adapted from an original work by
Dr. T. Andrew Mobley, Grinnell College

A Dramatic/ Molecular Interpretation of the Influenza Virus's Life Cycle

Goal

The purpose of this activity is for everyone to gain a basic knowledge of the influenza virus on a molecular level and to relate it to a most important global issue…….. a possible flu pandemic. This activity will serve as a capstone for all of the concepts and principles of organic chemistry that were considered throughout the entire course of study.

Activity

Your class will produce and perform a dramatic enactment (a play) depicting the life cycle of a flu virus. Each of six groups will take on one particular aspect of the life cycle. You and the other members of your assigned group will enact a particular part. It will be a very short play, since we do not need to understand all of the details involved in the viral replication process. You will need to interact within your own group to determine what stage actions the "influenza actors" need to perform and develop any speaking lines they may need to deliver. You may want to have a narrator, although it is not necessary. Your group will need to work with the other groups in the performance who precede you and who follow you. They will require information concerning the details of your scene. In fact, some of your lines/actions will need to be performed with the other groups, so careful coordination is a necessity.

Spend some time today organizing your scene, considering your actions and spoken lines, and coordinating with any groups preceding or following you.. The play will be presented in class on Thursday, May 20. Remember that although the purpose of this activity is to learn and use chemistry in an important interdisciplinary context, having fun while doing it is highly recommended. Humor, melodrama and surprise are encouraged.

The Life cycle of the Flu Virus (A Play In Four Scenes With Two Possible Endings)

The actors

Group 1: The Parent Virus Group (Hemagglutinin Proteins)
Group 2: The Cell Membrane Group
Group 3: The Cell Cytoplasm Group
Group 4: The Viral Progeny Group
Group 5: The Neuraminidase Group (Sialidase Enzyme)
Group 6: The Anti-influenza Group (Neuraminidase Inhibitors)

The Play

Scene 1: The Parent Virus recognizes The Cell Membrane and infects the cell
Scene 2: Viral RNA is transported into the cell and cellular machinery in the Cell Cytoplasm builds new virions
Scene 3: The Viral Progeny escape from the cell only to clump together
Scene 4: Neuraminidase cleaves frees Viral Progeny from clumping
Alternate Ending:
Scene 4: The Neuraminidase inhibitor keeps Neuraminidase from freeing Viral Progeny

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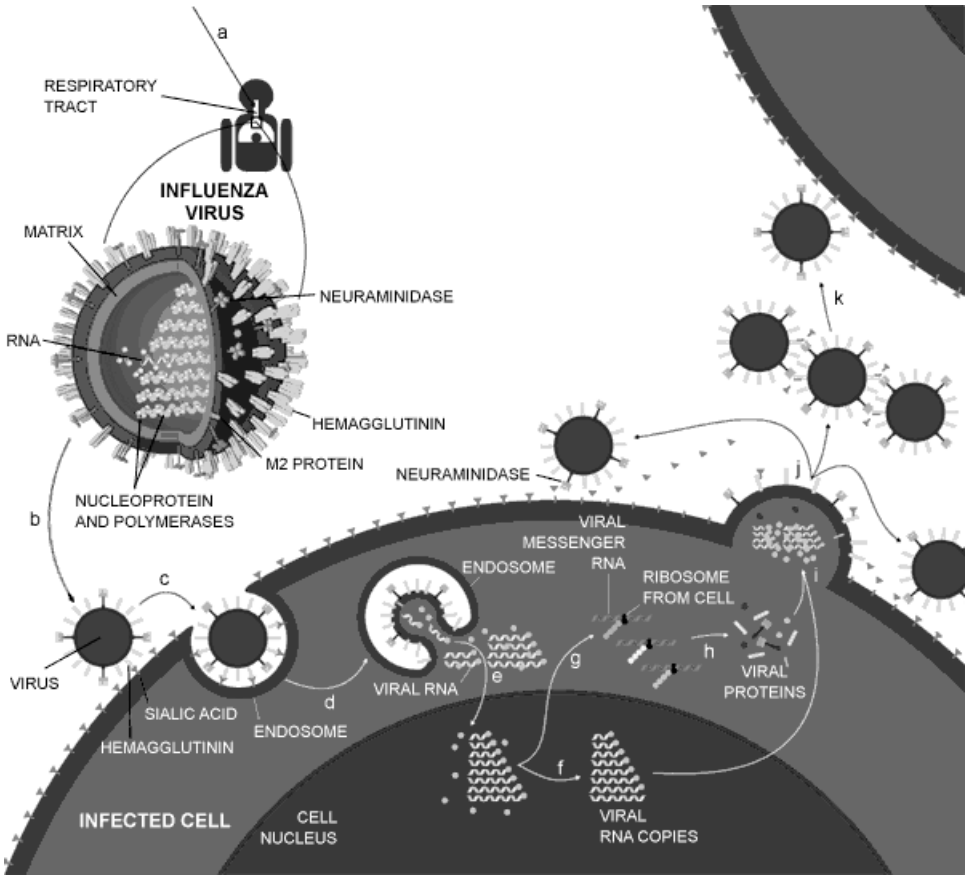
Information on the Flu Life Cycle

