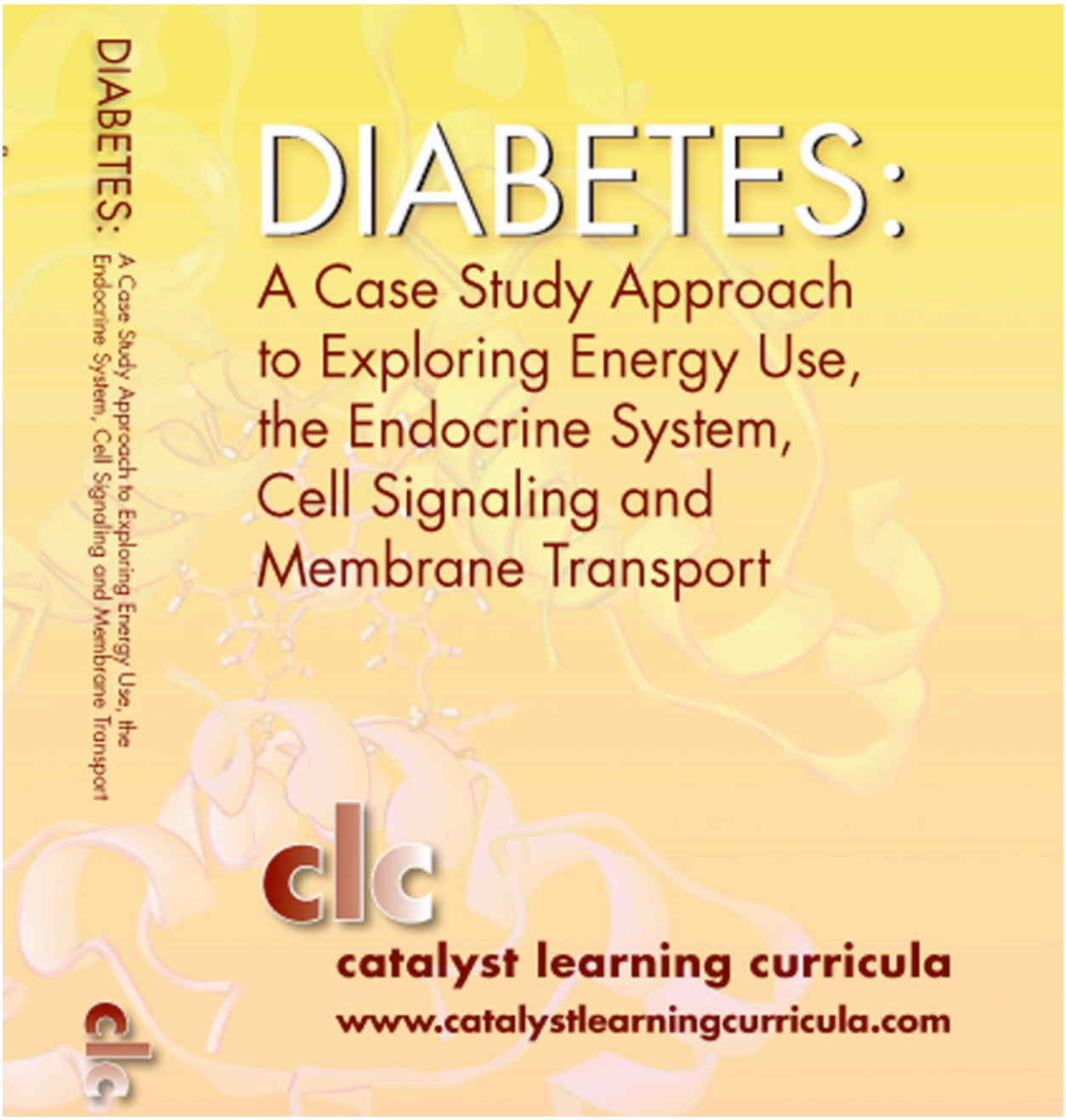

DIABETES:
A Case Study Approach to Exploring Energy Use, the Endocrine System, Cell Signaling and Membrane Transport
clc
DIABETES:
A Case Study Approach
to Exploring Energy Use,
the Endocrine System,
Cell Signaling and
Membrane Transport
clc
catalyst learning curricula
www.catalystlearningcurricula.com

DIABETES:

22 experiential, higher-order thinking activities for Advanced Placement* (AP) or International Baccalaureate** (IB) high school or college students, covered in 30-42 class periods

With the rising incidence of obesity and Type 2 Diabetes diagnoses, most students will be touched directly or indirectly by this disease sooner rather than later. In this case study, students will learn about the importance of carbohydrates as the basis of cellular energy and the consequences that occur when supply exceeds demand. They will examine the delicate control mechanisms that regulate appetite and satiety to insure a constant supply of energy for cellular processes. The interaction of insulin and glucagon with various tissues allows the students to observe the logistics of cell membrane transport, cell communication and regulatory feedback, all of which make homeostasis possible. Students will discover the impact unregulated blood sugar has on the kidneys and other organs and what actions can be taken to avoid irreparable damage. Together, the lessons in this case study will impress upon students that, while there are issues of genetic predisposition that affect metabolism, the choices they make now will largely determine their probability of developing diabetes as adults.

This case study was specifically designed to provide complete, stand-alone coverage of the following topics required by the AP College Board and the IB program:

Photosynthesis and Other Energy Acquisition Processes
The Biochemistry of Carbohydrates
Cell Respiration
Cell Membrane Transport
Kidney Structure and Function
Homeostatic Regulation of Blood Sugar
Cell Communication
Signal Reception and Response Mechanisms
Synthetic Production of Proteins
Translation of mRNA for Secretory Proteins
Diet and Nutrition

This case study also offers partial coverage of the following required topics:

Population Genetics
Bioethics

By Kristen Dotti
Catalyst Learning Curricula ©2012
59 Clemmons Street, Asheville, NC 28801
828-687-0807 • kristen.dotti@catalystlearningcurricula.com

Diabetes: A Case Study for Advanced Science
Table of Contents

(This sample contains Activity 11 and 15 only, which do not correspond with the page numbers listed below. The full curriculum can be purchased at the Catalyst Learning Curricula website)

Activity Eleven: The Insulin Message

Teaching time: 1 class period of 50 minutes

Objectives:

a) For students to understand how a concentration of glucose triggers the pancreas to release insulin
b) For students to visualize how insulin changes the level of sugars in the bloodstream
c) For students to observe how cell signals regulate homeostasis
d) For students to connect the symptoms of diabetes to problems occurring at the cellular level

Materials:

For each student: 1 pair of scissors, 1 sealable plastic bag, and 1 copy of each of the following handouts, all of which follow this lesson plan: "Insulin Signal Cascade and Cell Response Puzzle," "Cell Signal Pathway for Insulin," and "Insulin Cell Signal Pathway Reflection Questions." For the class: 1 skein of yarn, signs that read, "Small Intestine," "Pancreas," "Bloodstream," "Muscle/Brain/Adipose/Liver Cell" and "New Cell," and 1 shirt sticker for each role listed in step 6 below.

