BioEd℠ Teacher's Guide

the science of HIV/AIDS

Making Copies of an HIV Particle

The Deadly Cycle

by

Gregory L. Vogt, Ed.D.

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RESOURCES

Free, online presentations, downloadable activities in PDF format, and annotated slide sets for classroom use are available at www.bioedonline.org or www.k8science.org.

CONTENT ADVISORY

See the following resources for additional information about HIV/AIDS and advice for discussing HIV/AIDS with students.

- National Institute of Allergy and Infectious Diseases, National Institutes of Health (NIH), offers resources on understanding HIV/AIDS: niaid.nih.gov/topics/hivaids/and aidsinfo.nih.gov.
- National Institute on Drug Abuse, NIH, offers facts about drug abuse and the link between it and HIV/AIDS: hiv.drugabuse.gov.
- The Centers for Disease Control and Prevention provides up-to-date information on HIV/AIDS prevention: cdc.gov/hiv/topics.

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BioEd℠

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CCIT The Center for Collaborative and Interactive Technologies

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Many microscopic images used in this guide, particularly images obtained from the Public Health Image Library of the CDC, are part of an online library containing other images and subject matter that may be unsuitable for children. Caution should be used when directing students to research health topics and images on the Internet. URLs from image source websites are provided in the Source URL list, to the right.

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SOURCE URLs

AMERICAN DENTAL EDUCATION ASSOCIATION
explorehealthcareers.org

BAYLOR COLLEGE OF MEDICINE
BIOED ONLINE TEACHER RESOURCES
bioedonline.org | k8science.org
BAYLOR-UT CENTER FOR AIDS RESEARCH
bcm.edu/cfar
MOLECULAR VIROLOGY AND MICROBIOLOGY
bcm.edu/molvir

DARTMOUTH COLLEGE
ELECTRON MICROSCOPE FACILITY
dartmouth.edu/~emlab/

THE HENRY J. KAISER FAMILY FOUNDATION
kff.org

JOURNAL OF NANOBIOTECHNOLOGY
jnanobiotechnology.com/content/3/1/6

NATIONAL INSTITUTES OF HEALTH
LIFEWORKS
science.education.nih.gov/lifeworks

NATIONAL CENTER FOR RESEARCH RESOURCES
ncrr.nih.gov
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www.niaid.nih.gov
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ANIMATED TUTORIALS: MICROBIOLOGY
http://sumanasinc.com/webcontent/animation.html

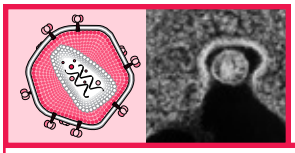
U.S. CENTERS FOR DISEASE CONTROL AND PREVENTION (CDC)
HIV/AIDS PREVENTION
cdc.gov/hiv/topics
PUBLIC HEALTH IMAGE LIBRARY
phil.cdc.gov

U.S. CENTRAL INTELLIGENCE AGENCY
THE WORLD FACTBOOK
https://www.cia.gov/library/publications/the-world-factbook/geos/us.html

WELLCOME IMAGES
images.wellcome.ac.uk

WHAT IS PUBLIC HEALTH
whatispublichealth.org

WIKIMEDIA COMMONS
commons.wikimedia.org



HIV virus particle. Charles P. Daghlian, Ph.D., Louisa Howard, Dartmouth College.

INTRODUCTION

Microbial Challenges

Infectious diseases have plagued humans throughout history. Sometimes, they even have shaped history. Ancient plagues, the Black Death of the Middle Ages, and the "Spanish flu" pandemic of 1918 are but a few examples.

Epidemics and pandemics always have had major social and economic impacts on affected populations, but in our current interconnected world, the outcomes can be truly global. Consider the SARS outbreak of early 2003. This epidemic demonstrated that new infectious diseases are just a plane trip away, as the disease was spread rapidly to Canada, the U.S. and Europe by air travelers. Even though the SARS outbreak was relatively short-lived and geographically contained, fear inspired by the epidemic led to travel restrictions and the closing of schools, stores, factories and airports. The economic loss to Asian countries was estimated at $18 billion.

The HIV/AIDS viral epidemic, particularly in Africa, illustrates the economic and social effects of a prolonged and widespread infection. The disproportionate loss of the most economically productive individuals within the population has reduced workforces and economic growth in many countries, especially those with high infection rates. This affects the health care, education, and political stability of these nations. In the southern regions of Africa, where the infection rate is highest, life expectancy has plummeted in a single decade, from 62 years in 1990–95 to 48 years in 2000–05. By 2003, 12 million children under the age of 18 were orphaned by HIV/AIDS in this region.

