

# Exploring Phagocytosis and Bacterial Pathogens with a Multimedia Learning Object

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## Abstract

This activity consists of a worksheet written to accompany a Flash multimedia learning object in the ASM Visual Resources library. As students explore the multimedia learning object, they collect information about phagocytosis in macrophages and about how plague and tuberculosis bacteria subvert phagocytosis so they can use macrophages as a host cell for pathogenesis. By the time they have explored fully, students will have all the information needed to fill out the worksheet.

## Activity

**Invitation for User Feedback.** If you have used the activity and would like to provide feedback, please send an e-mail to [MicrobeLibrary@asmusa.org](mailto:MicrobeLibrary@asmusa.org). Feedback can include ideas which complement the activity and new approaches for implementing the activity. Your comments will be added to the activity under a separate section labeled "Feedback." Comments may be edited.

### INTRODUCTION

#### Prep Time Required.

10 to 30 minutes to preview the multimedia learning object, modify the worksheet (if desired), and print out the worksheet.

#### Class Time Required.

5 minutes (instructor demonstrates animation); students will spend an additional 20 to 30 minutes outside of class to explore the multimedia learning object and fill in the worksheet.

#### Learning Objectives.

At the completion of this activity, students will be able to explain how phagocytosis works and how certain pathogens can block this process and use phagocytes as a host cell.

#### Background.

Some exposure to the concepts of nonspecific immune defenses and pathogenic virulence factors will be helpful.

### PROCEDURE

#### Materials.

Computer linked to the internet with Shockwave-Flash plug-in installed (see below).

#### Student Version.

[Student Worksheet](#)  
[Phagocytosis and Bacterial Pathogens Animation](#)

#### Instructor Version.

- Students find animations engaging and fun to watch. However, there is no guarantee that they will pay sufficient attention to learn as much as instructors hope they will learn. This activity has been designed as something of a "visual scavenger hunt," in which students must hunt for answers to questions as they examine a complex multimedia learning object. In this learning object, unlike a simple animation, students control their journey by interacting with various control buttons. In the process, they should learn substantially more about aspects of phagocytosis and the ways certain bacterial pathogens block this process and grow at the expense of phagocytes.
- The organisms chosen for this learning object include one extracellular (*Yersinia pestis*) and one intracellular (*Mycobacterium tuberculosis*) pathogen. This activity will help students appreciate similarities and differences in the virulence mechanisms between these two types of organisms.
- Some of the content of the learning object is very basic and would be appropriate for students at all levels. Some content (e.g., the mechanism of action of different Yop virulence factors) is more detailed and would be more appropriate for microbiology majors. Instructors may wish to modify or shorten the list of questions based on student level. The student worksheet can easily be pasted into word processing software and modified to individual tastes.
- Unless you know that the computers your students will use already contain an up-to-date version of the Flash plug-in, it will help to briefly review the use of plug-ins. These are free software programs that allow Web browsers such as Internet Explorer and Netscape Communicator to display certain file types not supported by browser

technology. They must be installed in a "plug-in" folder (directory) within the browser folder (directory). Depending on the operating system and browser, installation may be automatic or may require closing the browser and running an installer, then restarting the browser.

- The multimedia learning object was authored in Macromedia Flash, and technically only requires the Flash plug-in. However, if an update is necessary, it would be advisable to upgrade the Macromedia Shockwave plug-in since this includes Flash and will serve more purposes.
- Most computers will have some version of the Shockwave plug-in already installed, as this plug-in is shipped with all versions of Internet Explorer and comes preinstalled on most new computers sold. However unless the plug-in is sufficiently recent to support Flash 5 files, the pop-up information screens of the multimedia learning object will not work, making it impossible to access some information needed to complete the worksheet. A test page to see if the correct plug-in is installed is located at <http://www.macromedia.com/shockwave/welcome/>; if a browser does not load and display the test animation, then the Shockwave plug-in should be downloaded and installed from the Macromedia download page: <http://www.macromedia.com/downloads/>. Alternatively, one only has to try to open the pop-up window on the first screen of the learning object. A link is also provided directly from the Flash object to the Shockwave download page.
- The instructor should review the Flash learning object to see how it will integrate with material presented in class. [Phagocytosis and Bacterial Pathogens Animation](#)
- If classroom facilities allow internet access to websites, the instructor may want to show at least a portion of the Flash multimedia learning object during a class presentation on host defenses or mechanisms of pathogenesis. This will add interest to the topic, as well as prepare students for what to expect when they access the multimedia learning object on their own.

