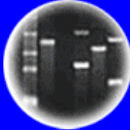


DNA  
Genetic Code of Life



Entire Genetic Code  
of a Bacteria



DNA Fingerprinting



Cloning: Ethical Issues  
and Future Consequences



Plants of Tomorrow

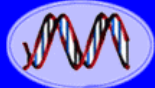
# HC70A & SAS70A Spring 2017 Genetic Engineering in Medicine, Agriculture, and Law

**Professors Bob Goldberg  
& John Harada**

## Lecture 2 The Age of DNA: What Is Genetic Engineering-Part Two

**UCLA**

**UC DAVIS**  
UNIVERSITY OF CALIFORNIA



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Entire Genetic Code  
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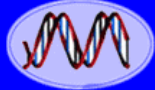
Cloning: Ethical Issues  
and Future Consequences



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## **THEMES**

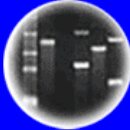
1. The Scientific Process
2. The Significance of Genetic Engineering
3. What Are the Tools of Genetic Engineering?
4. What Can Be Done With Genetic Engineering-  
Some Examples
5. What Does Genetic Engineering Tell Us About  
Basic Genetic Processes in All Organisms?
6. Genetic Engineering - Anything New?
7. Are Vegetables Engineered - Demonstration
8. Science & Ideology - A Tragic Combination
9. Classical vs. 21st Century Genetic Engineering
10. Understanding How Genetic Engineering Uses  
Natural Rules of the Cell (i.e., It Isn't  
Magic)!



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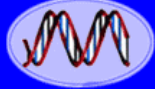


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# WHAT IS SCIENCE?

**WEBSTER:** Knowledge about, or study of, the natural world based on facts learned through experiments and observation.

Technology, like Genetic Engineering, is the application of science knowledge



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# HOW IS SCIENCE CARRIED OUT?



Scientific Knowledge is Based on Observation, Hypothesis Testing, Rigorous Experimentation, Results, Facts, and Verification

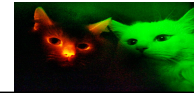
*What Are the Data?  
What Is the Evidence?*

Science is **NOT** "Hocus Pocus" or Based on Opinions and Beliefs

# What Is the Significance of Genetic Engineering?

1. Specific DNA Sequences and Genes Can Be Isolated From Any Organism
2. DNA Segments of Any Kind From Any Organism Can Be Combined (Genetic Engineering!!!!!!)
3. Isolated Genes Can Be Engineered and Re-Inserted Into the Chromosomes of Any Organism and Made to Work
4. Genes and Genomes Can Be Synthesized, Edited, and Made To Work in Any Organism


*There Are No Genetic Limits. All Biological Organisms Use the Same Genetic Rules. The Implications Are Enormous!!*



  
DNA  
Genetic Code of Life

  
Entire Genetic Code  
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DNA Fingerprinting

  
Cloning: Ethical Issues  
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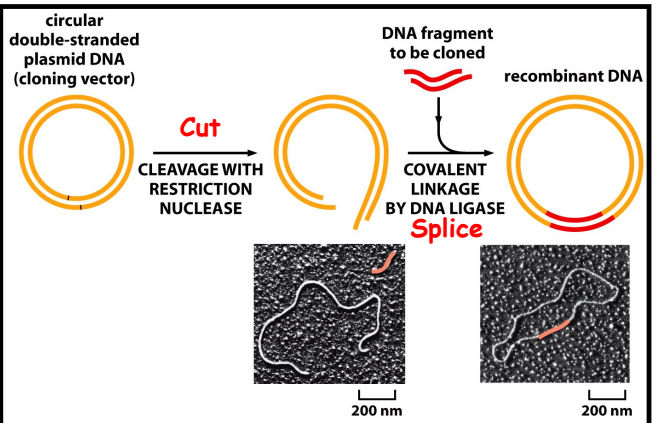


