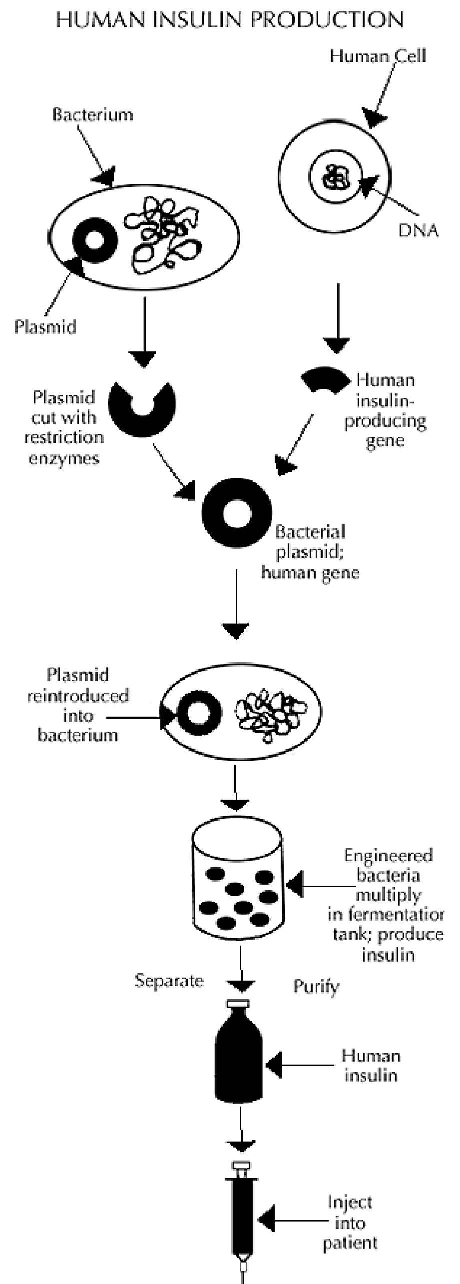
Genetic Engineering Activity

NC Essential Standard 3.3.2

Summarize how transgenic organisms are engineered to benefit society.

Follow the steps below to create a transgenic organism**.**



Pharmaceuticals produced with genetic engineering technology are administered to patients by traditional methods.

Step #1: **CLEAVE DONOR DNA**.

Cut out the gene for the trait you want to transfer from the donor organism's DNA by using a restriction enzyme. In this example, we will be using the restriction enzyme EcoRI to cut out the gene that makes human insulin. EcoRI recognizes the DNA sequence CTTAAG and GAATTC, cleaving (cutting) between the A/G and G/A.

a. Find the human cell. Extract the donor DNA by cutting along the SOLID line.
b. Represent the cuts made by the restriction enzyme EcoRI by cutting along the DOTTED lines. You have now cut out the gene to make human insulin – keep this! Notice how the gene has sticky ends. The remaining donor DNA is no longer needed. Discard it.

Step #2: CLEAVE PLASMID.

A vector carries the desired gene of the donor to the host cell, which could be a bacterium, an egg cell or a virus. In this activity, our host (target) cell will be a bacterium. The most commonly used vectors are viruses and plasmids. In this activity, the vector will be a plasmid. Remember, a plasmid is a circular form of DNA found in a bacteria cell.

a. Find the bacteria cell. Extract the plasmid by cutting along the SOLID line.
b. To make this DNA sequence a true plasmid, form it into a ring and tape it together. Observe your plasmid. Note how it contains a gene for tetracycline resistance. This is a genetic marker.
c. Represent the cuts made by the restriction enzyme EcoRI by cutting along the DOTTED lines. (This will separate your plasmid ring.) Notice how the plasmid now has sticky ends.

Step #3: PRODUCE RECOMBINANT DNA.
In order to produce recombinant DNA, you must mix the plasmid with the donor gene. Since both molecules were cut with the same restriction enzyme, they will have the same sticky ends. The enzyme DNA ligase will "glue" the two molecules together. The plasmid will be placed back into its host cell, which in our activity, is a bacteria cell. Through transformation, the bacteria cell will pick up the new genetic material.

a. Insert the gene for human insulin into the plasmid. Make sure your base pairs match up. Use tape to represent the work of DNA ligase.

Step #4: CLONE CELLS.
The transformed bacteria cells are grown on agar, which is a medium on which bacteria are grown in a lab setting. The agar has tetracycline on it, an antibiotic used to kill bacterial infections.

Step #5: SCREEN CELLS.
Only the bacteria containing the plasmids with both the tetracycline resistant and desired genes will grow and survive on the agar. The bacteria without the tetracycline resistant gene will be killed by the antibiotic. This ensures that only the bacteria with the desired gene, in this activity the gene for human insulin, will survive. These cells can then be used to manufacture insulin to be sold to patients with diabetes.

A T T G C C T T A A G Gene for Tetracycline Resistance C T T C G C T T A G T T A C C G T

T A A C G G A A T T C Resistance G A A G C G A A T C A A T G G C A

Plasmid found in Bacterial Cell

A T G G C T T A A G Gene for Human Insulin G G C T T A A G

T A C C G A A T T C Insulin C C G A A T T C

Human Donor DNA

A T T G C C T T A A G Gene for Tetracycline Resistance C T T C G C T T A G T T A C C G T

T A A C G G A A T T C Resistance G A A G C G A A T C A A T G G C A

Plasmid found in Bacterial Cell

A T G G C T T A A G Gene for Human Insulin G G C T T A A G

T A C C G A A T T C Insulin C C G A A T T C

Human Donor DNA

A T T G C C T T A A G Gene for Tetracycline Resistance C T T C G C T T A G T T A C C G T

T A A C G G A A T T C Resistance G A A G C G A A T C A A T G G C A

Plasmid found in Bacterial Cell

A T G G C T T A A G Gene for Human Insulin G G C T T A A G

T A C C G A A T T C Insulin C C G A A T T C

Human Donor DNA

Analysis

1. What is the function of the restriction enzymes in this process?
2. How did we simulate the role of restriction enzymes in this activity?
3. What is the function of a vector in this process?
4. In this activity what is the vector?
5. What type of cell is the host cell in this activity?
6. What are "sticky ends"?
7. What is the function of the enzyme DNA ligase in this process?
8. How did we simulate the role of DNA ligase in this activity?
9. After the donor gene has been inserted into the plasmid vector, the resulting DNA is termed ______________________________ DNA.
10. After the plasmid vector has been taken up by the bacterial cell, the bacteria is termed a ____________________________ organism (aka "genetically modified organism" or GMO).
11. Explain why every cell that is produced from the genetically modified bacterium will be able to produce insulin.
12. <u>Explain</u> the role of the tetracycline resistance gene that is used as a genetic marker.
13. Several decades ago, diabetics did not have access to human insulin; they were injected with the insulin of sheep or other mammals. State two possible advantages of using genetic engineering in the production of human insulin for diabetics.
    a.
    b.