Procedure:

1. Prior to class, use yarn to outline a section of small intestine villi, a pancreatic cell, a muscle cell and a capillary (see the "Cell Signal Pathway for Insulin" diagram for the recommended design of these items) on the classroom floor or in an open space nearby that you can use during class (for example, on the stage of the school auditorium, in an empty classroom, the library, an outside yard or the lunchroom). Within each space outlined with yarn, place the appropriate sign ("Small Intestine," "Pancreas," etc.).
2. Distribute the "Insulin Signal Cascade and Cell Response Puzzle" and "Cell Signal Pathway for Insulin" handouts and ask the students to cut apart the descriptions listed on the puzzle sheet.
3. Allow the students time to arrange the descriptions on the "Cell Signal Pathway for Insulin" diagram such that each description matches the flow of the cell communication pathway for insulin.
4. As students complete the puzzle, allow them to share their final guesses with a neighbor to compare answers and rearrange their puzzle descriptions as needed. They may need a few minutes to discuss and defend their answers before agreeing on a final arrangement.
5. Go over the puzzle when all the students have completed it to make sure their final arrangement is correct. Tell the students to flip each description over and write the number that corresponds to the correct location on the diagram so that they are able to check their work when they solve the puzzle again, later.

When finished, they can place the puzzle pieces in a sealable plastic bag, to save the puzzle for use as a pre-exam review game.

6. Choose sixteen students to demonstrate the insulin cell signal pathway. Give each student one of the following roles:

(3) Glucose	1 Glucose signal receptor
	1 Glucose secondary messenger
(1) Insulin	1 Tyrosine kinase (alpha subunit)
	1 Tyrosine kinase (beta subunit)
	1 Secondary messenger for tyrosine kinase
(3) PI-3K	1 ATK secondary messenger for PI-3K
(2) GLUT-4	1 PIP secondary messenger for PI-3K
(1) MAP-K	1 Secondary messenger for MAP-K

7. Ask the students to act out the process of the insulin signal cascade while you narrate the steps in the appropriate order using the Teacher's Version of the puzzle (which follows this lesson plan) and giving each student who is playing a role time to step into the simulation when called on. During the first run-through of the simulation, just follow the basic steps of the descriptions the students arranged in the puzzle, so that they make the leap from the 2-dimensional sequence to the 3-dimensional simulation. Ask the students to point out the message or chemical signal, the transduction and the response when they see each taking place. Tell the students who are participating in the simulation that when they have acted out their role correctly, in a memorable way, they will be asked to choose a member of the audience to take their place during the subsequent simulation (encourage them to get into their roles and have fun).
8. After completing the first run-through of the simulation, ask the students who have mastered their part to either stay in for the second run or teach another student how to perform their role. In the second run-through, add the information bookended by asterisks on the Teacher's Version of the puzzle, to provide greater depth and detail for those students who are ready for it.
9. After the simulation, ask the following questions:
    a. Which molecules are acting as chemical messages or signals? *(Glucose acts as a signal for the beta cells of the pancreas and insulin acts as a signal for the muscle, brain, liver and adipose cells.)*
    b. How is each signal received? *(Glucose will pass through a glucose-specific GLUT-2 channel embedded in the membrane of a beta cell. When glucose arrives on the interior side of the cell membrane, it initiates a cascade of signals that allows the release of insulin. Insulin docks on the outside of a cell using a protein receptor but never moves to the cell interior. Some cell signals are small, nonpolar molecules and are able to pass through the cell membrane without a protein receptor or protein channel. These molecules, often hormones, may interact with the nuclear envelope, enter the nucleus through a pore, and possibly initiate transcription directly.)*

c. Can all cells receive the same signals? *(No, cells only receive the signals for which they have a matching cell surface receptor protein or specific channel protein.)*
d. Which cells would you guess have insulin receptors, or stated another way, which cells have a high demand for glucose? *(Most cells use glucose, but some cells, like muscle, liver, brain and adipose cells, have a higher demand and can lower blood glucose concentrations as needed.)*
e. Describe one positive feedback mechanism in the insulin cell signal pathway. *(AKT and P70 are both examples of positive feedback because they push the pathway further from the normal range, which results in an even greater response. This is an example of an amplification of a signal.)*
f. What is the result when positive feedback is given to this pathway? *(The signal is amplified and the result takes the cell further from its normal rate of glucose use.)*
g. Explain one way this pathway may move into a negative feedback loop. *(Any of the phosphorylation pathways can be blocked, or the enzymes that phosphorylate secondary messengers can be regulated, inhibited, or otherwise kept from completing their role in the pathway.)*

10. Ask the students to work on the Reflection Questions (either alone or with a partner). When everyone has finished, discuss their answers (or grade their answers as an independent assessment).

Insulin Signal Cascade and Cell Response Puzzle

Cut out the descriptions on this page and place them in the appropriate places, numbered 1-18, on the "Cell Signal Pathway for Insulin" sheet, to depict the normal sequence of events that occurs when blood sugars rise.

The phosphorylation of phosphatidylinositol-3-kinase (PI-3K) initiates a cascade of other reactions in the cell.

Molecules triggered by MAP-kinase promote the expression of genes in the nucleus that are responsible for growth and division.

After a person has eaten food, glucose is absorbed from the lumen of the small intestine into the bloodstream, which raises the concentration of circulating blood sugars.

The mitogen-activated protein kinase (MAP-kinase) supplies energy to other molecules in the cytoplasm by adding phosphate groups to them so they become active.

The dimer shape of the cell receptor triggers the addition of a phosphate group to two enzymes on the interior surface of the muscle cell.

Molecules triggered by PI-3K initiate cell division (mitosis).

Glucose entering the beta cell transmits a message through the cytoplasm to the DNA in the nucleus, telling it to transcribe and translate the protein insulin.

Excess glucose is stored as glycogen.

Glucose is sent to the mitochondria to supply the cell with ATP (used to phosphorylate molecules in the signal pathway as well as perform other actions).

GLUT-4 channels in the cell membrane allow glucose to diffuse from the exterior to the interior of the cell.

Insulin engages with a receptor on the exterior surface of a muscle cell.

Molecules triggered by PI-3K restrict apoptosis (cell death).

Circulating glucose molecules enter beta cells through GLUT-2 channels embedded in the membrane.

Molecules triggered by PI-3K promote the expression of genes used for making chains of glycogen, proteins and lipids.

The cell receptor (called a tyrosine kinase) forms a dimer by bringing together its alpha and beta subunits.

Molecules triggered by MAP-kinase initiate the cell cycle (mitosis).

Insulin is secreted into the bloodstream by the beta cells of the pancreas.

PI-3K binds to vesicles containing GLUT-4 channel proteins and allows them to merge with the cell membrane.