## What "Tools" Do You Need For Genetic Engineering?



# What "Tools" Are Needed For Genetic Engineering?

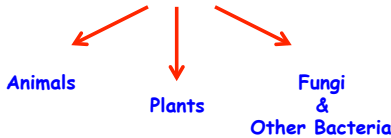




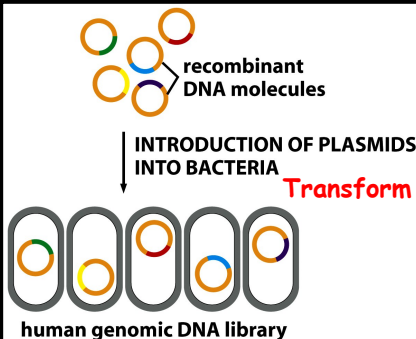


The diagram shows a circular double-stranded plasmid DNA (cloning vector) being cut with a restriction nuclease. A DNA fragment to be cloned is then inserted, and the DNA is resealed by DNA ligase, creating recombinant DNA. Two electron micrographs below show the plasmid and recombinant DNA at 200 nm scale.

1. DNA
2. Vector (Plasmids)
3. Enzymes
  1. Restriction Enzymes (Cut)
  2. Ligase Enzyme (Splice)
4. Cells (*E. coli*)

Lab Engineered DNA



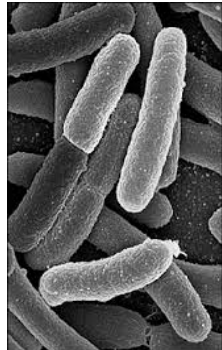


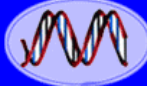
recombinant DNA molecules

INTRODUCTION OF PLASMIDS INTO BACTERIA


**Transform**

human genomic DNA library

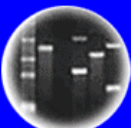





DNA  
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## What Can Be Done With Genetic Engineering?

### Some Examples

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Plants of Tomorrow

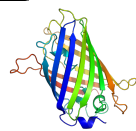
## Using a Jellyfish Gene to Make Bacteria, Animals, and Plants Glow!!!!



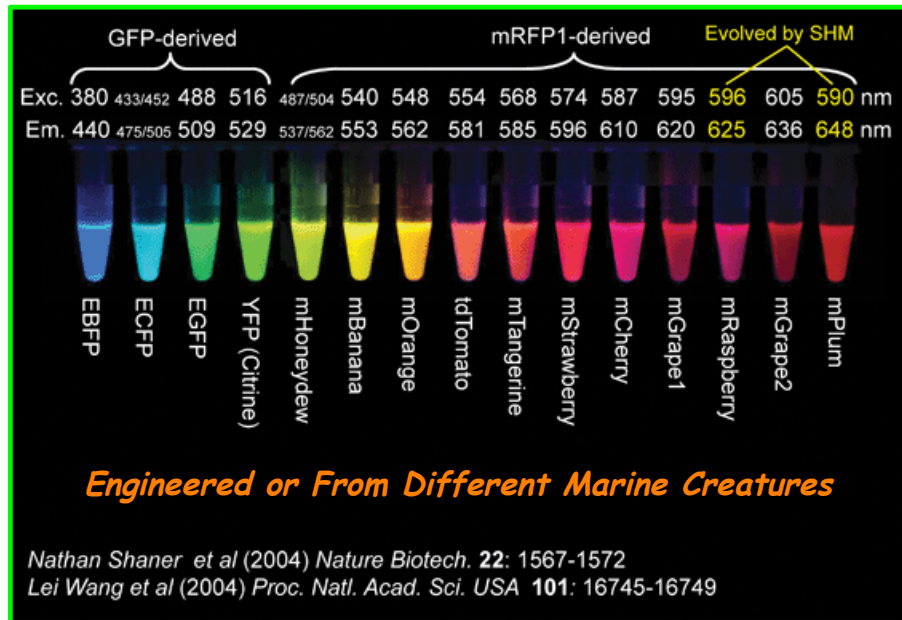
### Green Fluorescence Protein (GFP)

(238 amino acids)