For an emerging disease to become established, at least two events must occur: 1) the infectious agent has to be introduced into a vulnerable population, and 2) the agent has to have the ability to spread readily from person to person and cause disease. The infection also must be able to sustain itself within the population and continue to infect more people.

Despite significant advances in infectious disease research and treatment, control and eradication of diseases are slowed by the following challenges.

- The emergence of new infectious diseases
- An increase in the incidence or geographical distribution of old infectious diseases
- The re-emergence of old infectious diseases
- The potential for intentional introduction of infectious agents by bioterrorists
- The increasing resistance of pathogens to current antimicrobial drugs
- Breakdowns in public health systems.

Baylor College of Medicine, Department of Molecular Virology and Microbiology, bcm.edu/molvir.

Using Cooperative Groups In The Classroom

Cooperative learning is a systematic way for students to work together in groups of two to four. It provides organized group interaction and enables students to share ideas and to learn from one another. Students in such an environment are more likely to take responsibility for their own learning. Cooperative groups enable the teacher to conduct hands-on investigations with fewer materials.

Organization is essential for cooperative learning to occur in a hands-on science classroom. Materials must be managed, investigations conducted, results recorded, and clean-up directed and carried out. Each student must have a specific role, or chaos may result.

The Teaming Up! model* provides an efficient system for cooperative learning. Four "jobs" entail specific duties. Students wear job badges that describe their duties. Tasks are rotated within each group for different activities so that each student has a chance to experience all roles. For groups with fewer than four students, job assignments can be combined.

Once a model for learning is established in the classroom, students are able to conduct science activities in an organized and effective manner. Suggested job titles and duties follow.

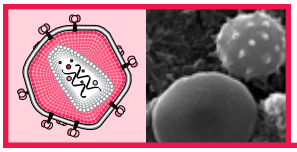
Principal Investigator
- Reads the directions
- Asks the questions
- Checks the work

Maintenance Director
- Follows the safety rules
- Directs the cleanup
- Asks others to help

Reporter
- Records observations and results
- Explains the results
- Tells the teacher when the group is finished

Materials Manager
- Picks up the materials
- Uses the equipment
- Returns the materials

* Jones, R.M. 1990. *Teaming Up!* LaPorte, Texas: ITGROUP.



Red (round) and white (knobby) blood cells. CDC\7320 Janice H. Carr.

ESSAY

The Deadly Cycle

An HIV virus particle is far too small to be seen with an ordinary light microscope. More than one hundred times smaller than the white blood cells they invade, HIV virus particles look like miniature cells—but they are not cells. Rather, HIV particles, like all viruses, are best described as containers of genetic material.

The HIV particle is surrounded by an envelope of cell membrane material, taken from the cell from which it emerged. Inside, the HIV virus contains enough genetic material (in the form of RNA molecules) to direct a host cell to make new virus copies. Viruses cannot live, grow and reproduce on their own. Instead, they must invade the cells of living organisms and force those cells to produce more viruses. This is how viruses cause disease. The term, "virus particle" (or "virion") usually refers to the infectious version of the virus, as it exists outside a host cell.

The surface of an HIV particle typically has between 14 to 73 small projections, referred to as glycoprotein spikes. Glycoproteins (gp) are protein molecules with carbohydrates incorporated into their structure. They are represented by concentric circles on the outside of the paper model used in the previous activity. Two different glycoproteins, gp120 and gp41, comprise each spike on an HIV particle. The numbers, 120 and 41, refer to each protein's molecular weight (an indicator of a molecule's size). The gp120 glycoproteins allow the HIV virus particles to attach to, or "dock" with certain kinds of white blood cells.

HIV cannot survive for long outside the body, and only can be transmitted to another person through body fluids from someone who already has the infection. Once inside the body, HIV particles enter the blood stream and make contact with leukocytes, or white blood cells, the body's chief defenders against infectious diseases. There are five different kinds of leukocytes. However, HIV most often attacks one kind, called a CD4+ cell. CD4+ cells get their name from a particular protein, called CD4, found on the outside cell surface (in other words, these cells are "positive" for the presence of a CD4 protein). CD4+ cells sometimes are referred to as T-cells.