Insulin Signal Cascade and Cell Response Puzzle

Teacher's Version

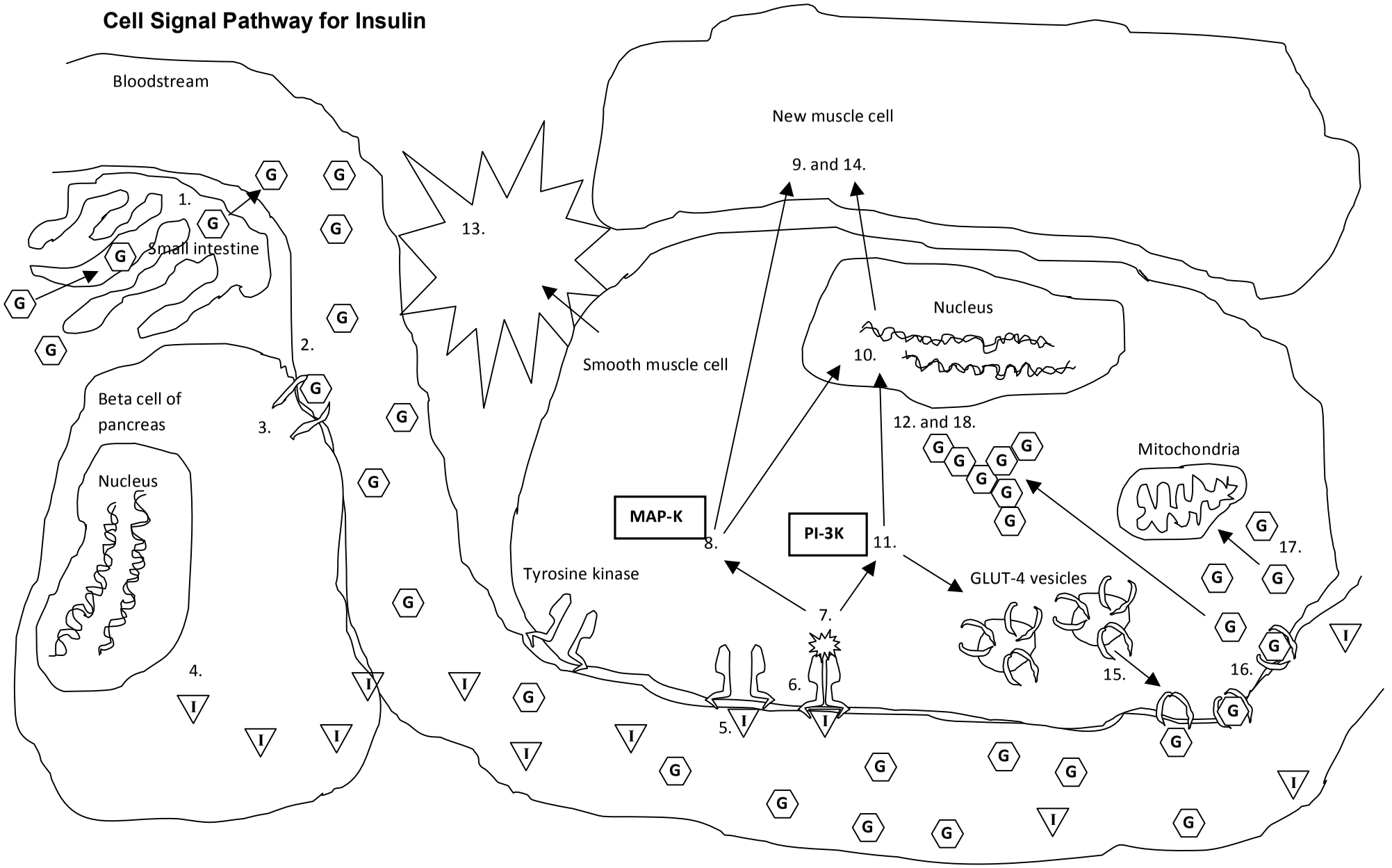
Following is the correct sequence of the descriptions the students have been asked to arrange; use this to check that they have arranged their puzzle pieces correctly on the "Cell Signal Pathway for Insulin" diagram. The information bookended with asterisks gives more details about the signal pathway than students will need when first working on the puzzle. You can share these details when the students perform a simulation of the pathway, building an additional layer of depth and understanding after the students understand the basic process of message→receptor→transduction→secondary messenger→response.

1. After a person has eaten food, glucose is absorbed from the lumen of the small intestine into the bloodstream, which raises the concentration of circulating blood sugars.
2. Circulating glucose molecules enter beta cells through GLUT-2 channels embedded in the membrane. *This is the ligand or chemical message to the beta cells.*
3. Glucose entering the beta cell transmits a message through the cytoplasm to the DNA in the nucleus, telling it to transcribe and translate the protein insulin. *This is the transduction. There is a series of secondary messengers that pass the signal through the nucleus, adhere to the DNA and activate the gene for transcription.*
4. Insulin is secreted into the bloodstream by the beta cells of the pancreas. *This is the response. Point out that many of the secondary messengers that control a cell signal pathway like this one can be interrupted (negative feedback), promoted (positive feedback), or regulated.*
5. Insulin engages with a receptor on the exterior surface of a muscle cell. *All cell surface receptors are specific to a single ligand and are not found on all cells or on every cell of a particular type in the same concentration. The presence or absence of each type of cell receptor is controlled by genes that are regulated by some other pathway.*
6. The cell receptor (called a tyrosine kinase) forms a dimer by bringing together its alpha and beta subunits. *There are many different tyrosine kinase receptors, each with a different type of ligand-binding site. Tyrosine kinases are a class of cell receptors that act as switches to turn something on or off inside a cell.*
7. The dimer shape of the cell receptor triggers the addition of a phosphate group to two enzymes on the interior surface of the muscle cell. *Phosphorylation can be spontaneous or it may occur using secondary messengers. In this case, secondary messengers are used.*
8. The mitogen-activated protein kinase (MAP-kinase) supplies energy to other molecules in the cytoplasm by adding phosphate groups to them so they become active. *This occurs through several secondary messengers which are specific to the pathways that lead to steps 9 and 10, below.*
9. Molecules triggered by MAP-kinase initiate the cell cycle (mitosis).
10. Molecules triggered by MAP-kinase promote the expression of genes responsible for growth and division. *Secondary messengers like cyclic AMP (cAMP) are used to promote and regulate events concerning growth and cell division.*
11. The phosphorylation of phosphatidylinositol-3-kinase (PI-3K) initiates a cascade of other reactions that are activated by PI-3K. *The presence of certain promoter molecules such as AKT and P70 push this process along in a positive feedback loop

while the presence of too much glucose in the cell or other factors cancel the action of PI-3K, sending the signal into a negative feedback loop.*

12. Molecules triggered by PI-3K promote the expression of genes used for making chains of glycogen, proteins and lipids. *This occurs through several secondary messengers along a complex signal pathway that include a chemical called ATK. These materials allow cell growth which is part of the positive feedback loop also being promoted by MAP-K.*
13. Molecules triggered by PI-3K restrict apoptosis (cell death). *This occurs through several secondary messengers along a complex signal pathway that includes a chemical called ATK. ATK is a positive feedback mechanism that pushes the cell toward growth and division instead of self-destruction.*
14. Molecules triggered by PI-3K initiate cell division (mitosis). *This occurs through several secondary messengers along a complex signal pathway that includes ATK. Notice that the MAP-K pathway also leads to this result, reinforcing the positive feedback loop.*
15. PI-3K binds to vesicles containing GLUT-4 channel proteins and allows them to merge with the cell membrane. *This occurs when PI-3K phosphorylates PIP2 to convert it to PIP3. PIP3 goes on to phosphorylate two more secondary molecules, unlocking the waiting GLUT-4 vesicles and allowing them to move to the cell membrane.*
16. GLUT-4 channels in the cell membrane allow glucose to diffuse from the exterior to the interior of the cell.
17. Glucose is sent to the mitochondria to supply the cell with ATP (used to phosphorylate molecules in the signal pathway as well as perform other actions for cell growth and division). *The demand for readily available phosphate groups should be apparent based on how often phosphorylation is used to energize cell processes.*
18. Excess glucose is stored as glycogen. *If the cell taking up the glucose is an adipose cell, the cell will swell and the person will gain weight.*

Cell Signal Pathway for Insulin



Insulin Cell Signal Pathway Reflection Questions

Please answer the following reflection questions on a separate sheet of paper using your "Insulin Signal Cascade and Cell Response Puzzle" pieces and the "Cell Signal Pathway for Insulin" diagram as needed.

