Campbell and Reece Biology, 7th Edition. Ch. 43 The Immune System
2. Article: Galimore and Godkin "Regulatory T cells and tumour immunity – observations in mice and men" 2007 (http://onlinelibrary.wiley.com/doi/10.1111/j.1365-2567.2007.02748.x/abstract;jsessionid=B23FA5125ED1B629A217AD8E9555F4D9.d04t01)
3. Article scaffolding questions (attached)
4. Hemocytometry lab kit: Flinn Scientific "Counting Simulated Red Blood Cells Using a Hemocytometer" (*Catalog No. FB186* - http://www.flinnsci.com/store/Scripts/prodView.asp?idproduct=21834)
5. Hemocytometry pre and post lab questions (attached)
6. Compound microscope
7. Explanation and rubric for immune response group role play activity (attached)
8. Various materials (pictures, paper, art supplied, etc) for creation of your visual aids and figures for your group role play (see attached sample)

Procedure:

Part A: Research

Pre-research & class discussion:

Before going on, use your textbook and notes from class discussion to clarify the function and importance of the following terms and concepts:

- Innate vs. Acquired immunity
- Antigens
- Lymphocytes (B and T)
- Macrophage
- Inflammatory response
- Antibodies/Immunoglobulins

- Memory cells
- Clonal selection
- Humoral vs. cell mediated responses
- Helper T Cells
- Cancer

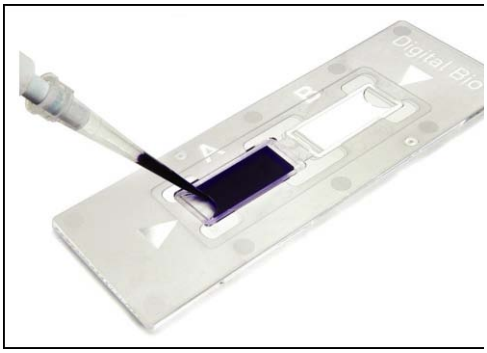
What are regulatory T cells?

Obtain the peer reviewed article "Regulatory T cells and tumour immunity – observations in mice and men" by Galimore and Godkin (2007). Read it and use the scaffolding questions on pg. 9 for deeper understanding.

Part B: Hemocytometry

Hemocytometry is a common laboratory technique used in immunology for counting blood cells under a compound microscope. You will be using a simulated suspension of red blood cells (Safranin dye and active yeast) in lieu of real blood.

1. Place a 10µl sample into the opening on your hemocytometer.
2. Place the hemocytometer under the microscope and focus it up to 40x.
3. Count the number of RBCs in each designated shaded square and determine the number of RBCs per mm^3.
4. Complete the data table and answer the analysis questions



Disposable Hemocytometer
montreal-biotech.com

See Flinn Scientific's "Bio Fax!" kit and instructions for the complete lab procedure (2007. Catalog No. FB1863, Publication No. 10819).

Part C: Group Immunology Role Play

You will be organized into groups of 6-8 students. After class discussions and research on the immune system, you will be designing and presenting a role-play that depicts the mechanisms of the acquired/specific immune response. Your role-play should:

- ✓ Show clear contrast between humoral and cell-mediated immunity
- ✓ Make clear the roles of at least 5 lymphocytes, including B and T cells. One of them must be Treg cells
- ✓ Show how the body might respond to at least two types of antigens, including a tumor.
- ✓ Highlight the difference between an appropriate response from Treg cells and a potentially inappropriate response that may lead to diseases like cancer or autoimmunity.
- ✓ Be creative!
- ✓ Include all group members equally
- ✓ Be clear and easy to understand
- ✓ Be at least 7-10 minutes in length
- ✓ Be accompanied by visual aids (at least one for each character)

Data Collection
Part B: Hemocytometry

Create a data table that displays the number of cells counted for both Sample A and B in each of the 5 squares. Also, include the total number of cells counted for each sample.