HIV particles—specifically the exterior glycoprotein spikes—attach to CD4 molecules on the surface of CD4+ cells. This connection is similar to that between a lock and key. Once attached, the virus particle fuses with the cell membrane and releases its contents into the cell. After this stage in the infection process, the HIV particle and white blood cell together can begin to reproduce more HIV particles.

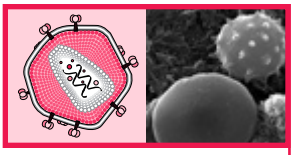
Inside the fatty envelope of an HIV particle is a bullet-shaped core, called the capsid. Made of proteins, the capsid holds the virus's genetic material and triggering enzymes. HIV's genetic material consists of two single-stranded RNA molecules (or ribonucleic acid). The viral RNA strands contain just nine genes, compared to the 20,000 or 25,000 genes in humans. Once HIV RNA is inserted into a cell, an enzyme called reverse transcriptase transcribes, or changes the RNA strands into double-stranded DNA. The viral DNA then integrates with the host DNA in one chromosome within the cell's nucleus. From this point, the virus may remain inactive for many years. Eventually, though, the viral DNA is activated and the cell begins replicating the parts required to make new HIV virus particles—by the hundreds of thousands. In essence, HIV hijacks the cell's functions and turns the cell into a kind of virus factory. Raw materials inside the cell are reworked into new strands of RNA, proteins, and enzymes, which gather just inside the cell wall. Then, the new HIV virus particles bud from the wall of the host cell into the bloodstream.

The HIV replication process eventually overwhelms the host cell until it dies. New HIV particles, millions of them, pass through the blood stream to attach and insert themselves into other leukocytes and begin the replication process again. Over time, the number of white blood cells declines to the point where they can no longer provide protection. Other components of the immune system, such as the lymph nodes, also are affected, and the host body becomes less and less able to defend itself against diseases. A person infected with HIV is diagnosed with AIDS when he or she has one or more serious illnesses associated with HIV, such as pneumonia or tuberculosis, and has dangerously low numbers of infection-fighting white blood cells.

For further details about this process, see the Microbiology animated tutorial "Life Cycle of HIV, a Retrovirus" at http://sumanasinc.com/webcontent/animation.html.

Overview

Students will learn the internal structure of HIV and about its replication cycle.



Red (round) and white (knobby) blood cells. CDC\7320 Janice H. Carr.

TIME

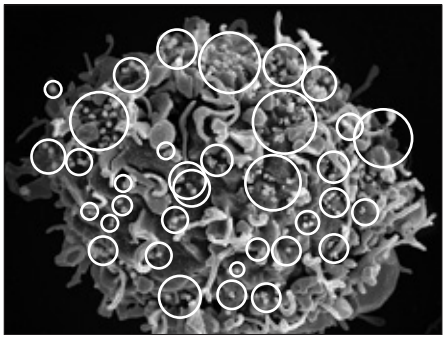
Setup: 30 minutes

Activity: 1 class period

MAKING COPIES OF AN

HIV Particle

Many biologists do not consider viruses to be "living" organisms, because they cannot carry out many of the functions that define life. For example, viruses cannot use food; nor are they able or make copies of themselves ("reproduce") without invading a living cell and redirecting the cell's internal mechanisms to make new virus copies. Outside cells, viruses exist as genetic material (DNA or RNA) surrounded by a protective coat of protein, called a capsid. HIV's capsid contains two strands of RNA.



Numerous tiny HIV particles (in circles) are shown erupting out of a single CD4+ cell.
Photo: National Institute of Allergy and Infectious Diseases, NIH.

Some viruses also wrap themselves in a modified form of the cell membranes from which they emerge. This modified membrane, called an envelope, is studded with proteins that enable the virus to latch onto and infect other cells. HIV and the influenza (flu) virus are examples of viruses that are surrounded by an envelope.

The complete, assembled viral package—consisting of the genetic material, capsid and envelope (when present)—is referred to as a "virus particle" (or virion) to distinguish it from the virus components present inside host cells.

MATERIALS

Teacher (see Setup)

- Images of HIV particles for projection or display (see Setup)
- Slides or transparencies of HIV Virus Particle and HIV Replication sheets
- LCD or document projector, "smartboard" or overhead projector

Per Student

- Assembled HIV particle model with capsid structure from the activity, "Modeling an HIV Particle"
- Copies of HIV Virus Particle and HIV Replication sheets

SETUP

Assemble images of HIV particles into a presentation for projection in your classroom. Images can be found on the following websites.