Nobel Prize in Chemistry - 2008 - Shimomura, Chalfie, & Tsien



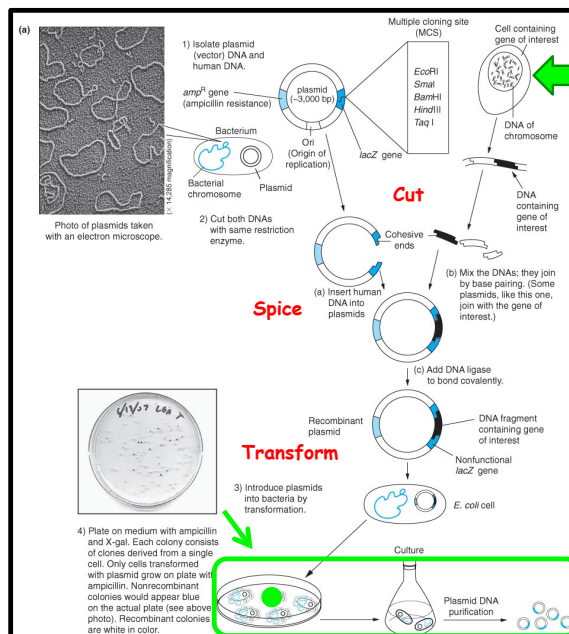
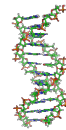
# There Are Many Different Kinds of Fluorescing Proteins!



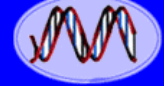
Nobel Prize in Chemistry - 2008 - Shimomura, Chalfie, & Tsien

-  DNA Genetic Code of Life
-  Entire Genetic Code of a Bacteria
-  DNA Fingerprinting
-  Cloning: Ethical Issues and Future Consequences
-  Plants of Tomorrow


## Using Recombinant DNA to Isolate the Jellyfish GFP Gene



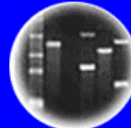
How Find the E. coli colony With the GFP Gene?




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DNA Fingerprinting



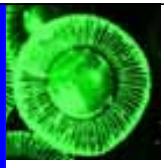
Cloning: Ethical Issues  
and Future Consequences

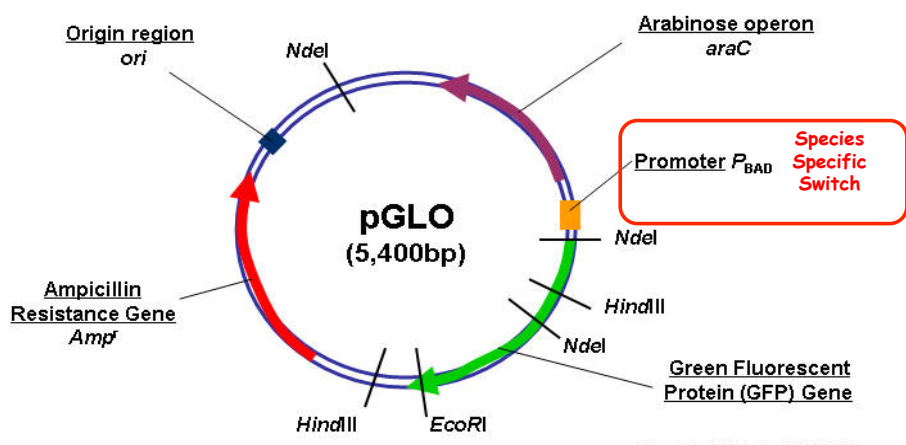


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## A Recombinant Plasmid Containing the GFP Gene


How Make it Active in Living Cells?





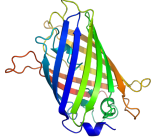
**pGLO (5,400bp)**

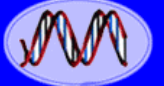
Graphic©E.Schmid/2003




Bacterial Switch

Need a **Species-Specific Switch** to  
Allow a Gene To Function in a  
Specific Organism