Discussion/Analysis

Part A: Galimore Research Article Questions

1. There are many different types of lymphocytes, including T cells. This article focuses on the role of a specific population called Treg cells. Before reading, review the different types of T cells and their different roles.
2. For better comprehension, get in the habit of looking up unfamiliar words. Look up and define the following terms: fibrosarcoma, concomitant, ablating CD4, CD8, CD25, *in vitro* and *in vivo*. Continue with additional unknown words throughout the article as needed.
3. According to Robert North's studies in the 1980s, why were T cells unable to stop tumor growth?
4. Summarize the correlation between rejection of tumor cells and regulatory T cell (Treg) activity.
5. What did the drug cyclophosphamide result in? How/why?
6. What is CD4? Why were CD4-depleting antibodies used? What was the result?
7. What is the most reliable marker used to identify Treg cells?
8. Look up on the internet: what population of T cells are CD8+ and help with tumor rejection?
9. In Shimizu's study, what happened to mice inoculated with tumor cells after the all the CD25+ cells (Tregs) were depleted?
10. What is meant by the "immune surveillance" concept? How did it become better supported in recent years?
11. Why do you think human studies are more restricted, making drawing a correlation between Treg function in mice and Treg function in humans difficult?
12. Even though immunosuppressive drugs are used widely for many diseases in humans, still the greatest increase in tumors is due to what other variable?
13. Studies indicate there's a positive correlation between better cancer patient outcomes and increased numbers of lymphocytes in their tumors (tumor-infiltrating lymphocytes, or TILs). What might this suggest about how your body responds to cancer?
14. Describe the concept of "Duke's staging" in reference to CRC?
15. What is the difficult question regarding lymphocyte reaction and tumor progression?
16. Summarize the conclusion of the Galon *et al*. study.
17. What are 4 potential reasons for why tumor antigens seem to be ineffective at triggering an immune response?
18. In paragraph 2, on page 160, what is meant by "CD4+ lymphocytes appeared to downregulate antitumor CD8+ T cell responses"? Look up "downregulation", then review what the difference is between CD4+ and CD8+ cells and paraphrase that statement.
19. What is the overall trend in Treg involvement of regulating response to tumors? What is the implication of this observation?
20. Determine what the term "ascites" means in reference to ovarian cancer.
21. In the 2004 Curiel study of 104 biopsied tumors, what conclusion did they draw between numbers of Tregs and stage of the disease and patient survival?
22. What is Denileukin diftitox or "Ontak"? What is it being tested to do?
23. What is one inadvertent side effect of targeting CD25+ cells in order to deplete Tregs?
24. Autoimmunity is mentioned in paragraph 4 of pg. 161. What is autoimmunity, and how are Tregs related to this?
25. Why do you think the author cites numerous different scientists and their studies of T cell involvement in different kinds of cancers, even though they all derived similar conclusions?

Part B: Hemocytometry Analysis

1. Calculate the number of red blood cells per mm^3 for each sample using the following equation: (sum of the 5 squares) x 5 x 10^4 = Number of cells in 1 mm^3
2. Is there a difference in the cell counts for each sample? Research the normal range for RBC's and propose a reason why the cell counts might not fit the normal range.
3. Compare the size and shape of a RBC to that of a white blood cell and a tumor cell, like the 4T1 breast cancer cell line. Draw a picture depicting how these three cells might look in comparison to each other if they were all observed in the same sample.
4. Assume you used a method called Flow Cytometry that sorts cells according to size and complexity, in order to count specific cell types in suspension. Your "Flow" data indicates the number of CD4+ Fox P3+ T cells (Tregs) in suspension is approximately 282,500 for Sample A, and 423,000 in Sample B. Calculate the percentage of Treg cells in each sample, and compare. Research and determine if they are within normal rage. If not, suggest a possible cause for the data.
5. Using Hemocytometry as part of your methodology, design a simple controlled experiment to test the following hypothesis: Mice with advanced stage breast cancer show significantly higher numbers of Treg cells in both the tumor and tumor-draining lymph nodes, compared to healthy control mice.

Acknowledgments

I would like to thank Professor Douglas Steeber, Ph.D, and doctoral student Jessica Loppnow from the University of Wisconsin at Milwaukee for greatly contributing to my research and development of knowledge on the Immune System in order to build this curriculum. Dr. Steeber generously extended this opportunity to me and Jessica Loppnow worked many hours one on one with me to perform the lab techniques, acquire data and generate ideas for my curriculum plan.