- Centers for Disease Control and Prevention, Public Health Image Library (phil.cdc.gov)
- *Journal of Nanobiotechnology* (www.jnanobiotechnology.com/content/3/1/6)

You also may download related slides directly from BioEd Online (bioedonline.org). Conduct this as a whole-class activity.

Continued

SCIENCE EDUCATION CONTENT STANDARDS

Grades 5–8

Life Science

- Living systems at all levels of organization demonstrate the complementary nature of structure and function.
- Cells carry on many functions needed to sustain life.
- Disease is a breakdown in structures or functions of an organism. Some diseases are the result of damage by infection by other organisms.
- Every organism requires a set of instructions for specifying its traits. Heredity is the passage of these instructions from one generation to the next.

Grades 9–12

Life Science

- Cells have particular instructions that underlie their functions. Every cell is surrounded by a membrane that separates it from the outside world.
- Cells use and store information to guide their functions. The genetic information stored in DNA is used to direct the synthesis of the thousands of proteins that each cell requires.
- In all organisms, the instructions for specifying the characteristics of the organism are carried in DNA [usually], a large polymer formed from subunits.

HOW CAN AN HIV PARTICLE BE SEEN WHEN IT IS SO SMALL?

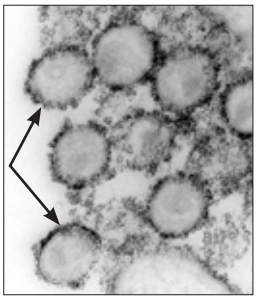
Though 800 times smaller across than a human hair, the HIV particle is larger than most other viruses. Even so, it was very challenging to discover what the HIV virus looks like and how it is constructed. You cannot observe a virus particle on the stage of a normal optical microscope, which works with visible light and has a practical limit for magnification.

An optical microscope's diffraction limit, or resolution (ability to separate two closely spaced objects) is based on the wavelengths of visible light, which range from about 400 to 700 nm (violet to red). The minimum practical resolution (or distance between two objects) is less, about 200 nm. Any specimens closer together than 200 nm appear as a single object under an optical microscope. Consequently, the useful magnification power of optical microscopes is limited to approximately 1,500x. Pushing to a magnification power higher than that leads to hopelessly fuzzy images that are impossible to resolve clearly. Thus, an HIV particle, which measures 120 nm across, is smaller than optical microscopes will allow us to view, even at maximum resolution.

Researcher Jenelyn Ramos, with the National Center for Macromolecular Imaging at Baylor College of Medicine, uses a transmission electron microscope to isolate and examine virus particles and their components.

The National Center for Research Resources, National Institutes of Health, supports several centers dedicated to visualizing 3-D structures within cells and viruses. Photo: National Center for Macromolecular Imaging.

Because virologists (scientists who study viruses) must be able to "see" objects as small as a single nanometer, they require microscopes with much greater magnification power. However, "seeing" is not quite what they do. Rather, they employ a variety of sophisticated microscopes that create images on a computer screen.



In this TEM image of HIV-1 virions, glycoprotein spikes appear darkest gray. Photo: CDC\949 Edwin P. Ewing, Jr., Ph.D.

One such instrument is the transmission electron microscope, or TEM, which directs a beam of electrons through a very thin specimen. The electrons interact with the specimen and are shifted slightly as they pass through. Then, they fall onto a fluorescent screen or a detector, similar to a CCD chip in a digital camera, where the TEM's image is created. Typically, electron microscopes are able to produce useful magnifications one million times the actual size. But under special circumstances, 50 million times magnification has been achieved.

PROCEDURE

1. Use the student-constructed models as a basis for a class discussion about the structure and function of the HIV particle. For example, ask, *What is contained inside the particle?* [capsid and genetic material] *What does the capsid do?* [contain and protect genetic material] *Why might some virus particles also have an envelope?* [provides a way to dock with certain kinds of cells and fuse with the cell membrane]
2. Discuss the main parts of the HIV particle, and their functions. Refer to the illustration on the student sheet, "HIV Virus Particle," (p. 10) to provide more detail.
3. Project microscopic images of the HIV particle and have students compare the outsides of their models to the images. Mention that the double circles on the exterior of the envelope on their models represent the glycoprotein spikes needed by the virus particle to attach to the CD4+ white blood cells.
4. Have students remove their capsid models from the inside of the viral envelope. Ask them to examine the inside of the capsid. Point out the RNA strands and discuss their function: to transmit genetic information to the host cell. Describe the RNA strands as an instruction manual that directs the cell to make virus components. Also mention the reverse transcriptase enzyme and its function, which is to transform the genetic information on the RNA strands into DNA, the genetic code within each host cell.
5. Depending on the ages of your students, you may want to examine the HIV life cycle in more detail.