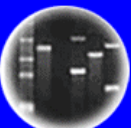





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DNA Fingerprinting




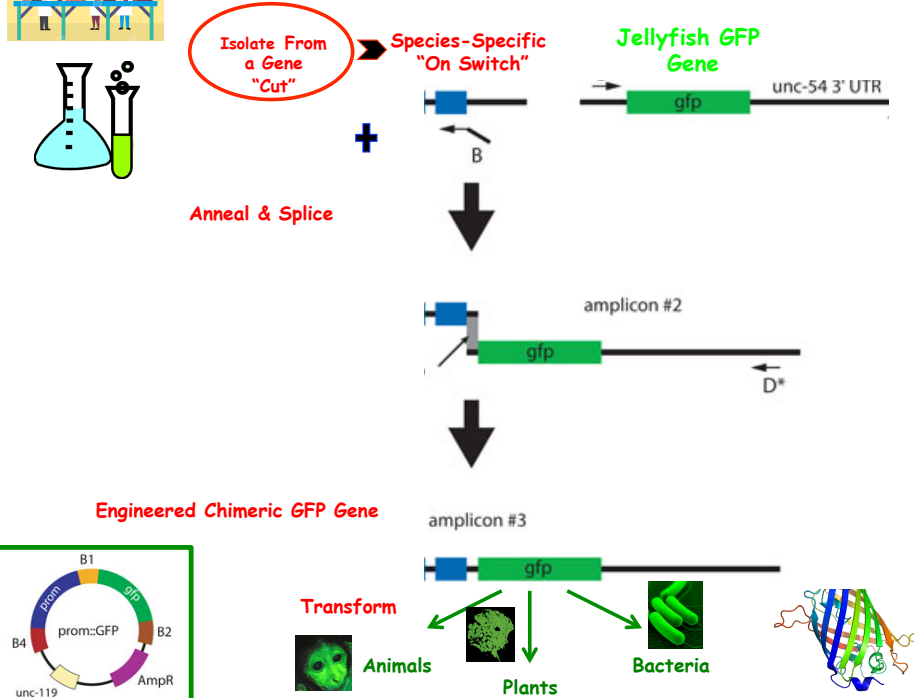
Cloning: Ethical Issues  
and Future Consequences




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## Engineering the Jellyfish GFP Gene to Be Active in Different Organisms







Engineered Chimeric GFP Gene



Animals



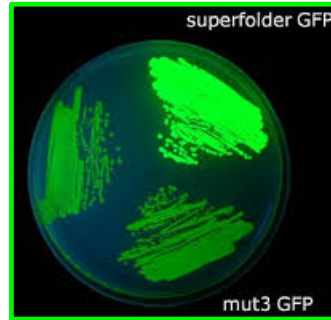
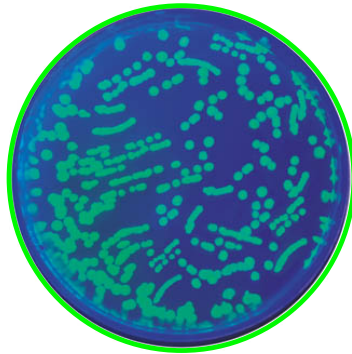
Plants



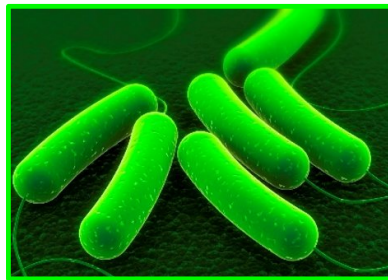
Bacteria

# GloColi - Engineering *E. coli* With the Jellyfish GFP Gene!

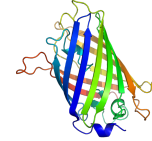
What Are the Conceptual Implications of This Experiment?



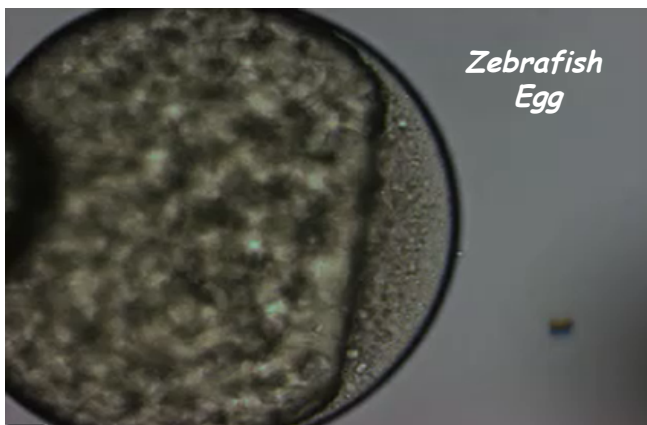
*E. coli* Switch + Jellyfish GFP Gene



*E. coli* Synthesizes GFP Protein!