Shown below is the picture from the Scientific American article (Laver *et al*) about the flu virus. The description given here (also taken from the article) refers to that picture.

Life Cycle of the influenza virus often involves transmission from one person's airways to another's via water droplets emitted during a sneeze (a). An individual virus (detail, above) enters a cell lining the respiratory tract after a molecule called hemagglutinin on the virus binds to sialic acid on the cell (b). This binding induces the cell to take up the virus (c), which soon dispatches its genetic material, made of RNA, and its internal proteins to the nucleus (d and e). Some of those proteins then help to duplicate the RNA (f) and to produce messenger RNA, which the cell's protein-making machinery uses as a template for making viral proteins (g and h). Next, the viral genes and proteins assemble into new viral copies, or particles (i), and bud from the cell. The particles emerge coated with sialic acid. If that substance remained on the virus and on the cell, the hemagglutinin molecules on one particle would soon attach to the sialic acid on other particles and on the cell, causing the new viruses to clump together and stick to the cell. But neuraminidase on the virus clips sialic acid from the offending surfaces (j), leaving the new particles free to travel on (k) and invade other cells.



Group 1: The Parent Virus Group (Hemagglutinin Protein)

The task of Group 1 is to understand the basics of the cell recognition process. The cell surface is recognized by the virus by recognition of sialic acid by the protein Hemagglutinin.

Group Information

Hemagglutinin is a viral surface protein which is able to detect the presence of sialic acid (a sugar) on the surface of a cell membrane. Many cells (including the epithelial cells lining our lungs) have sialic acid as a terminal sugar on cell-membrane bound carbohydrates. The influenza virus uses this fact to "grab" ahold of the cell membrane surface. Each hemagglutinin protein is actually a trimer of three individual protein strands and hence each hemagglutinin binds to three sialic acids on the cell surface. Following this binding event, the hemagglutinin protein brings the surface of the cell in close contact with the surface of the virus and the virion is absorbed into the cell by a process called endocytosis. Following endocytosis, the virus fuses its membrane with that of the endosome and releases the viral RNA into the cell cytoplasm. This process is described by parts a-d of the Scientific American figure.

Your part in the play is to describe this recognition process. You will work closely together with the Cell Membrane Group to accomplish this task.

Group Instructions

1. Your first duty is to find the Cell Membrane Group and hand them the sealed envelope marked **viral-RNA Instructions**. (This is to simulate the viral infection process. The viral RNA passes from the parent virion through the cell membrane into the cell cytoplasm. The viral membrane group will pass these instructions on to the Cell Cytoplasm Group.)
2. Understand the cell recognition process. Discuss this amongst yourselves until you have a good understanding of it.
3. Determine from the cell membrane group the shape of sialic acid.
4. Design a "protein" capable of recognizing sialic acid. The parts necessary for your hemagglutinin are in your packet. You will need to compare the parts that you have to those being used by the cell membrane people to have one interactions with the sialic acid. Since the hemagglutinin protein is a trimer, you need to have three copies of your recognition site which will simultaneously bind.
5. Working with The Cell Membrane Group work out the lines/actions which describe the hemagglutinin/sialic acid recognition event.
6. Work out the lines/actions which describe the endocytosis process (including passage of the viral RNA information to the cell cytoplasm). This must also be done in conjunction with the Cell Membrane Group.
7. Your last action in the play will be the passing of the viral RNA information through the Cell Membrane to the Cell Cytoplasm. For the play, you should retrieve the **vRNA Instructions** envelope and pass it.