1. A high concentration of glucose in the cell can trigger a negative feedback mechanism to halt the actions of PI-3K such that GLUT-4 channels are not transferred to the cell membrane. Explain three things that would occur if the GLUT-4 vesicles did not move to the cell membrane.
2. Cancer is defined as the uncontrolled growth of cells. Explain how a diet with an overabundance of simple carbohydrates might be linked to cancer.
3. Why do you think cells are programmed to absorb excess glucose and store it in adipose tissue instead of releasing the molecules through an excretion process?
4. Type 1 diabetes occurs when a person is not able to secrete insulin. Using your puzzle pieces and the "Cell Signal Pathway for Insulin" diagram, determine which steps would occur in a type 1 diabetic and which steps would be hindered or would not occur. List the steps that would occur in a type 1 diabetic.
5. Using your answer to question #4 above, what problems would a type 1 diabetic have, based on the gaps in their cell signal pathway?
6. Hypothesize three things that may be occurring in a person's body that would result in a low level or absence of insulin.
7. In type 2 diabetes, a person is able to make insulin but their cells no longer respond to the insulin signal. Most type 2 diabetics have responded to insulin for some portion of their life and did not always have diabetes. Use your puzzle pieces and the "Cell Signal Pathway for Insulin" diagram to determine which steps would occur in a type 2 diabetic and which steps would be hindered or lost. List the steps that would occur in a type 2 diabetic below:
8. Using your answer from question #7 above, what problems would a type 2 diabetic have, based on the gaps in their cell signal pathway?
9. Hypothesize three things that may be occurring in a person's body that would result in a low level of insulin or lack of response to insulin.
10. Prior to treatment, type 1 diabetics tend to lose weight, while type 2 diabetics tend to gain weight. Explain why each situation might occur based on what you understand about the interplay between insulin and glucose.

Insulin Cell Signal Pathway Reflection Questions

Teacher's Version

1. A high concentration of glucose in the cell can trigger a negative feedback mechanism to halt the actions of PI-3K such that GLUT-4 channels are not transferred to the cell membrane. Explain three things that would occur if the GLUT-4 vesicles did not move to the cell membrane. *(Glucose would not be able to diffuse into the cell, blood sugar concentrations would remain elevated, the cell may not have glucose available to the mitochondria, the cell may not have a surplus of glucose and therefore may not make glycogen, triglycerides or other molecules for growth and storage.)*

2. Cancer is defined as the uncontrolled growth of cells. Explain how a diet with an overabundance of simple carbohydrates might be linked to cancer. *(Cells that are growing out of control need a steady supply of glucose to continue growth, the formation of additional proteins/lipids/glycogen and cell division. When glucose is present in the bloodstream, cells take it up and it promotes cell growth and cell division. In addition, an increase of glucose leads to an increase in insulin. The insulin signal halts apoptosis and promotes cell growth and division. Apoptosis is a normal process that allows cells to die when they have divided too many times or when they are at risk of having errors in their DNA. Halting apoptosis may lead to higher rates of cancerous growth particularly when mitosis is promoted by the same cell signal.)*

3. Why do you think cells are programmed to absorb excess glucose and store it in adipose tissue instead of releasing the molecules through an excretion process? *(Because most organisms must deal with periods when food is not available, they have evolved mechanisms to absorb excess energy when it is available rather than dispose of it. It is only in recent evolutionary history that humans have had such an abundance of calories and nutrients that can be eaten in a constant manner. Prior to this point, a mechanism for glucose storage was advantageous for survival rather than detrimental.)*

4. Type 1 diabetes occurs when a person is not able to secrete insulin. Use your puzzle pieces and the "Cell Signal Pathway for Insulin" diagram to determine which steps would occur in a type 1 diabetic and which steps would be hindered or lost. List the steps that would occur in a type 1 diabetic: *(Only step #1 could be assumed to occur in a type 1 diabetic since steps #2 and 3 may or may not occur and step #4 is known not to occur. Steps #5-18 cannot occur without the presence of insulin in the bloodstream.)*

5. Using your answer from step #4 above, what problems would a type 1 diabetic have, based on the gaps in their cell signal pathway? *(Type 1 diabetics would have high levels of sugars in their bloodstream and not enough glucose in their cells for cell processes. High circulating blood sugar would give them the symptoms of hyperglycemia: they may become sleepy and weak because*

their brain, muscle and liver cells are low on ATP, they may get headaches, be thirsty, and/or have trouble concentrating or have blurred vision.)

6. Hypothesize three things that may be occurring in a person's body that would result in a low level or absence of insulin. *(Type 1 diabetes is caused by the loss of beta cells and students may state this in their response, or they may hypothesize any situations or scenarios that would lead to a lack of insulin response. For example, some might hypothesize that the beta cells may not have the glucose signal receptor on the surface of their cell membranes, the beta cells may have a defect in any of the secondary messengers that relay the glucose signal to the DNA for transcription, or the beta cells may have a mutation in the gene that makes insulin such that the molecule is not made or is incorrectly transcribed, translated or exported.)*

7. In type 2 diabetes, a person is able to make insulin, but their cells no longer respond to the insulin signal. Most type 2 diabetics have responded to insulin for some portion of their life and did not always have diabetes. Use your puzzle pieces and the "Cell Signal Pathway for Insulin" diagram to determine which steps would occur in a type 2 diabetic and which steps would be hindered or lost. List the steps that would occur in a type 2 diabetic: *(In a type 2 diabetic, steps #1-7 should occur, since their body can respond to glucose and can produce insulin. However, it is likely that some or all of steps #8-18 would be hindered or stopped because of an interference or negative feedback mechanism in the signal pathway. If the person has unregulated blood sugar levels that are too high, the cells may have an excess of glucose and the negative feedback mechanism would shut down the process that would activate the GLUT-4 channels.)*

8. Using your answer from question #7 above, what problems would a type 2 diabetic have, based on the gaps in their cell signal pathway? *(A type 2 diabetic would have circulating blood sugar levels that are too high because their cells are not taking up the glucose that is available. High blood sugar levels would give them the symptoms of hyperglycemia, which include thirst, frequent urination, headaches, blurred vision and confusion. Type 2 diabetics can go into a coma if dehydration becomes too severe.)*

9. Hypothesize three things that may be occurring in a person's body that would result in a low level of insulin or a low level of response to it. *(A person may have a low level of insulin or low response to insulin because their cells are sufficiently supplied with glucose and negative feedback mechanisms are cancelling the cell signal. There may also be a defect or mutation in any of the secondary messengers of these pathways or the GLUT-4 channel proteins, but these possibilities are less likely since the person has been able to respond to insulin in the past and a mutation in a single cell would not be able to spread to other cells to cause a systemic failure. In the United States,*

excessive weight and lack of physical exercise account for the onset of 95% of type 2 diabetes cases.)

