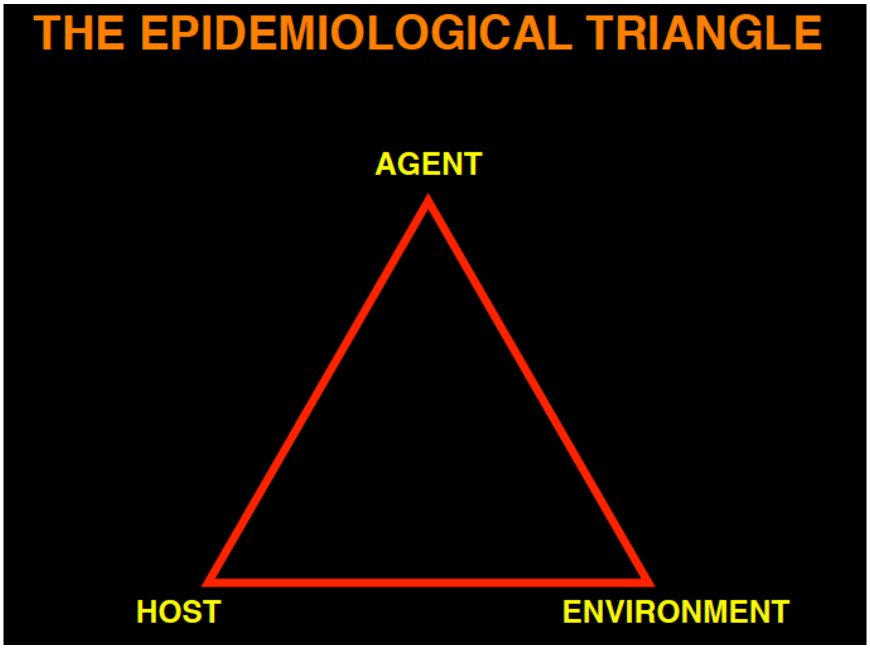
BIOS E-65C
Human Anatomy & Physiology II
Immunology Lab

INTRODUCTION

In 1980, variola virus (Smallpox) was the first ever disease to be completely eradicated through the combination of public health awareness and vaccinia virus vaccination (Fenner 1993). However, this eradication is only limited to nature. Seventy-five laboratories throughout the world ranging from: Africa (5), Americas (18), Southeast Asia (14)*, Europe (29), eastern Mediterranean (3), and western Pacific (6) claimed to have stocks of variola for research purposes in 1975 (Tucker 2009). By 1983, all nations provided written documentation to the World Health Organization (WHO) that all viral stocks of variola were either destroyed or transferred to one of two official WHO Collaborating Laboratories - The US Centers for Disease Control and Prevention (CDC), Atlanta, GA, or The State Research Center for Virology and Biotechnology (VECTOR), Novosibirsk, Russia (Henderson 2014). The WHO has recommended against vaccinating individuals with the vaccinia virus vaccination do to potential side effects. Since 1972 the United States has halted mandatory vaccination, much like the rest of the world. This leaves much of the world population at risk of developing smallpox, should variola virus be introduced back into nature, either accidentally or deliberately.

In 2001 the National research council identified infectious disease and the environment to be one of four areas of environmental science research to be most deserving of research investment. According to the NRC, a program is needed to understand the ecology and evolutionary aspects of infectious and environmental diseases. However, it is challenging to understand these aspects of infectious diseases while simultaneously developing an understanding of interactions among pathogens, receptors/hosts, and the environment in which they thrive, and thereby making it possible to prevent changes in the infectivity and virulence of pathogens that threaten human, animal, and plant health at the population level. Further inquiry is required in examining the effects of environmental changes as selection effects pathogen virulence and host resistance, while exploring its impact on disease etiology, vectors and toxic organisms and developing new approaches to surveillance and monitoring. But what are the factors that are commonly considered important and that contribute to the "epidemiological triangle?" Many researchers look at diseases in terms of their epidemiological triangle, which characterizes a particular disease. (Fig. 1)

* Although China did not respond to the WHO survey, samples of the smallpox virus were being held at the Institute for the Control of Drugs and Biological Products in Beijing.



'Figure 1: A representation of the epidemiological triangle for a disease.