**Safety Issues.** None.

## ASSESSMENT and OUTCOMES

### Suggestions for Assessment.

1. The Flash multimedia learning object has been designed so that much of the information needed to answer questions is found in pop-up boxes controlled by a "mouse over" command. If students attempt to electronically copy the information so they can paste it into the worksheet, the pop-up window will close, so that some reading and writing is necessary. Other information is present on different screens in graphic format and cannot be copied and pasted. Thus students will be forced to read the material and write their own version of the answer in the worksheet.
2. Unless the worksheet is graded, it is unlikely that students will complete it. The worksheet can be assigned as one of a number of graded assignments, depending on the grade structure for the course.
3. In my course, I reserve 50 points out of 500 for various short assignments such as this, without knowing at the beginning of the semester how many such activities I will use or how many points I will assign. The remaining 450 points are assessed by exams and labs. This structure gives me the flexibility to assign a number of short activities that are graded differently than exams and that allow students to demonstrate mastery of certain topics. An assignment like this might be worth 5 to 10 of those 500 points (1 to 2%).
4. The worksheet can also be assigned as an extra credit or make-up assignment for certain students who need an opportunity to improve their grade.

### Field Testing.

The activity was tested during a summer course with 15 students. Reaction was very positive, as indicated in the following comments:

- "Animations were very useful, helped in visualization of process."
- "I thought animations made things clearer, would have liked to see them in other science classes."
- "These visual representations of processes are more clear to me than the same process described in words."

## SUPPLEMENTARY MATERIALS

### References.

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3. **Cornelis, G. R., A. Boland, A. P. Boyd, C. Geuijen, M. Iriarte, C. Neyt, M. P. Sory, and I. Stainier.** 1998. The virulence plasmid of *Yersinia*, an antihost genome. *Microbiol. Mol. Biol. Rev.* **62**:1315.
4. **Cornelis, G. R., and H. WolfWatz.** 1997. The *Yersinia* Yop virulon: a bacterial system for subverting eukaryotic cells. *Mol. Microbiol.* **23**:861–867.
5. **Galán, J. E., and A. Collmer.** 1999. Type III secretion machines: bacterial devices for protein delivery into host cells. *Science* **284**:1322–1328.
6. **Helmuth, L. A.** 2000. Weak link in TB bacterium is found. *Science* **289**:1123–1125.
7. **Silhavy, T. J.** 1997. Death by lethal injection. *Science* **278**:1085–1086.

### Answer Key.

[Answer Key for Student Worksheet](#)

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Name: \_\_\_\_\_ Date: \_\_\_\_\_

**Student Worksheet: Phagocytosis & Bacterial Pathogens**Instructions:

- Print out a paper copy of this worksheet.
- Be sure the Web browser you will use contains a recent version of the Shockwave plug-in needed to view all features of the animation. Visit the Macromedia Web Player Test page at (<http://www.macromedia.com/shockwave/welcome/>). If no animation is visible, you will need to download and install the current version of the Shockwave plug-in, which includes the Flash plug-in necessary for this activity. Download the Shockwave plug-in at (<http://www.macromedia.com/downloads/>). Follow instructions listed on the page.
- Explore the Web-based Flash animation "Phagocytosis & Bacterial Pathogens" by Thomas Terry, located at this URL: <http://www.microbelibrary.org/images/tterry/anim/phago053.html>
- As you explore, look for answers to the questions and fill in the worksheet.
- Return the completed worksheet to your instructor as directed.