10. Prior to treatment, type 1 diabetics tend to lose weight, while type 2 diabetics tend to gain weight. Explain why each situation might occur based on what you understand about the interplay between insulin and glucose. *(Type 1 diabetics lose weight because they do not have enough glucose in their cells for normal cell functions. There is no storage of excess glucose in adipose tissue because these cells are not able to take up glucose as needed. Type 2 diabetics tend to gain weight because this condition is often brought on by continuous levels of high blood sugars. High levels of blood sugars can be due to alcohol consumption that is excessive and routine, the consumption of sugary drinks throughout the day or a constant supply of high calorie foods that are rich in carbohydrates. After the muscle cells have taken in what they need, adipose and liver cells take up the excess glucose and store it as triglycerides or glycogen, causing weight gain and eventually a lack of cellular response to the insulin signal.)*

Activity Fifteen: Regulating the Translation of Insulin

Teaching time: 1 class period of 50 minutes

Objectives:

a) For students to understand the general process of translation
b) For students to discover some examples of additional information that may be present in an RNA message
c) For students to recognize some of the mechanisms that allow proteins to be secreted by a cell

Materials:

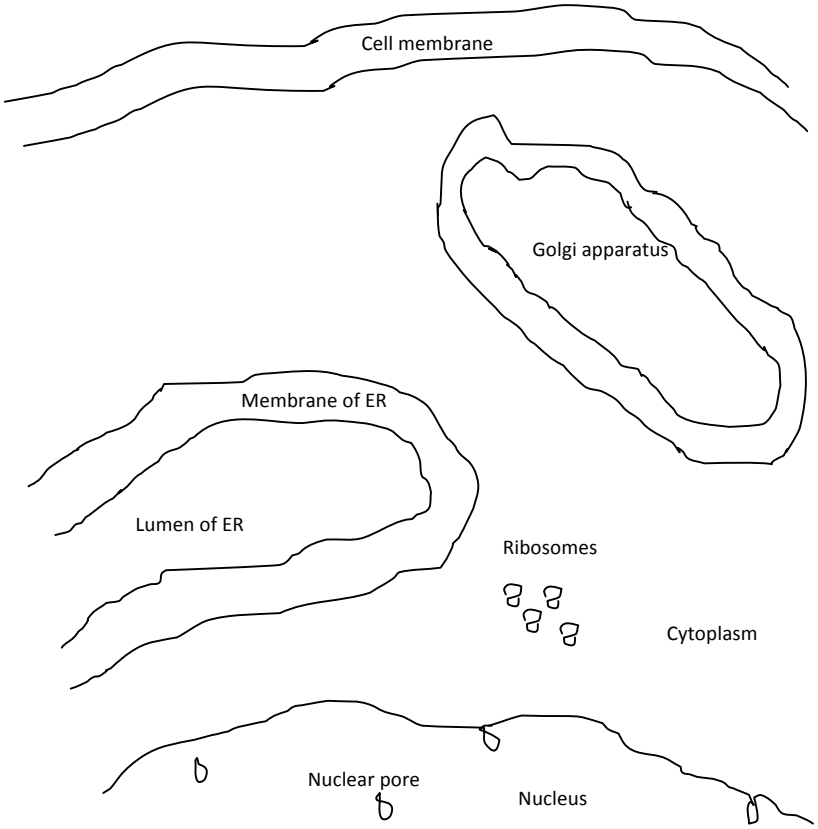
For each pair of students: 1 copy each of "The Translocation and Exportation of Insulin," "Properties of Amino Acids," and "Simulation Labels" handouts, all of which are located within or following this lesson plan, 4 chenille sticks or pipe cleaners, each of a different color, 1 6cm section of a foam swim noodle (pool toy) (obtain swim noodles in two different colors for the class) or foam pipe insulation (if you use this, you won't be able to have two different colors, which is okay), 1 15cm section of a foam swim noodle (pool toy) (the entire class can have the same color) or foam pipe insulation, 4 sheets of colored cardstock paper (in a single color) or one sheet of easel paper, plus a couple of sheets of paper in a different color (for use to create cut-outs), 2-4 markers, 1 sheet of white copy paper, 1 small bowl or cup full of yellow beads (exactly 68 yellow beads total will be used by each pair of students if none are lost during the activity), 1 small bowl or cup full of blue beads (7 total), white beads (19 total), red beads (10 total) and green beads (6 total) (other colors can be substituted if necessary, but you will need to make notations on the handout, in which specific colors of beads are referenced), three twist ties, scissors and tape (scissors and tape can be shared among the class), and 1 video recording device such as a camera, smartphone, etc. (optional). For the class: 1 computer with Internet access so you can show a short animation of the mechanism that occurs due to the insertion signal: http://en.wikipedia.org/wiki/File:Translation.gif and a short animation of the movement of a secretory protein from the ER to the Golgi and out of the cell: http://www.phschool.com/science/biology_place/biocoach/cells/endoreview2.html.

Special Note: To achieve the same objectives using a different activity, you may choose to purchase the "Insulin mRNA to Protein Kit" from 3D Molecular Designs (www.3dmoleculardesigns.com) or borrow the class kit from Milwaukee School Of Engineering's lending library (http://cbm.msoe.edu/teachRes/library/index.html) and perform the main activity with your students.

Procedure:

1. Pair the students or allow them to pair themselves, and ask them to pick up supplies for today's activity then guide the students through the steps below.

2. Ask the pairs of students to each use a large sheet of easel paper (or multiple sheets of colored cardstock taped together to form one big piece of paper) to create a backdrop of the interior of a cell. Their backdrop should show the nucleus (only partially) and nuclear pores, the cytoplasm, ribosomes, a close-up of the membrane and interior of the endoplasmic reticulum (with at least 10cm in the lumen of the ER), a close-up of the Golgi apparatus and the cell membrane, with all structures labeled appropriately (if you choose, you can let them use the labels that follow this lesson plan). An example backdrop is shown below:



3. Ask the student pairs to each cut long strips about 1cm wide down the length of their piece of white copy paper.
4. Using the printed mRNA sequence the students "found" during their RNA-RNA hybridization simulation in Activity Fourteen, ask the students to tape the strips of blank paper they have just created behind the sequence to make the strip wide enough along its entire length to allow labeling of each region. The students may have taped the B chain sequence and the A chain sequence to the mRNA strip when they performed the hybridization, but if they did not already do this, ask them to do so now. Ask them to label the following regions in the space above each corresponding stretch of sequence: signal peptide, B chain, C chain, A chain.
5. The students might have noticed that there are additional nucleotides upstream from the nucleotides that code for the protein chains and downstream from the

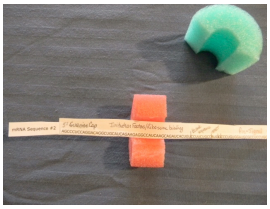
end of the A chain. Ask the students if they can imagine what role this additional information might play in the translation of insulin. Discuss any ideas the students might have, or explain some of the sequence information presented below. (These are just a few of the most commonly discussed examples of mRNA translational regulation; there are new regulatory mechanisms based on mRNA sequences currently being discovered. If you have time, an excellent extension activity would be to ask the students to each look into a different gene regulatory mechanism specifically for mRNA.):