Every epidemiological triangle defines agent, host and environmental characteristics of the disease. An etiologic agent may be any microorganism that causes an infection. When considering the agent it is important to identify the pathogenicity of the agent, it's transmission, it's life cycle and what evasion strategies the agent may use.

Pathogenicity of an agent is its ability to cause disease, which can be characterized by describing its virulence and invasiveness. Virulence refers to the severity of the infection and can be expressed by describing the morbidity (incidence of disease) and mortality (death rate) of the infection. The invasiveness of an organism refers to its ability to invade tissue. *Vibria cholera* organisms are noninvasive, casing symptoms by releasing into the intestinal canal an exotoxin that acts on tissues. In contrast, *Shigella* organisms in the intestinal canal are invasive and migrate into tissue. The life cycle and transmission route of a parasite are usually related. The life cycle of an agent is a series of changes in form than an organism goes through before returning to its starting state. For example the *Plasmodium* that causes the disease malaria has a very specific life cycle (Fig. 2) where the *Anopheles* mosquito introduces the parasites into the persons blood. The parasites travel to the liver where they mature and reproduce. When a fertilized mosquito bites an infected person, immature gametes are taken up in the blood and mature in the mosquito gut where the fuse and form a motile zygote. The zygote develops into a new sporozoite that migrates to the insect's salivary glands and can infect a new vertebrate host.

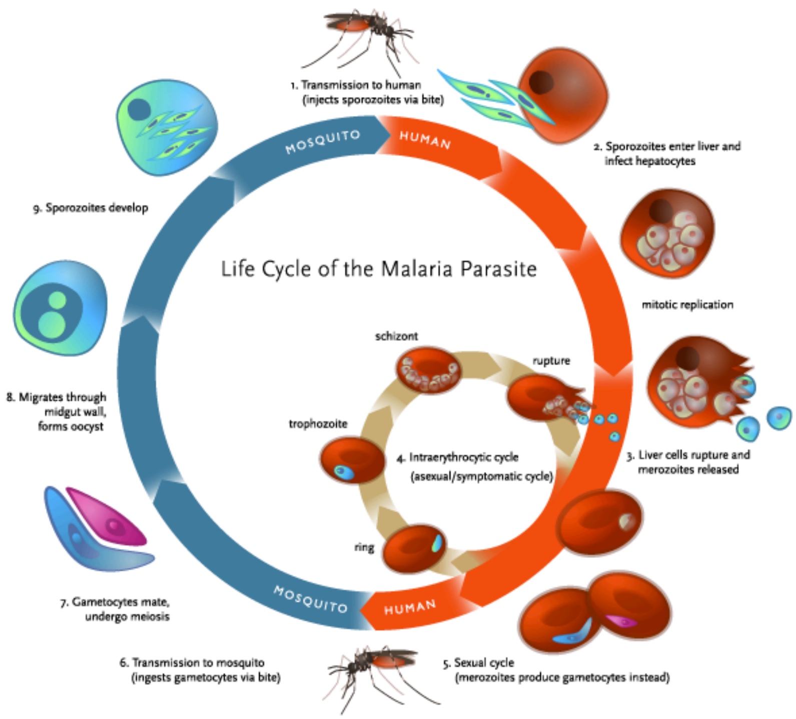


Figure 2: The life cycle of the malarial parasite.

In terms of evasion strategies there are four strategies we are going to consider they are: outrun, lay low, outwit and disable. When an agent outruns it's host it reproduces at a rate that is faster than the host organism can mount an immune response. Organisms can also hide out in various parts of the body effectively "laying low" until conditions are right for the organism to cause a disease (ex. Immune system is suppressed). African sleeping sickness (*Trypanosoma brucei*) is a salivary trypanosome which is usually transmitted by the tsetse fly and it continuously changing its surface glycoproteins which allows the parasite to outwit the immune system. Finally HIV can both outwit and disable the immune system. HIV can hide in the immune system and destroys T-helper cells which disables the immune system.