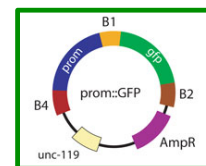


## Engineering a "GloFish"



Zebrafish Egg

Zebrafish *Danio rerio*

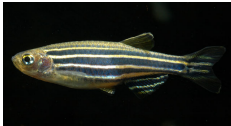
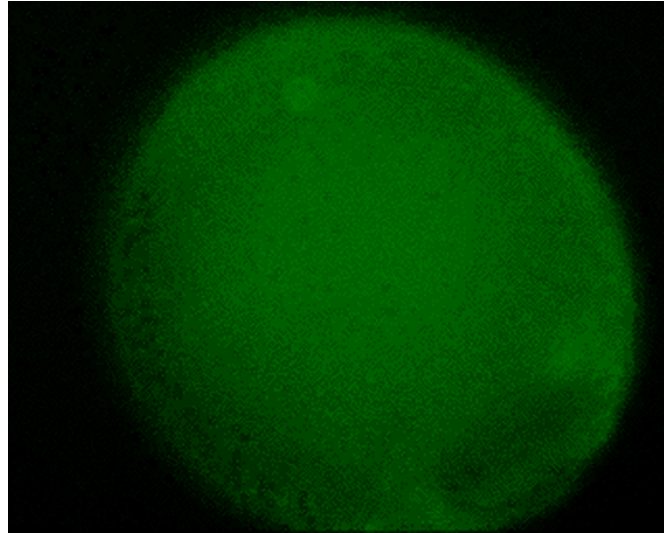


Using Genetic Engineering To Insert An Engineered Jellyfish GFP Gene into a Zebrafish Egg!

What Switch Used?



## A “GloFish” Embryo!!



*Zebrafish - Danio rerio*

## *Genetically Engineered “GloFish!!”*



Note Different Fluorescing Colors - Due to Different Engineered Jellyfish Genes



DNA  
Genetic Code of Life



Entire Genetic Code  
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DNA Fingerprinting



Cloning: Ethical Issues  
and Future Consequences



Plants of Tomorrow

## Can GloFish Can Be Sold In California?

- **Cal. Depart. of Fish and Game Code § 15007 (2007) Regulation** Makes it illegal to spawn, cultivate, or incubate any transgenic fish in the state controlled waters of the Pacific Ocean.
- **Cal. Depart. of Fish and Game Code Ruling (2015)** The Dept. of Fish and Game will propose the addition of an exception to Section 1.92 that would allow the sale of transgenic tropical aquarium fish that the Dept. has determined pose no foreseeable risk or harm to native fish or wildlife.

### Genetic Engineering & The Law!



## How About a GloFly!



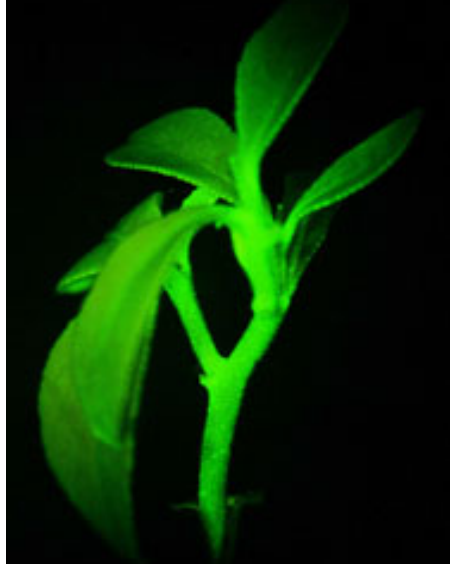
## *What About "GloMice!!!"*



## *And Glo Monkeys, Cats and Pigs as Well!!*



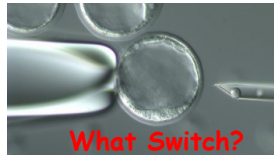
## *Engineering a GloPlant With the Same Jellyfish Gene!!!*