a. Some of the additional information on an mRNA strand includes the sequence of nucleotides that tells the ribosome where to begin translating the amino acid chain. The most important signal is the start codon, which will be the first amino acid that begins the polypeptide chain. Ask the students to look for the AUG start codon about 60 nucleotides from the beginning of their mRNA strand. Ask them to label the AUG "start codon." All of the sequence information that follows the start codon but precedes the B chain is used to translate the signal peptide.
b. The nine nucleotides prior to the AUG start codon and one nucleotide after the AUG start codon are called the Kozak sequence. These nucleotides give additional help to the ribosome to confirm the location of protein synthesis. Certain combinations of nucleotides in the Kozak sequence give the ribosome a stronger start signal while other combinations are not as clear and so the rate and frequency of translation is regulated by how closely this sequence matches the strongest signal. Ask the students to label the nine nucleotides upstream (to the left) of the start codon the "Kozak sequence."
c. Prior to the amino acid start sequence (AUG) and following the sequence for the A chain, the mRNA has additional nucleotides that act as gene regulatory mechanisms. For example, upstream of the start codon, the 5' end of the mRNA strand will have a guanine base that has been phosphorylated and methylated—this modified base is called the 5' guanine cap. The 5' guanine cap helps in transporting the mRNA out of the nuclear pore and prevents degradation of the sequence by allowing the translation initiation proteins (eIF-4F and eIF-4G) to bind on this region of the mRNA. Specifically, the translation initiation factors block the enzymes that would normally break down mRNA stands in the cytoplasm, and they help the ribosome find the proper location on the mRNA on which to bind. This binding site, which is upstream of the start codon (called the Shine-Dalarno sequence in prokaryotes), will determine how strongly the mRNA can attract ribosomes. Ask the students to write "5' guanine cap" on the far end of their mRNA strip and write "Initiation factors" and "Ribosome binding site" in the space between the cap and the Kozak sequence.
d. At the end of the mRNA strand, there are additional regulatory sequences, one of which is the repeating adenosine nucleotides that are called the Poly-A tail. Because the chemistry of the cytosol slowly cuts nucleotides apart from one another, starting from the 3' end, the length of the Poly-A

tail determines how long the mRNA will be present in the cytoplasm and available for translation. As the mRNA tail slowly degrades, the message will eventually inhibit ribosomes from translating the information. A messenger RNA molecule can be present in the cytoplasm for a few minutes or up to a few hours, which directly results in either a lesser or greater quantity of a protein being synthesized. Ask the students to label the Poly-A tail found at the 3' end of their mRNA sequence.

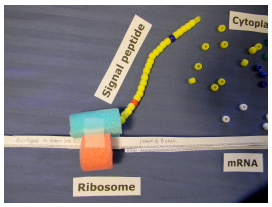
e. Other topics you may choose to introduce, discuss or ask the students to research regarding the regulation of translation include but are not limited to: initiation factor binding sites (eIF-2a, eIF-4E, eIF-4F, eIF-4G, etc.), phosphorylation of the binding factors, mRNA repression, 48s ribosome repression, structured internal ribosome entry sites (IRES), initiation transacting factors (ITAFs), etc. One reference for some of these topics: http://genomebiology.com/2006/7/12/332

6. Ask the student pairs to take their 6cm section of foam swim noodle, cut it in half and trade one piece with another student who has a segment of a different color. Ask the students to now cut in half the segment they received from their neighbor, such that there are two segments that are each approximately 3cm wide, and ask them to set one of these 3cm segments aside. The original foam segment they cut and one of the halves cut from the piece they obtained from trading will be used to simulate the large and small subunits of a ribosome.
7. Ask the students to place on their desk the cell backdrop they created, and ask them to lay their labeled mRNA strand in the nucleus. Narrating the entire process aloud to themselves, ask the students to explain how the mRNA transcript leaves the nucleus through a pore and enters the cytosol. As they narrate, they should attach the small subunit of their foam ribosome to the region of the mRNA labeled "Initiation factors" or "Ribosome binding site" and then move the small subunit forward along the mRNA strand until it is lined up with the AUG start codon. At the start codon, the students can add the large subunit of the ribosome and tape these two pieces together before beginning the next step (while narrating the process aloud throughout).



Attachment of the small subunit to codon.

Attachment of large subunit at AUG start initiation factors.

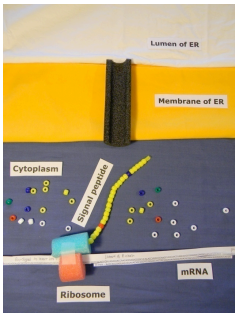


First part of translated protein chain emerging from the ribosome.

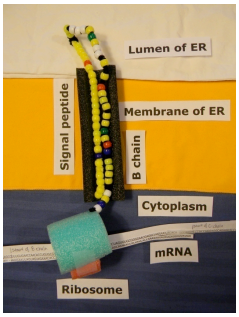
8. Using their cup of colored beads, a chenille stick and their Codon-Anticodon Pairing Chart from the previous activity or from their textbook, ask the students to translate the signal peptide of the mRNA sequence into a polypeptide chain by sliding beads that correspond to the color key on their "Properties of Amino

Acids" chart onto a piece of chenille stick—yellow beads for nonpolar amino acids, white beads for polar amino acids, etc.

9. After all the students have translated the signal peptide, ask them what characteristics the peptide chain exhibits. The students should notice that 22 of the 24 amino acids in this peptide chain are hydrophobic (nonpolar) and thus readily associate with oily environments rather than watery environments. Explain that this sequence of nonpolar amino acids will act as a signal for the ribosome to bind to a channel in the exterior wall of the endoplasmic reticulum (ER). This membrane protein, called a translocation channel, allows growing polypeptide chains to enter the lumen of the ER. Let the students know that any protein chain used in the cytosol will be translated directly into the cytosol, while any protein that is destined to be secreted will need to enter the endomembrane system by being inserted into the ER during translation. Show the students the following short animation so they can see the mechanism that occurs due to the insertion signal: http://en.wikipedia.org/wiki/File:Translation.gif. The animation shows the small and large subunits of the ribosome coming together at the start codon of a strand of mRNA. The transfer RNA molecules are shown bringing amino acids to the ribosome and the amino acids attached to each tRNA are being connected to produce a growing protein chain that is emerging out of the top of the large subunit. After the signal peptide is produced, the ribosome migrates to the ER and the signal peptide feeds the rest of the polypeptide into the lumen of the ER through the translocation channel. You may have the students watch the animation once, then play it again but this time narrate it, then play it a third time, asking the students to each narrate the process to themselves as they watch.
10. Ask the students to use the 15cm segment of a foam swim noodle or foam pipe insulation they were given to make a translocation channel across the membrane of the ER on their cell backdrop. Show them the diagram on the next page or the photos below, so they can see how to set up the channel and insert the signal peptide into the channel such that it feeds the growing polypeptide into the lumen of the ER.



Protein chain emerging from the ribosome.

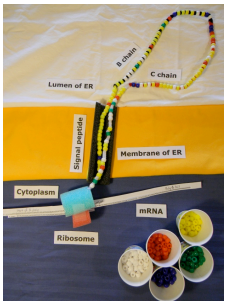


B chain spooling into the ER through the channel.

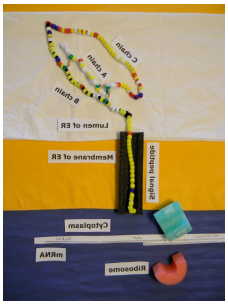
11. Ask the students to narrate the process quietly to themselves as they attach the signal molecule to the translocation channel and feed the peptide chain into the ER. When they have set up the signal protein in the translocation channel, ask

the students to move the ribosome down the mRNA strand and continue making the B chain of insulin by threading the appropriate color beads onto a new color of chenille stick attached to the end of the first chenille stick. The ribosome should be up against the exterior membrane of the ER at the opening of the translocation channel from this point on in the simulation.

12. When the students have completed the B chain, they can feed most of the chain into the lumen of the ER through the translocation channel and move the ribosome down to the C chain sequence. They should then add another color of chenille stick to the growing polypeptide molecule and select the appropriate amino acids (beads) to create the C chain.
13. When the students have completed the C chain, they can feed most of the chain into the lumen of the ER through the translocation channel and move the ribosome down to the A chain sequence. They should then add another color of chenille stick to the growing polypeptide molecule and select the appropriate amino acids (beads) to create the A chain.