The second point of the epidemiological triangle is environmental factors. These include encounter rates, both with agents and with individuals that are susceptible to the pathogen, vectors from one organism to another, reservoirs and the seasonality of any pathogen. As populations have evolved from hunter-gatherers to more agrarian societies the types of diseases we encountered have shifted. In hunter-gatherer societies directly transmittable disease were probably rare and instead pathogens with long latencies, dormant life cycle stages, non-human reservoirs and transmission vectors were probably more common.

Do to the nature of the world we live in we must also prepare for bio terroristic events that we may be forced to deal with in the future. Research plays a vital role in developing cures and vaccines to protect society from these threats. As we consider the advantages that research confers upon us to peer into the natural workings of infectious agents, we must also consider the horrible implications that may ensue if said agents are accidentally or maliciously introduced into our world. In recent history, laboratory accidents have caused infectious agents to take the lives of research personal and civilians alike (Silver 2014). On July 1, 2014 six vials, dated to the 1950s, labeled, "variola virus," were found in a refrigerator of a storage facility on the campus of the National Institutes of Health (NIH) in Bethesda, Maryland (Reardon 2014). The vials were found to contain viable DNA from variola. There could quite possibly be other unknown samples of thriving in unknown locations. Furthermore, in 2011 construction works accidentally unearthed a mummified body that had traces of variola virus on it from the mid 1800s. Consider the consequences of introducing an infectious agent, such as variola, into society.

Today's lab will allow you to look at how the interactions between agent factors, host factors and the environment will affect disease transmission. The game Plague Inc. for Ipad/Android devices is a mix of strategy and simulation where your goal is to evolve a deadly, global plague that will wipe out humanity.

LAB PROCEDURE

Open up the plague application and select the "How to Play" button and review the how to play the game. Answer the questions below:

1) Define infectivity, severity and lethality. (2 points)

Once you are familiar with how to play return to the main menu and click on "Play Game" then "New Game". You will start out using a bacteria to take over the world. Click on the "Bacteria" and "Casual" for difficulty level feel free to name your disease anything you would like and answer the questions below:

2) What is the name of your disease? (0.5 points)
3) In which country did you start your disease? (0.5 points)

Once the game begins click on the disease menu in the bottom left corner of the screen and answer the questions below:

4) What are the routes of transmission for your disease? (2 points)
5) What are the symptoms of the disease that you can evolve and which of the body's systems is involved with each of the symptom groups? (2 points)
6) What are the four abilities that you can have your disease evolve with? (2 points)

Proceed through the game evolving your disease as you continue forward. Play the game to its conclusion then answer the questions below.

7) How much of the earth's population were you able to kill? (0.5 points)
8) Which symptoms did you evolve? How did those symptoms increase the transmission of your disease? (0.5 points)
9) Which routes of transmission did you choose? How did the transmission rate change as you evolved the transmission traits? (0.5 points)

10) Which forms of resistance did you evolve in your disease? How did this effect the rate of transmission? (0.5 points)
11) If you did not kill everyone on planet the first time how would you change your game play to be more successful next time? (0.5 points)

If you started your first game in a developed country (ex. USA) try starting the second game in a less developed country (or vice versa). If you started your game is a warmer country try starting it in a cold country for your next game. Based on your second game answers questions 2, 3, 7 – 11 again (0.5 points each). When you have completed two games answer the questions below.

12) How did your mutation rate change with time? (1 point)
    a. Why was the mutation rate slow at the beginning and end of the game? (1 point)
13) How was the cure rate for your pathogen change as you evolved your disease? (1 point)
14) In your opinion how did the simulation accurately reflect what you have seen happening in the real world? How was it inaccurate? (2 points)

AritaI. The Smallpox Eradication Saga. Hyderabad, India: Orient Blackswan; 2010:16-20

D. A. Henderson, I. Arita, Biosecur. Bioterror. **12**, 117 (2014).

G Girard., Plague. Annual Review of Microbiology, Vol. 9: 253 -276 (Volume publication date October 1955) DOI: 10.1146/annurev.mi.09.100155.001345

Jonathan B. Tucker, The Smallpox Destruction Debate: Could a Grand Bargain Settle the Issue? Arms Control Association (2009); http://www.armscontrol.org/act/2009_03/tucker

S. Reardon, Sci. Am. (2014); www.scientificamerican.com/article/vials-of-smallpox-virus-found-unsecured-at-nih/.