Questions:

1. Where are macrophages found in the human body?
2. What cells carry out phagocytosis?
3. What is a phagosome?
4. What is in a lysosome?
5. How is a phagolysosome formed?
6. What happens during an "oxygen burst"?
7. What are the end products of phagocytosis?
8. What happens to the products of phagocytosis?
9. What bacterium causes plague?
10. Is plague infection an intracellular or extracellular process?
11. What is the role of the Type III secretory apparatus during plague infection?
12. What are Yops?
13. Identify the mode of action of at least one Yop.
14. What is a common symptom of plague infection?
15. What bacterium causes tuberculosis (TB)?

16. Is TB infection an intracellular or extracellular process?
17. How many people worldwide are estimated to be infected with the TB pathogen?
18. How many of these people will actually develop some clinical symptoms of TB?
19. What happens to the TB bacteria in those people who don't have any clinical symptoms?
20. How many people die each year as a result of TB?
21. What prevents TB bacteria from being killed by phagocytosis?
22. What organ(s) in the human body can be infected with TB?

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**Answer Key****Student Worksheet: Phagocytosis & Bacterial Pathogens**Questions:

1. Where are macrophages found in the human body?

**A.** Mainly in lymph nodes.

2. What cells carry out phagocytosis?

**A.** Certain white blood cells, including macrophages and granulocytes.

3. What is a phagosome?

**A.** A membrane-bounded structure resulting from the invagination of a cell membrane to enclose particulate matter, such as a bacterial cell.

4. What is in a lysosome?

**A.** Dozens of degradative enzymes called acid hydrolases, including DNases, RNases, proteases, phosphatases, and lipases.

5. How is a phagolysosome formed?

**A.** By fusion of a phagosome with a lysosome.

6. What happens during an "oxygen burst"?

**A.** Stored sugar is used to make NADPH, which combines with O<sub>2</sub> to make toxic peroxide (H<sub>2</sub>O<sub>2</sub>) and superoxide.

7. What are the end products of phagocytosis?

**A.** Smaller molecules resulting from enzymatic digestion of biological molecules such as DNA, RNA, protein, and lipid.

8. What happens to the products of phagocytosis?

**A.** They are released from the cell.

9. What bacterium causes plague?

**A.** *Yersinia pestis*.

10. Is plague infection an intracellular or extracellular process?

**A.** Extracellular.

11. What is the role of the Type III secretory apparatus during plague infection?

**A.** It acts like a syringe, allowing virulence factors to be injected into the host macrophage.

12. What are Yops?

**A.** *Yersinia* outer proteins.

13. Identify the mode of action of at least one Yop.

**A.** (any one of the following)

YopE: disrupts host cell microfilaments (actin), prevents movement of phagosomes to lysosome to form phagolysosomes.

YopJ: a cysteine protease that cleaves macrophage proteins, including those that would normally send

an "SOS" signal to other immune cells. More specifically, YopJ blocks two signaling pathways: the "mitogen-activated protein kinase" (MAPK) pathway and the nuclear factor kB.

YopH: a serine phosphatase that removes phosphate from certain enzymes needed to promote attachment of cytoskeletal fibers to adhesion zones on the macrophage surface needed for phagocytosis.

YopT: function not well known.

14. What is a common symptom of plague infection?

A. Swollen lymph glands or "buboes" filled with dead macrophages

15. What bacterium causes tuberculosis (TB)?

A. *Mycobacterium tuberculosis*.

16. Is TB infection an intracellular or extracellular process?

A. Intracellular.

17. How many people worldwide are estimated to be infected with the TB pathogen?

A. About 2 billion people (1 out of 3), according to World Health Organization estimates.

18. How many of these people will actually develop some clinical symptoms of TB?

A. Less than 10% of those infected.

19. What happens to the TB bacteria in those people who don't have any clinical symptoms?

A. The bacteria grow briefly, then stop growing and enter a latent state inside host macrophages.

20. How many people die each year as a result of TB?

A. 3 million.

21. What prevents TB bacteria from being killed by phagocytosis?

A. The bacteria synthesize virulence proteins which inhibit fusion of phagosomes with lysosomes, thus preventing the formation of phagolysosomes.

22. What organ(s) in the human body can be infected with TB?

A. Lungs, of course, but almost every organ in the body, such as kidney, liver, muscle, etc., can be infected.