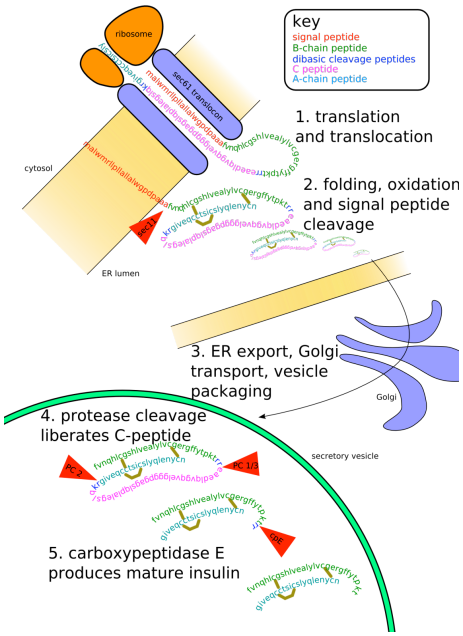
Entire insulin protein spooled into the ER.



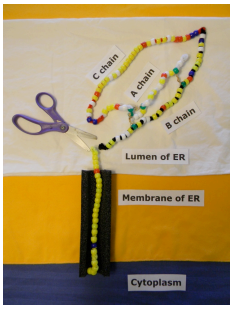
Ribosome detaching at the STOP codon.
Disulfide bridges forming between cysteines.

14. When the students, and the ribosome, arrive at the stop codon UAG, the ribosome should be untaped so that each subunit can fall free, releasing the mRNA and the polypeptide. The remaining A chain should be fed into the lumen of the ER, folding in such a way that the molecule arrangement resembles the diagram on the next page (you may choose to project this image on a screen for students to refer to).
15. Point out the folded shape that the molecule has taken due to the presence of the C chain, showing the students the diagram. Ask the students to notice the position of the cysteine residues and ask them if they know what interesting property these amino acids have. If the students are unaware, have them refer to their "Properties of Amino Acids" chart and ask them to note the position of the sulfur atom on these amino acids (the cysteine residues). Remind the students that the sulfur atoms on the residues can form salt bridges with sulfur atoms on adjacent cysteine amino acids. Ask the students to use the three twist ties to simulate covalent bonds that form between the sulfur atoms of the cysteine residues. Students can reference the diagram to determine which residues pair with one another on the A and C chains.

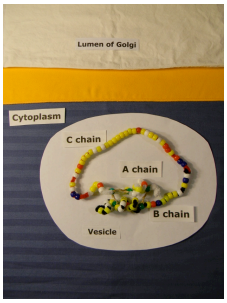
The Translocation and Exportation of Insulin



16. Explain to the students that once the entire polypeptide is translocated into the lumen of the ER, the B, C and A chains will be cut free of the signal sequence by an enzyme located in the lumen of the ER. Ask the students to simulate the enzyme with the scissors and cut the insulin molecule free of the signal molecule.



Signal peptide is cut free by enzymes in the ER.

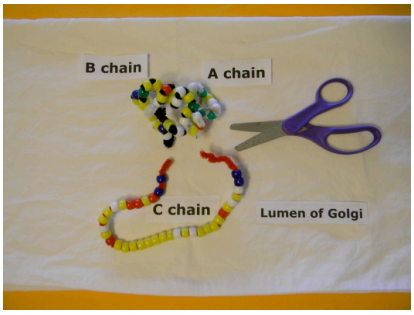


Insulin molecule is shipped to the Golgi.

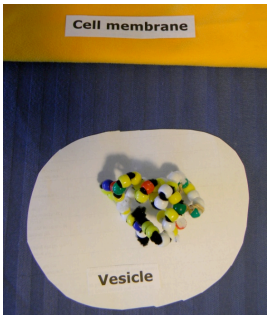
17. Ask the students to model the movement of the insulin protein from the lumen of the ER into a vesicle by having them simulate pinching off a portion of the ER with the insulin molecule trapped inside (they can do this by cutting a piece of cardstock the same color as the ER or using some other available prop to serve the same purpose). Remind them to quietly narrate the entire process aloud to themselves as they perform the simulation.
18. Tell the students to move the vesicle to the *cis* side (the side closer to the nucleus and ER) of the Golgi apparatus and allow it to merge with the membrane of the Golgi so the insulin molecule can be released into the interior

of the Golgi. Again, the students can decide how best to depict this process with the supplies available, but they should narrate the process while they simulate it, being sure to use the appropriate terminology. Here is a link to a short animation of the movement of a secretory protein from the ER to the Golgi and out of the cell, so that your students can see this process in sequence: http://www.phschool.com/science/biology_place/biocoach/cells/endoreview2.html

19. Tell the students that the C chain of the molecule will be cut out by enzymes in the lumen of the Golgi and the A and C chains will coil into alpha helices to create the final tertiary structure needed to become a mature molecule of insulin. Ask the students to simulate the enzymatic process using their scissors and then coil the two strands into a globular, three-dimensional shape. Encourage the students who are driven to do so to look up the three-dimensional structure of insulin on the Internet and fold their beaded strands into the tertiary form of this molecule (remind the students that some of the diagrams on the Internet will be of the insulin hexamer and the molecule they have created thus far is a monomer).



C chain is removed by enzymes in the Golgi.



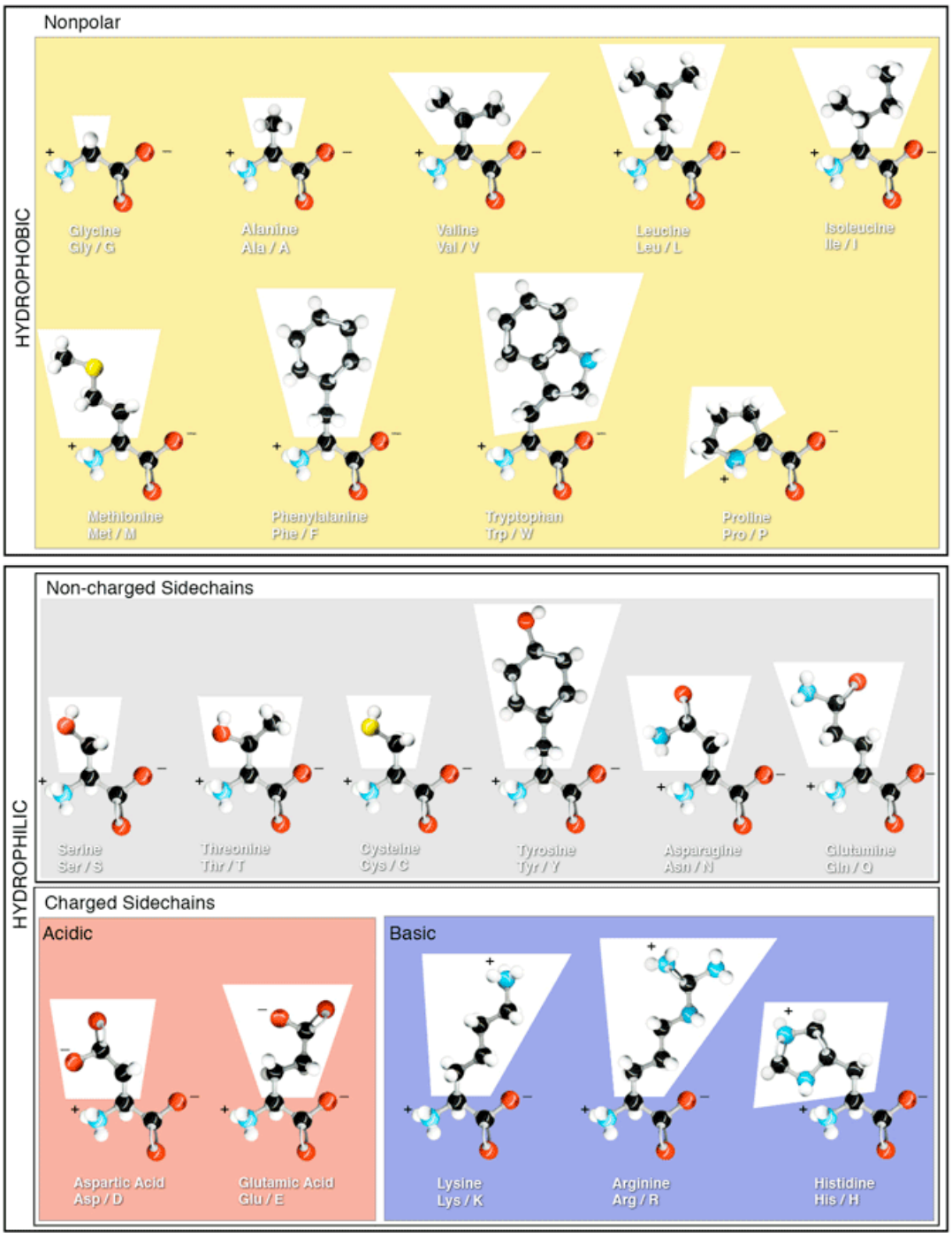
Inactive insulin is held ready in cytoplasm.