References

1. Campbell, Neil A. and Jane B. Reece. Biology. AP Edition, 7th Edition. Pearson Education, Inc. 2005.
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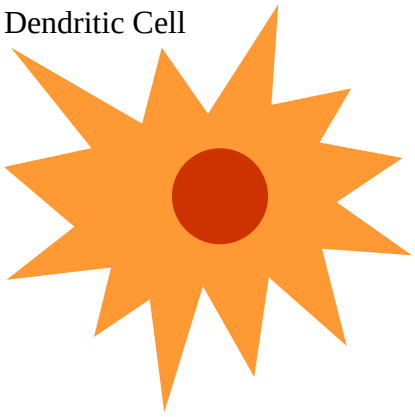
4. Kindt, Thomas J., Barbara Anne. Osborne, and Richard A. Goldsby. Kuby Immunology 6th Edition. W.H. Freeman and Company 2007.
5. Mailloux, AW,.Young, MRI. "Regulatory T-cell Trafficking: From Thymic Development to Tumor-Induced Immune Suppression." Crit. Rev. Immunol. 30:435, 2010.
6. Watanabe, MA, Oda, JM, Amarante, MK, Voltarelli, JC. Regulatory T cells and breast cancer: implications for immunopathogenesis. Cancer Metastasis Rev. 29:569, 2010.

Appendix A
Sample rubric for role play

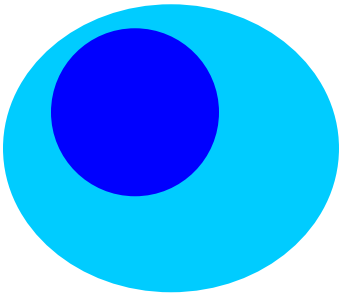
	Excellent	Average	Below Average
Humoral vs. cell-mediated	Complete, accurate and detailed 5	Complete and accurate 3-4	Inaccurate, incomplete 0-2
Roles of WBCs, B, T, Treg cells	All cells defined and examples given 5	All cells defined 3-4	Incomplete or inaccurate 0-2
Imm Resp to antigens & tumor	Complete, accurate and detailed 5	Complete and accurate 3-4	Inaccurate, incomplete 0-2
Appropriate vs. inappropriate Response of Treg	Complete, accurate and detailed 5	Complete and accurate 3-4	Inaccurate, incomplete 0-2
Creativity	Novel, unique, Engages most students 5	Relevant, engages most students 3-4	Not relevant or relatable to topic 0-2
All members included equally	95-100% involvement 5	80-94% involvement 3-4	0-79% involvement 0-2
Clarity & ease of understanding	Very clear and understandable 5	Clear and understandable 3-4	Unclear, difficult to understand 0-2
Time length	7-10 minutes 5	5-6 minutes 3-4	0-4 minutes 0-2
Visual Aids	6+, large, easy to read 5	At least 5, large, clear 3-4	4 >, unclear, small 0-2

Sample figures for Role Play activity

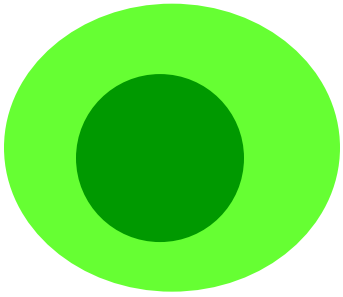
Dendritic Cell



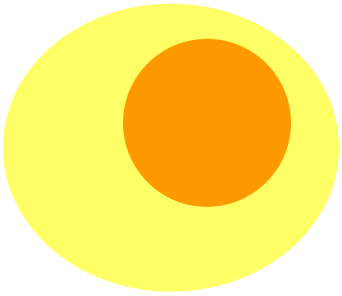
Cytotoxic T Cell



Helper T Cell



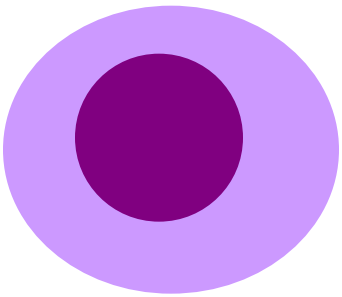
B Cell



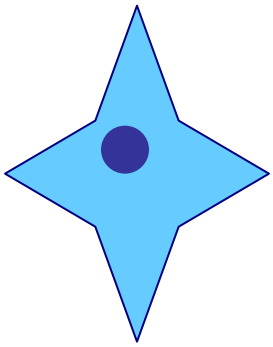
Antigen

Antibody

T $_{Reg}$ Cell



Cancer Cell



Appendix B
Textbook Alignment

Chapter 43: The Immune System
(Campbell Biology 7th Edition)