Interaction Scheme

Group 1 (The Parent Virus Group) will be interacting heavily with Group 2 (The Cell Membrane Group).

Group 2: The Cell Membrane Group

The task of Group 2 is to understand the basics of the cell recognition process. The cell surface is recognized by a foreign virus by recognition of the sugar sialic acid by the protein Hemagglutinin.

Group Information

The surface of the cell is covered by (among other things) many different complex carbohydratesThese complex carbohydrates are typically capped at the end by sialic acid. One role for these cell membrane carbohydrates is cell-cell recognition. Unfortunately, the virus has taken advantage of the fact that these sialic acid units are always there and "learned" to recognize them as well. Hemagglutinin is the viral surface protein able to detect the presence of sialic acid on the surface of the cell membrane. Each hemagglutinin protein is actually a trimer of three individual protein strands and hence each hemagglutinin binds to three sialic acids on the cell surface. Following this binding event, the hemagglutinin protein brings the surface of the cell in close contact with the surface of the virus and the virion is absorbed into the cell by a process called endocytosis. Following endocytosis, the virus fuses its membrane with that of the endosome and releases the viral RNA into the cell cytoplasm. This process is described by parts a-d of the Scientific American figure.

Your part in the play is to describe this recognition process. You will work closely with The Parent Virus Group to accomplish this task. After the virus has successfully broken through the cell membrane, you will transmit the viral RNA to The Cell Cytoplasm Group.

Group Instructions

1. Your first duty is to find the Parent Virus Group and receive from them the sealed envelope marked **viral-RNA Instructions**. (This is to simulate the viral infection process. The viral RNA passes from the parent virion through the cell membrane into the cell cytoplasm. You will then pass these instructions on to the Cell Cytoplasm Group.)
2. Understand the cell recognition process. Discuss this amongst yourselves until you have a good understanding of it.
3. In cooperation with the Cell Cytoplasm Group, you will determine the shape of sialic acid. The parts necessary for this task are available from the Cell Cytoplasm Group. These two groups must construct a sialic acid fairly quickly because both the viral hemagglutinin and the viral neuraminidase must build its recognition template from your sialic acid.
4. Working with The Parent Virus Group work out the lines/actions which describe the hemagglutinin/sialic acid recognition event.
5. Work out the lines/actions which describe the endocytosis process (including passage of the viral RNA information to the cell cytoplasm). This must also be done in conjunction with The Parent Virus Group.
6. Your last action in the play will be the passing of the viral RNA information through the Cell Membrane to the Cell Cytoplasm.

Interaction Scheme

Group 2 (The Cell Membrane Group) will be interacting heavily with Group 1 (The Viral Parent Group).

Group 3: The Cell Cytoplasm Group

The task of Group 3 is to understand the basics of the viral replication process. The protein machinery of the cell is hijacked by the RNA from the virus and used to produce viral proteins.

Group Information

Following cell surface recognition by the virus, the virus fuses with the cell membrane and introduces viral RNA into the cell cytoplasm. The viral RNA then is used for two processes. The first process is controlled by viral proteins and is the process of transcribing many new copies of viral RNA. The second process is handled by cell protein machinery and involves the translation of viral RNA into new viral proteins. These proteins are then assembled into two major structures. One of these structures is the capsid (or nucleocapsid after the incorporation of viral RNA) which contains several viral proteins. The second structure assembles itself at the cell membrane and is primarily new hemagglutinin and neuraminidase proteins. The capsid then moves to the cell membrane and buds as a progeny virion. This process is described by parts e-i of the Scientific American figure.