*What are the Biological Implications of These Experiments?*

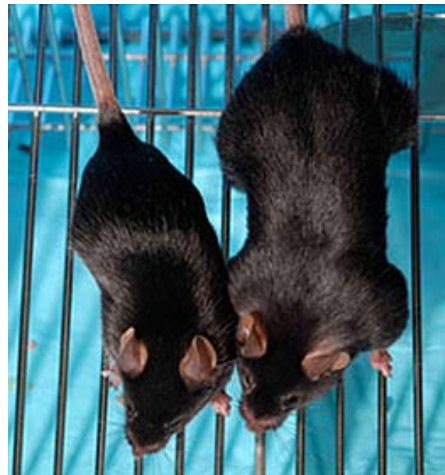
## **Dramatic growth of mice that develop from eggs microinjected with metallothionein–growth hormone fusion genes**

**Richard D. Palmiter<sup>1</sup>, Ralph L. Brinster<sup>1</sup>, Robert E. Hammer<sup>1</sup>, Myrna E. Trumbauer<sup>1</sup>, Michael G. Rosenfeld<sup>2</sup>, Neal C. Birnberg<sup>3</sup> & Ronald M. Evans<sup>1</sup>**



Nature,  
December, 1982  
~33 Years Ago!

**Engineering "Mighty Mouse" With a Rat Growth Hormone Gene**



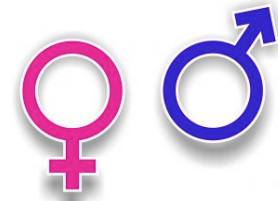
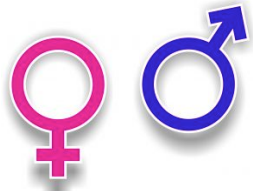
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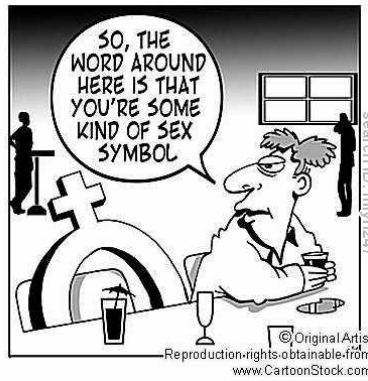
 DNA Fingerprinting

 Cloning: Ethical Issues  
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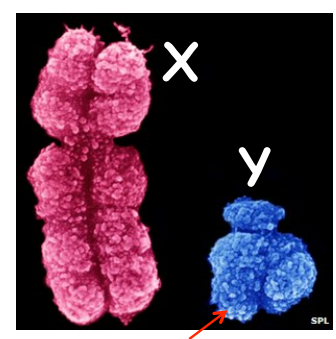
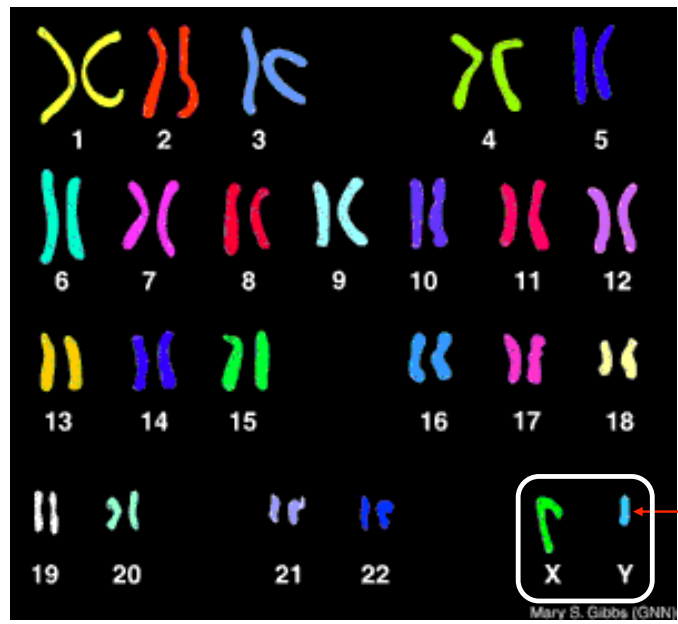
 Plants of Tomorrow