Background vocab – Define the following:

Microorganism

Pathogen

Antibiotic

Antibody

Antigen

Define Innate Immunity:

What makes up the innate immunity in your body?

Define Acquired Immunity:

Give an example of acquired immunity:

43.1 Innate Immunity provides broad defenses against infection

Describe External Defenses of your body:

What does Internal Defenses of your body mainly depend on? _____

Which cells typically carry out phagocytosis of invading microorganisms?

How do phagocytes find their prey?

What structures on these cells are recognized by phagocytes?

How are microbes destroyed once they are phagocytized?

It is the job of phagocytes to identify and engulf foreign microoranisms. Why do we worry about the pathogenic threat of microbes?

Complete the following table regarding Phagocytic Cells:

Type of White Blood Cell	Abundance	Description/ mechanism of action	Location
Neutrophils			
Macrophages			
Eosinphils			
Dendritic Cells			

What is the Lymphatic System?

What is the role of the lymphatic system in immune response?

How do Interferon proteins limit the spread of viruses?

What is the Inflammatory Response?

What is the result of the release of Histamine from mast cells?

How does the inflammatory response aid in creating innate defense?

What is the role of chemokines in this response?

What kinds of cells do Natural Killer (NK) cells target?

How do NK cells lead to cell death?

What is apoptosis?

43.2 In acquired immunity, lymphocytes provide specific defenses against infection

	B Cells	T Cell
Cell Receptor		
Antigens recognized		

Where do lymphocytes originate from?

What is the role of the Thymus?

What is meant by “Clonal Selection” of Lymphocytes?

Compare/Contrast effector cells and memory cells.

Outline the events and differences between a primary immune response and a secondary immune response

Compare/Contrast Cell Mediated vs. Humoral immune responses

Discuss the roles of Helper T cells, Cytotoxic T cells, and B cells. Include CD4 and CD8 in your discussion.

Antibody Classes

	Roles	Special Features/Misc.
IgM		
IgG		
IgA		
IgE		
IgD		

Disposal of Antigens: what is a “MAC”

Compare and contrast Active vs. Passive immunity. Include vaccinations in your discussion.

43.4 The immune system's ability to distinguish self from nonself limits tissue transplantation

Blood Transfusions: Transfusion Reactions:

Blood type	Antibodies	A	B	AB	O
A					
B					
AB					
O					

Who is the universal donor? _____ universal recipient? _____

What is Rh factor?

43.5 Exaggerated, self-directed or diminished immune responses can cause disease

Explain why many people have "allergies." What does that really mean? What is your body doing? Include Anaphylactic Shock in your discussion.

What is an autoimmune disease? List several examples.

What are some types of inborn and acquired immunodeficiency diseases?

Discuss the role of stress.

Explain the effects of HIV on the body. Include discussion on reverse transcription.

Appendix C

Formative Assessment

Name: _____ Class: _____ Date: _____

Cells and Systems Test: Endocrine, Nervous, Immune

Multiple Choice

Identify the letter of the choice that best completes the statement or answers the question.

____ 1. Based on their effects, which pair below could be considered antagonistic?
   a. insulin and glucagon
   b. growth hormone and prolactin
   c. endocrine and exocrine glands
   d. hormones and target cells

____ 2. Why is it that some body cells respond differently to the same peptide hormones?
   a. Different target cells have different genes
   b. Each cell knows how it fits into the body's master plan.
   c. the response to the peptide is determined by the type of receptor molecule that the hormone binds to and the resulting action in the cell
   d. The circulatory system regulates responses to hormones by routing the hormones to specific targets.