20. Ask questions, such as the following, to check for understanding before the students continue:
    a. What is the purpose of the signal peptide? *(The signal peptide is necessary for helping insulin enter the endoplasmic reticulum so it can be secreted from the cell.)*
    b. What is the purpose of the C chain? *(The C chain helps hold the A and B chains together in the proper alignment so disulfide bridges can form between the chains using the correct cysteine amino acids.)*
    c. If the A and B chains will spontaneously combine when in the presence of one another, explain the evolutionary significance of the C chain. *(Organisms that are more efficient at metabolizing consumed food will be more fit to function, therefore dependably efficient metabolism favors the survival and reproduction of an organism. If an organism has the C chain as part of the insulin gene, the A and B chains of insulin will be guaranteed to "find" one another in the lumen of the ER and therefore the molecule translated will be guaranteed to finish the tertiary and quaternary folding necessary to eventually become an active protein. And although*

the A and B chains will spontaneously combine in the correct arrangement to make a functional protein in a test tube or when secreted by separate genes in a host organism, when these two chains are translated as one continuous peptide, they may be held too close together for the disulfide bridges to form between them without errors occurring. The spacer region that is made by the C chain may favor the accuracy of covalent bonding and allow a greater number of proteins to form successfully, resulting in an organism with efficient metabolism.)

21. Ask the students to complete their simulation of the process by which insulin is made: ask them to pinch off a vesicle from the Golgi apparatus and anchor the vesicle in the cytoplasm near the cell membrane. Tell the students that when this beta cell receives an influx of glucose through the GLUT2 channels, a signal cascade will lead to the movement of the vesicle to the cell membrane, so that the insulin can be released into the bloodstream. Ask the students to pick up any available prop (that can be used to represent a glucose molecule) to simulate glucose approaching the outside of the cell to trigger the release of insulin into the bloodstream. Remind the students to narrate the steps as they simulate them, using the proper terms. **If you choose to extend this activity, you can challenge the students to uncover the details of the signal pathway that begins with an influx of glucose and results in the release of insulin in a beta cell; and the students can then add these steps to their simulation.**
22. When all of the students have completed the simulation, ask that each student in a pair take a turn simulating the entire process in a fluid, continuous manner, with their partner recording their actions (if possible). Tell the students that the process can be narrated by either the person who is simulating it or the person who is recording it, however, each student must get experience narrating the simulation as well as performing the actions of the simulation. The videos of the students' simulations can be turned in for a grade, posted to the class Web site or shown on parents' night or open house so that adults or other students can ask them about this process.

Properties of Amino Acids



Key:

Nonpolar amino acids will be represented with yellow beads
Polar amino acids will be represented with white beads
Basic amino acids will be represented with blue beads
Acidic amino acids will be represented with red beads
Cysteine will be represented with a green bead

Simulation Labels

Lumen of ER

Membrane of ER

Membrane of ER

Ribosome

B chain

C chain

A chain

Cytoplasm

Cytoplasm

Cell membrane

Lumen of Golgi

Vesicle

Golgi

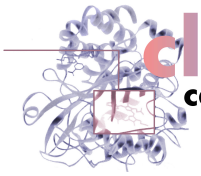
mRNA

Signal peptide

Outside of cell

Nuclear pore

Nucleus



Special Topics Curricula — **$175** (per licensed user)

Incorporating Math and Statistics into AP/IB Science
(14 lesson plans covering 20-22 class periods)

AIDS: A Case Study Approach to Exploring the Immune System, Viruses and Community Health
(20 lesson plans covering 28-32 class periods)

Diabetes: A Case Study Approach to Exploring Energy Use, the Endocrine System, Cell Signaling and Membrane Transport
(22 lesson plans covering 30-42 class periods)

Hemoglobin: A Case Study Approach to Exploring Proteins, the Cardiorespiratory System and the Evolution of a Gene Family
(24 lesson plans covering 32-44 class periods)

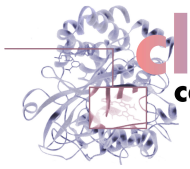
Full-year Curricula — **$570** (per licensed user)

AP® Biology Daily Lesson Plans
Full-year curriculum for AP*/IB Biology

Experimental Biology Daily Lesson Plans
Full-year curriculum for general, Pre-AP*/IB or Honors Biology using inquiry methods

AP® Environmental Science Daily Lesson Plans
Full-year curriculum for AP*/IB or general Environmental Science

** AP College Board is not involved in the production or the endorsement of this curriculum*



Billing Information:

Name of Billing Contact Person:

_____

Address:

_____

_____

Telephone: _____

Email: _____

Shipping Information:

Name of Shipping Contact Person:

_____

Address:

_____

_____

Telephone:_____

Email:_____

Name of School: _____

Name of Licensed User(s): _____Email of Licensed User(s): _____

How did you hear about Catalyst Learning Curricula Daily Lesson Plans? _____

A curriculum license extends to only one teacher. Additional copies must be purchased for each teacher who will be using the curriculum in a given school or school district. Please contact us using the email address below to receive a discount for multiple user licenses.

Product Title: AP Biology, APES, or Experimental Biology; or Name of the Case Study, Game or Syllabus	**Quantity** (number of users – one user per license)	**Format Desired** (CD or download)	**Price**

Shipping ($0 for download or $14/CD, $14 per game): _____

Taxes (6.75%-8.25% sales tax, depending on local tax rates, for NC residents only): _____

Total: _____

Payment Method:

Purchase Order Number: _____

Checks should be made out to Catalyst Learning Curricula

Credit Card purchases will have an additional $22 charge added to the total.

Card Number: _____ Expiration date: _____

Name on card: _____3-digit security code: ____ Billing zip code: _____

You may either email a picture of this completed form to Kristen.Dotti@CatalystLearningCurricula.com or mail the completed form to:
Kristen Dotti, 59 Clemmons St., Asheville NC 28801 (Ph: 828-687-0807)