

Teacher Guide for Creating Transgenic Corn

For each pair of students:

One lab instruction sheet

yellow highlighter

blue highlighter

one pair of scissors

one pencil

scotch tape

White DNA strip

Pink DNA strip

Have extra DNA strips of each color to allow for mistakes. This lab requires the teacher to move from group to group approving each step of the process. This can be modified not to include these approval steps if the teacher wishes. It will depend on the teachers' assessment of the student's abilities to work unaided. The excel workbook has a teacher sheet that has each DNA step shown for easy comparison, along with a white DNA strip page for reproducing. The teacher will need to copy the corn DNA strips onto pink paper.

KEY: 1. Enzyme XbaI 2. DNA ligase 3. Answers will vary

Sources Used:

Primrose, S.B. and Twyman, R.M. Principles of Gene Manipulation and Genomics, 7th Edition. Balckwell Publishing, 2006.

http://parts.mit.edu/igem07/index.php/Wet_to_Dry (used slide 16 from Powerpoint for information)

Name _____ Name _____

Creating Transgenic Corn

In this activity you will recreate the steps in creating transgenic corn. In our discussions, we have learned that transgenic means having the genetic material from one species incorporated into another.

Agrobacterium sp. strain CP4, has a naturally occurring gene encoding a glyphosate-tolerant enzyme, CP4 EPSP synthase. In this activity we will remove this gene from the DNA of the *Agrobacterium* and incorporate it into the DNA of corn. Our purpose is to make the corn have the same glyphosate tolerance. Glyphosate is the main ingredient in a weed killer made by Monsanto which is called Roundup.

Why do you think corn with this tolerance would be desirable?

In our first step we must be able to remove the gene from *Agrobacterium*. Our simulation will not have the exact DNA sequence as the real *Agrobacterium*, but a much smaller version. We will be using a restriction enzyme called XbaI which recognizes the GAATTC sequence. Restriction enzymes cut DNA at specific sites leaving “sticky ends”.

On the white strip of DNA (our *Agrobacterium* DNA), using a yellow highlighter, highlight the GAATTC sequences on the 5' to 3' strand, and the same sequence that runs CTTAAG on the 3' to 5' strand. You should find two of them. When you have completed this, call your teacher over to initial here _____.

Now take a blue highlighter and highlight in between the yellow highlighted area. This is the gene that represents the glyphosate tolerance gene. With a pencil, draw lines that cut the GAATTC between the G and A on each sequence. It should look like the sequence below.



Your scissors represent the enzyme XbaI. Cut out the section in between the pencil marks. Now you have the gene that codes for the glyphosate tolerance.

Now we must somehow take this gene and put it into our corn plant. In the actual process, particle bombardment is used. Particle bombardment is when tiny microscopic metal pieces (1-4µm) are coated with the DNA pieces you want to incorporate, and are placed in a modified shotgun and fired at the plant cell at a velocity of approximately 250 m/s. This velocity allows the particles to penetrate the cell wall.

Our corn DNA is represented by the pink strip of DNA. We must take our XbaI enzyme and locate the GAATTC sequence, and cut it in the same way we did earlier to our *Agrobacterium* DNA, except this time we should only find one site.

On the pink strip of DNA locate the site, and with a pencil draw the cut marks you believe you should make. Call your teacher over for initials here _____. If approved, cut the corn DNA apart.

Now we need to join our bacterial DNA to the corn DNA. This is done with a ligase enzyme which we will call X-ligase. Our X-ligase will be scotch tape. Carefully match up the “sticky ends” of the corn DNA and the *Agrobacterium* DNA and tape them together. Call your teacher over to approve here _____. We have created transgenic corn DNA.

Questions:

1. In our activity, what did the scissors represent?

2. In our activity, what did the scotch tape represent?

3. Although this was a very simplistic exercise in how transgenic organisms are created, we can see the possibilities of other organisms being modified. With your partner, if you could create an organism with a trait from another species, what would you try to create? And why?

Staple your transgenic corn DNA to this sheet and turn in.

Agr 5' to 3' A T T G C T G C G A A T T C C A C C C G T A A G A A T T C G C
DNA 3' to 5' T A A C G A C G C T T A A G G T G G G C A T T C T T A A G C G

Agr 5' to 3' A T T G C T G C G A A T T C C A C C C G T A A G A A T T C G C
DNA 3' to 5' T A A C G A C G C T T A A G G T G G G C A T T C T T A A G C G

Agr 5' to 3' A T T G C T G C G A A T T C C A C C C G T A A G A A T T C G C
DNA 3' to 5' T A A C G A C G C T T A A G G T G G G C A T T C T T A A G C G

corn 5' to 3' A T T G C T G C G A A T T C G A A T T C G C
DNA 3' to 5' T A A C G A C G C T T A A G C T T A A G C G

corn 5' to 3' A T T G C T G C G A A T T C G A A T T C G C
DNA 3' to 5' T A A C G A C G C T T A A G C T T A A G C G

corn 5' to 3' A T T G C T G C G A A T T C C A C C C G T A A G A A T T C G C
DNA 3' to 5' T A A C G A C G C T T A A G G T G G G C A T T C T T A A G C G

Copy this onto white paper

Agr 5' to 3' A T T G C T G C G A A T T C C A C C C G T A A G A A T T C G C
DNA 3' to 5' T A A C G A C G C T T A A G G T G G G C A T T C T T A A G C G

Agr 5' to 3' A T T G C T G C G A A T T C C A C C C G T A A G A A T T C G C
DNA 3' to 5' T A A C G A C G C T T A A G G T G G G C A T T C T T A A G C G

Agr 5' to 3' A T T G C T G C G A A T T C C A C C C G T A A G A A T T C G C
DNA 3' to 5' T A A C G A C G C T T A A G G T G G G C A T T C T T A A G C G

Agr 5' to 3' A T T G C T G C G A A T T C C A C C C G T A A G A A T T C G C
DNA 3' to 5' T A A C G A C G C T T A A G G T G G G C A T T C T T A A G C G

Agr 5' to 3' A T T G C T G C G A A T T C C A C C C G T A A G A A T T C G C
DNA 3' to 5' T A A C G A C G C T T A A G G T G G G C A T T C T T A A G C G

Agr 5' to 3' A T T G C T G C G A A T T C C A C C C G T A A G A A T T C G C
DNA 3' to 5' T A A C G A C G C T T A A G G T G G G C A T T C T T A A G C G

Agr 5' to 3' A T T G C T G C G A A T T C C A C C C G T A A G A A T T C G C
DNA 3' to 5' T A A C G A C G C T T A A G G T G G G C A T T C T T A A G C G

Copy this onto pink paper

corn 5' to 3'

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

DNA 3' to 5'

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

corn 5' to 3'

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

DNA 3' to 5'

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

corn 5' to 3'

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

DNA 3' to 5'

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

corn 5' to 3'

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

DNA 3' to 5'

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

corn 5' to 3'

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

DNA 3' to 5'

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

corn 5' to 3'

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

DNA 3' to 5'

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

corn 5' to 3'

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

DNA 3' to 5'

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