____ 3. How does a steroid hormone initiate a response in target cells?
   a. by binding to cell membrane receptors
   b. through release outside the body
   c. by causing a negative feedback mechanism
   d. by binding to receptor proteins present inside of the cell

____ 4. The endocrine system and the nervous system are structurally related. Which of the following cells best illustrates this relationship?
   a. a neuron in the spinal cord
   b. a steroid-producing cell in the adrenal cortex
   c. a neurosecretory cell in the hypothalamus
   d. a brain cell in the cerebral cortex

____ 5. If a person drinks a large amount of water in a short period of time, he or she may die from water toxicity. ADH can help prevent water retention through interaction with target cells in the
   a. anterior pituitary.
   b. posterior pituitary.
   c. adrenal gland.
   d. kidney.

____ 6. Iodine is necessary in the production of Thyroxine. Which gland requires iodine to function properly?
   a. ovaries and testes
   b. adrenal
   c. thyroid
   d. pancreas

The question below refers to the following information.

In an experiment, rats' ovaries were removed immediately after impregnation and then the rats were divided into two groups. Treatments and results are summarized in the table below.

	Group 1	Group 2
Daily injections of progeterone (milligrams)	0.25	2.0
Percentage of rats that carried fetuses to birth	0	100

____ 7. The results most likely occurred because progesterone exerts an effect on the
   a. general health of the rat.
   b. size of the fetus.
   c. uterine lining growth
   d. number of eggs fertilized.

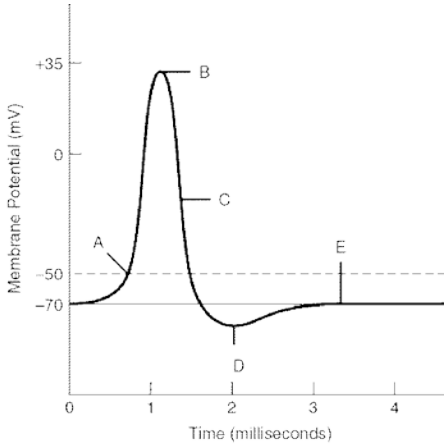
____ 8. Which of the following is secreted by the pancreas?
   a. prolactin
   b. glucagon
   c. thyroxine
   d. oxytocin

____ 9. Blood samples taken from an individual who had been fasting for 24 hours would have which of the following?
   a. high levels of insulin
   b. high levels of glucagon
   c. high levels of both insulin and glucagon
   d. low levels of glucagon

____ 10. Which combination of gland and hormone would be linked to winter hibernation and spring reproduction in bears?
   a. pineal gland, melatonin
   b. hypothalamus gland, melatonin
   c. anterior pituitary gland, gonadotropin-releasing hormone
   d. pineal gland, estrogen

____ 11. The sodium-potassium pump of neurons pumps
   a. Na^+ and K^+ into the cell.
   b. Na^+ and K^+ out of the cell.
   c. Na^+ into the cell and K^+ out of the cell.
   d. Na^+ out of the cell and K^+ into the cell.

For the following 2 questions, refer to the graph of an action potential in the figure below and use the letters to indicate your answer.



____ 12. The sodium gates open.
   a. A
   b. B
   c. C
   d. D
   e. E

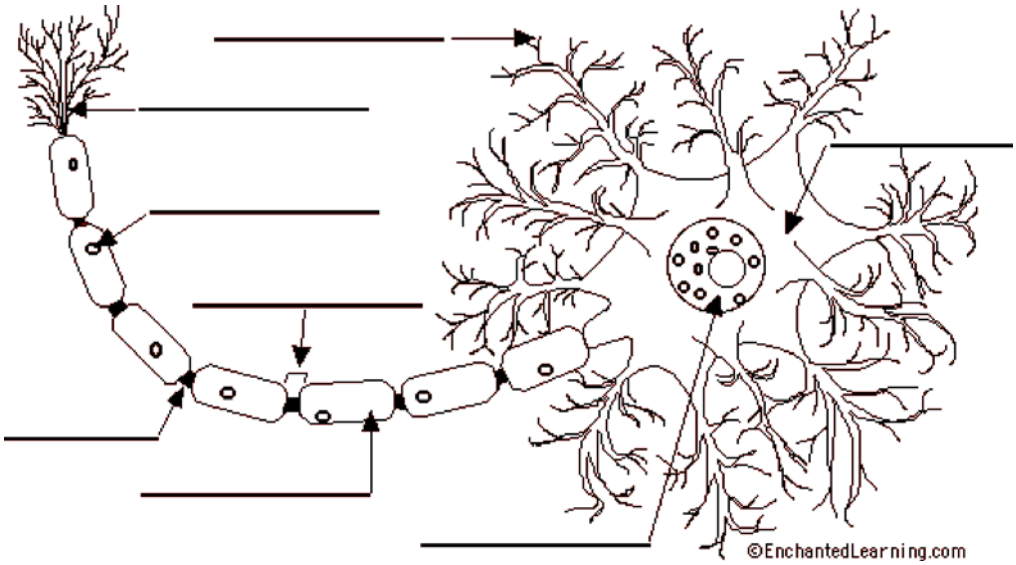
____ 13. The membrane is at resting potential.
   a. A
   b. B
   c. C
   d. D
   e. E