Your part in the play is to describe this replication process. You will work closely with The Progeny Virus Group.

Group Instructions

1. Your first duty is to find the Cell Membrane Group and receive from them the sealed envelope marked **viral-RNA Instructions**. This envelope will contain these instructions. (This is to simulate the viral infection process. The viral RNA passes from the parent virion through the cell membrane into the cell cytoplasm.)
2. Understand the viral replication process. Discuss this amongst yourselves until you have a good understanding of it.
3. With The Cell Membrane Group determine the shape of sialic acid.
4. Working with the Viral Progeny Group, you will need to find out from the Parent Virus Group the shape of the hemagglutinin receptor site and manufacture three copies.
5. Working with the Viral Progeny Group, you will need to find out from the Viral Progeny Group the shape of the neuraminidase active site and manufacture three copies.
6. Working with the Viral Progeny Group, you will need to make three copies of the Viral RNA Instructions.
7. Once you have all of the parts necessary, work with the Viral Progeny Group to assemble three complete new virions.
8. Work out the lines/actions which will describe the viral replication events.
9. Work out the lines/actions which describe the budding process. This must be done in conjunction with the Viral Progeny Group.
10. Your last action in the play will be the passing of the Viral RNA Instructions to the Viral Progeny Group.

Interaction Scheme

Group 3 (The Cell Cytoplasm Group) will be interacting heavily with Group 4 (The Viral Progeny Group).

Group 4: The Viral Progeny Group

The task of Group 4 is to understand the basics of the viral replication process and the budding process. The protein machinery of the cell is hijacked by the RNA from the virus and used to produce viral proteins. After assembly of new virus proteins, the new virus buds from the infected cell.

Group Information

Following cell surface recognition by the virus, the virus fuses with the cell membrane and introduces viral RNA into the cell cytoplasm. The viral RNA then is used for two processes. The first process is controlled by viral proteins and is the process of transcribing many new copies of viral RNA. The second process is handled by cell protein machinery and involves the translation of viral RNA into new viral proteins. These proteins are then assembled into two major structures. One of these structures is the capsid (or nucleocapsid after the incorporation of viral RNA) which contains several viral proteins. The second structure assembles itself at the cell membrane and is primarily new hemagglutinin and neuraminidase proteins. The capsid then moves to the cell membrane and buds as a progeny virion. Upon budding, however, the new virus is coated with sialic acid (both from the cell membrane and from intracellular fluid – mucus). Because the new viral particles have active hemagglutinin they bind to the sialic acid and clump together. This process is described by parts e-j of the Scientific American figure.

Your part in the play is to describe this replication and budding process. You will work closely with The Cell Cytoplasm Group.

Group Instructions

1. Your first duty is to find the Cell Cytoplasm Group and receive from them the sealed envelope marked **viral-RNA Instructions**. This envelope will contain these instructions. (This is to simulate the viral infection process. The viral RNA passes from the parent virion through the cell membrane into the cell cytoplasm which then follows the instructions encoded by the viral RNA. Upon assembly of progeny viral particles, each new virion receives a copy of the Viral RNA Instructions)
2. Understand the viral replication process. Discuss this amongst yourselves until you have a good understanding of it.
3. Working with the Cell Cytoplasm Group, you will need to find out from the Parent Virus Group the shape of the hemagglutinin receptor site and manufacture three copies.
4. Working with the Cell Cytoplasm Group, you will need to find out from the Viral Progeny Group the shape of the neuraminidase active site and manufacture three copies.
5. Working with the Cell Cytoplasm Group, you will need to make three copies of the Viral RNA.
6. Once you have all of the parts necessary, work with the Cell Cytoplasm Group to assemble three complete new virions.
7. Work out the lines/actions which describe the budding process. This must be done in conjunction with the Viral Progeny Group.
8. Work out the lines/actions which will describe the clumping of Progeny Virions due to sialic acid binding to hemagglutinin.
9. In scene 4 you will be freed from the sialic acid/hemagglutinin binding by neuraminidase. Work with the Neuraminidase Group to determine the lines/actions which will described the cleavage of sialic acid from the viral surfaces.