Use the "HIV Replication Cycle" sheet as a guide. Following are the steps involved in HIV infection of a cell.

a. **Attachment and entry.** The HIV virus bumps into a CD4+ white blood cell, attaches to it, and injects the capsid and its contents into the cell.

b. **Reverse transcriptase.** Once inside the cell, HIV genetic material (in the form of RNA) is converted into a form that is compatible with the cell's genetic information (DNA). In cells, DNA usually is used to produce new RNA through a process called transcription. When RNA is used as a template to produce DNA, as is the case with HIV infection, the process is referred to as "reverse" transcription.

c. **Integration.** The newly formed viral DNA moves into the cell nucleus, where it is spliced into the cell's human DNA. The HIV genetic material may remain dormant or inactive for many years. In this state, HIV is able to "hide" from the immune system and is unaffected by antiviral treatments.

d. **Transcription and translation.** The viral DNA becomes active and directs the cell's machinery to produce the virus components: viral RNA, viral envelope and capsid. This activation can occur many years after initial infection with HIV, and is not yet completely understood.

e. **Assembly and release.** The viral particle is assembled, fuses to the cell membrane and is released by "budding" off the surface of the cell. During the budding process, the new particle wraps itself in part of the host cell's membrane to create the viral envelope. The new virus particles now circulate within the body and are able to invade other cells.

VIRAL REPLICATION

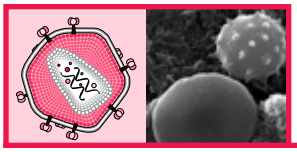
Viruses cannot live, grow and reproduce on their own. Instead, they must invade cells of living organisms and force these cells to produce more viruses. This invasion of healthy cells is how viruses cause disease.

APPLYING BASIC SCIENCE

Researchers use information about the HIV life cycle to develop anti-HIV treatments. One class of drugs blocks the integration of viral DNA into the DNA of the host cell. Another approach prevents the "reverse" transcription of viral RNA into DNA. Many drugs used to fight HIV have harmful side effects, which must be balanced carefully against their value in fighting the virus.

WHAT IS A RETROVIRUS?

HIV is one of a handful of viruses known to reverse the normal pathway through which genetic information is transmitted within cells. Usually, DNA is used to produce RNA, which then directs the assembly of proteins in cells. HIV, however, is able to use its own RNA as a template to produce viral DNA that can be spliced into the DNA of the human host cell.



Red (round) and white (knobby) blood cells. CDC\7320 Janice H. Carr.

HUMAN IMMUNODEFICIENCY VIRUS (HIV)

HIV Virus Particle

GLYCOPROTEIN - gp120
(identifies and initially docks with host cell)

CAPSID
(protein shell)

GLYCOPROTEIN - gp41
(completes docking and assists in fusion with host cell)

VIRAL ENVELOPE
(two-layer lipid membrane)

VIRAL RNA
(two identical strands of genetic material)

MATRIX
(protein shell)

REVERSE TRANSCRIPTASE
(uses viral RNA as a template to produce DNA once the particle is inside a cell)

Illustration by G.L. Vogt and M.S. Young © Baylor College of Medicine.

HIV Replication Cycle

Red (round) and white (knobby) blood cells. CDC\7320 Janice H. Carr.

HIV PARTICLE

HOST CD4+ WHITE BLOOD CELL

1 Attachment and fusion

CD4 Receptors

2 Reverse transcription

HIV RNA

HIV RNA produces viral DNA; enters host cell nucleus

3 Integration

HIV viral DNA

Host cell DNA

Host cell chromosome and DNA

New HIV RNA (genetic material)

mRNA (exits nucleus)

Ribosome (makes viral proteins)

4 Transcription and translation

Viral RNA and proteins

5 Assembly and release

HIV budding from host cell

Immature HIV particle

Mature HIV particle

(not to scale)

Illustration by G.L. Vogt and M.S. Young © Baylor College of Medicine.