____ 14. Neurotransmitters are released from presynaptic axon terminals into the synaptic cleft by which mechanism?
   a. exocytosis
   b. active transport
   c. diffusion
   d. endocytosis

____ 15. A neurotransmitter can trigger different responses in postsynaptic cells due to which of the following?
   a. sodium present
   b. types of receptors present
   c. level of resting potential
   d. exocytosis

____ 16. What is the main neurotransmitter of the sympathetic nervous system?
   a. acetylcholine
   b. cholinesterase
   c. thyroxine
   d. epinephrine

____ 17. The divisions of the nervous system that have antagonistic actions, or opposing actions are
   a. motor and sensory.
   b. sympathetic and parasympathetic.
   c. presynaptic and postsynaptic.
   d. forebrain and hindbrain.



____ 18. Neurons transmit electrical impulses. This would happen very slowly without the insulating effects of
   a. the axon
   b. dedrites
   c. nodes of ranvier
   d. myelin sheath

____ 19. The innate immunity that protects a person digging in the garden from developing a microbial infection includes all of the following except
   a. lymphocytes.
   b. the skin.
   c. mucous membranes.
   d. antimicrobial proteins.

____ 20. In the inflammatory response, the absence of which of the following would prevent all the others from happening?
   a. release of histamine
   b. increased permeability of blood vessels
   c. increased population of phagocytes in the area
   d. leakage of plasma to the affected area

____ 21. Which action below is affected by an antihistamine?
   a. blood vessel dilation
   b. phatocytosis of antigens
   c. MHC presentation by macrophages
   d. the secondary immune response

____ 22. Inflammatory responses may include all of the following except
   a. clotting proteins sealing off a localized area.
   b. increased activity of phagocytes in an inflamed area.
   c. reduced permeability of blood vessels to conserve plasma.
   d. release of substances to increase the blood supply to an inflamed area.

For the questions below, match the following answers with the phrase that best describes them.

A. cytotoxic T cells
B. B cells
C. helper T cells
D. macrophages

____ 23. These cells are involved in cell-mediated immunity and destroy virally infected cells.
a. A
b. B
c. C
d. D

____ 24. These cells are involved in humoral immunity, and release antibodies to target antigens in bodily fluids.
a. A
b. B
c. C
d. D

____ 25. Phagocytosis of microbes by macrophages is enhanced by
a. the binding of antibodies to the surface of microbes.
b. antibody-mediated agglutination of microbes.
c. the release of bacterial toxins.
d. A and B only

____ 26. What happens to people who receive flu vaccinations?
a. They develop active immunity to the flu strains in the vaccine.
b. They develop passive immunity to the flu strains in the vaccine..
c. At least 50% of people do not develop any immunity to the flu strains in the vaccine
d. Many people get sick from the flu vaccine itself.

____ 27. A patient reports severe symptoms of watery, itchy eyes and sneezing after being given a flower bouquet as a birthday gift. A reasonable initial treatment would involve the use of
a. a vaccine.
b. monoclonal antibodies.
c. sterile pollen.
d. antihistamines.