Interaction Scheme

Group 4 (The Viral Progeny Group) will be interacting heavily with Group 3 (The Cell Cytoplasm Group).

Group 5: The Neuraminidase Group (Sialidase Enzyme)

The task of Group 5 is to understand the basics of the sialic acid cleavage process. After budding from the infected cell, progeny virions are covered in sialic acid (both from the cell membrane and from intracellular fluid – mucus) and therefore clump together because of binding between the hemagglutinin receptor and the sialic acid. The neuraminidase enzyme cleaves the sialic acid from the membrane surface thus freeing the progeny virions.

Group Information

Upon budding from the cell membrane progeny virion clump together. This is due to the presence of sialic acid attached to their surfaces. The hemagglutinin protein on the viral membrane binds to the attached sialic acid and the various virion particles clump together. It is necessary to cleave the linkage between the sialic acid and the viral surface for the progeny viruses to be able to migrate from the site. This reaction is accomplished by the viral neuraminidase enzyme. It recognizes and tightly binds sialic acid and then cleaves the molecule from the surface. This process is described by part k of the Scientific American figure.

Your part in the play is to describe this sialic acid recognition and cleavage process.

Group Instructions

1. Understand the sialic acid recognition/cleavage process. Discuss this amongst yourselves until you have a good understanding of it.
2. Determine from the Cell Membrane Group the shape of sialic acid.
3. Design an "enzyme" capable of recognizing sialic acid. The parts necessary for your neuraminidase are in your packet. You will need to compare the parts that you have to those being used by the cell membrane people to have at least three interactions with the sialic acid. In addition to the interactions with sialic acid, you must provide one other type of possible interaction (this will be utilized by the anti-influenza drug).
4. Working with the Viral Progeny Group, work out the lines/actions which sialic acid recognition and cleavage by neuraminidase.
5. In the alternate ending, you will be working with the Anti-influenza Group (the Neuraminidase Inhibitors). Work out the lines/actions for the plugging of the neuraminidase active site by the Anti-influenza plug drug.

Interaction Scheme

Group 5 (The Parent Virus Group) will be interacting heavily with Group 4 (The Viral Progeny Group) and Group 6 (The Anti-influenza Group).

Group 6: The Anti-influenza Group (Neuraminidase Inhibitors)

The task of Group 6 is to understand the basics of the neuraminidase inhibition process. The active site of neuraminidase is inhibited by tight binding with a molecule quite similar to sialic acid, but even more strongly bound.

Group Information

Upon budding from the cell membrane progeny virion clump together. This is due to the presence of sialic acid attached to their surfaces. The hemagglutinin protein on the viral membrane binds to the attached sialic acid and the various virion particles clump together. It is necessary to cleave the linkage between the sialic acid and the viral surface for the progeny viruses to be able to migrate from the site. This reaction is accomplished by the viral neuraminidase enzyme. It recognizes and tightly binds sialic acid and then cleaves the molecule from the surface. If this neuraminidase could be inhibited, the success of the viral infection could be limited. The action of the new anti-influenza drugs is to act as a competitive inhibitor for the active site of the neuraminidase. This means that the influenza drug looks much like sialic acid, but one or two additional intereactions increase the binding constant of the drug such that it is a better binder than sialic acid. This results in competitive inhibition of the neuraminidase reaction, thus slowing the ability of the viral particles to escape from clumping.

Your part in the play is to describe this inhibition process.

Group Instructions

1. Understand the neuraminidase inhibition process. Discuss this amongst yourselves until you have a good understanding of it.
2. Determine from the Viral Progeny Group the shape of the active site for sialic acid cleavage.
3. Design a “drug” capable of binding more tightly in the pocket than sialic acid. The parts necessary for your hemagglutinin are in your packet. You will need to compare the parts that you have to those being used by the Viral Progeny Group to have at least four interactions with the active site.
4. Work out the lines/actions which describe the inhibition. This must be done in conjunction with the Viral Progeny Group.

Interaction Scheme

Group 6 (The Anti-influenza Group) will be interacting heavily with Group 5 (The Viral Progeny Group).