____ 28. Which choice below describes how HIV affects the body?
a. it attacks muscle cells which make the patient weak
b. it invades helper T cells preventing acquired immunity
c. it invades B cells preventing innate immunity
d. it attacks AIDS cells preventing acquired immunity

Cells and Systems Test: Endocrine, Nervous, Immune
Answer Section

MULTIPLE CHOICE

1. ANS: A TOP: Concept 45.2
2. ANS: C TOP: Concept 45.2
3. ANS: D TOP: Concept 45.2
4. ANS: C TOP: Concept 45.3
5. ANS: D TOP: Concept 45.3
6. ANS: C TOP: Concept 45.4
7. ANS: C TOP: Concept 45.4
8. ANS: B TOP: Concept 45.4
9. ANS: B TOP: Concept 45.4
10. ANS: A TOP: Concept 45.4
11. ANS: D TOP: Concept 48.2
12. ANS: A TOP: Concept 48.3
13. ANS: E TOP: Concept 48.3
14. ANS: A TOP: Concept 48.4
15. ANS: B TOP: Concept 48.4
16. ANS: D TOP: Concept 48.4
17. ANS: B TOP: Concept 48.5
18. ANS: D
19. ANS: A TOP: Concept 43.1
20. ANS: A TOP: Concept 43.1

21. ANS: A TOP: Concept 43.1
22. ANS: C TOP: Concept 43.1
23. ANS: A TOP: Concept 43.3
24. ANS: B TOP: Concept 43.3
25. ANS: D TOP: Concept 43.3
26. ANS: B TOP: Concept 43.3
27. ANS: D TOP: Concept 43.5
28. ANS: B TOP: Concept 43.5

Sample Additional Immunity questions for Unit Test

29. Which of the following substances or molecules could be considered antigens in your body?
    a. Viruses and bacteria
    b. Donor or transplant organs, tissues or blood
    c. Pollen, ragweed or cat dander
    d. All of the above
30. Which of the following types of white blood cells are typically involved in targeting malignant tumor cells for disposal?
    a. B Cells
    b. Cytotoxic T Cells
    c. Natural Killer Cells
    d. B and C are both correct
    e. A-C are all correct
31. What is the difference between B cells and T cells?
    a. B cells target pathogenic antigens like virus and bacteria while T cells target allergens, like pollen and dust.
    b. B cells are involved in innate immunity while T cells are involved in acquired immunity.
    c. B cells are involved in the humoral response while T cells are involved in the cell-mediated response.
    d. B cells originate in the bloodstream while T cells originate in the thyroid.
32. Autoimmunity results when
    a. The immune system doesn't differentiate self from non-self tissues.
    b. The immune system inappropriately reacts to non-pathogenic allergens, like pollen and cat dander.
    c. The immune system over reacts to a vaccine.
    d. A virus, like HIV targets T cells.
33. How is the body's inflammatory response different from a cell-mediated response?
    a. Inflammatory responses are specific, while cell-mediated responses are non-specific
    b. Inflammatory responses are non-specific, while cell-mediated responses are specific.
    c. Inflammatory responses utilize T cells, while cell-mediated responses use B cells.
    d. Inflammatory responses target infected cells, while cell-mediated responses target extra-cellular pathogens.
34. Which of the following best describes the role of a Regulatory T (Treg) Cell?
    a. Maintains immune homeostasis
    b. Suppresses the immune system when necessary
    c. Maintain self-tolerance
    d. All of the above

35. Which of the following best describes current research findings on the possible link between Treg cells and tumors?
    a. Numbers of Tregs have been positively correlated with tumor progression in mice
    b. Treg malfunction causes cancer
    c. Lower numbers of Tregs are correlated with more advanced stages of cancer
    d. Treg activity in mice with cancer is easily correlated with Treg activity in humans with cancer

36. What is Hemocytometry used for?
    a. Detection of B and T cell populations
    b. Counting blood cells in suspension
    c. Detecting the Foxp3 marker in Tregs
    d. Comparing cell size and shape

37. Which of the following antigen(s) is/are considered known causes of certain types of cancer?
    a. *Candida albicans* yeast
    b. Human Papillomavirus and Epstein barr virus
    c. *E.coli* and *Salmonella* bacteria
    d. *Trypanosoma* (Malaria) protist