# How About Engineering The Sex Of An Organism?



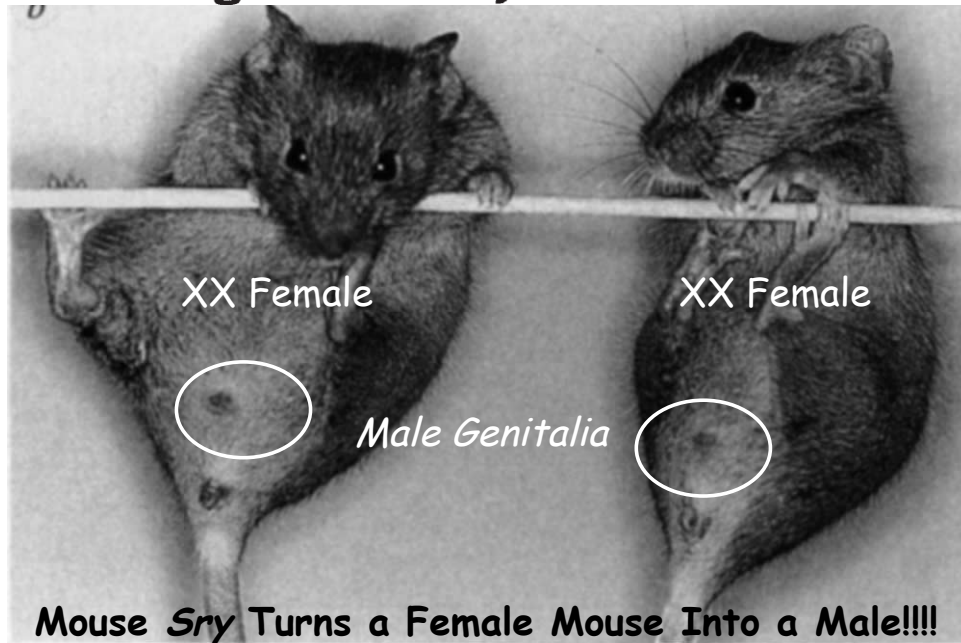
Males and Females Differ By the Presence or Absence Of the Y Chromosome (simplistically!!)



Male SRY Gene  
(Sex Determining  
Region Y)


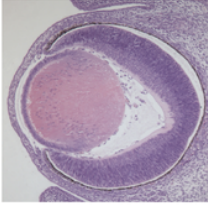
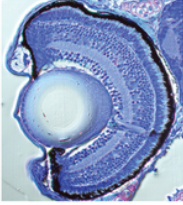

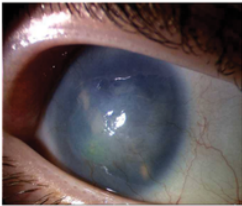
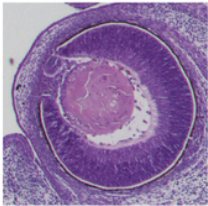
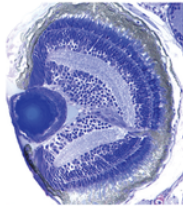
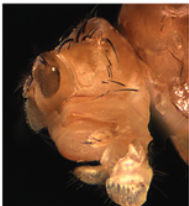
**The Human SRY Gene For Maleness Controls Gender**

# Male development of chromosomally female mice transgenic for *Sry* Nature, May 9, 1991

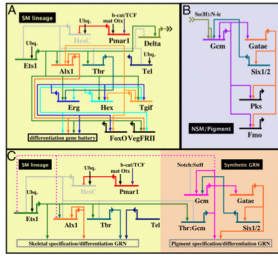


*Functional Proof That Sry Controls Male Development*

## Using Genetic Engineering to Change Body Architecture-Engineering Eyes on a Fly's Leg With a Single Gene!

	Human	Mouse	Zebrafish	<i>Drosophila</i>
WT				
mut				
	<b><i>PAX6</i><sup>-/-</sup></b>	<b><i>Pax6</i><sup>-/-</sup></b>	<b><i>pax6b</i><sup>-/-</sup></b>	<b><i>ey</i><sup>-/-</sup></b>
EQs	cornea opaque <u>iris absent</u> retina degenerate lens opaque aqueous humor of eyeball increased pressure	eye decreased size lens fused to cornea iris morphology anterior chamber absent	eye decreased size lens decreased size retina malformed	eye absent

# Induction of Ectopic Eyes by Targeted Expression of the *eyeless* Gene in *Drosophila* *Science* 267, 1788, 1995



Mouse PAX-6 Gene (*eyeless* in flies)  
Engineered To Work

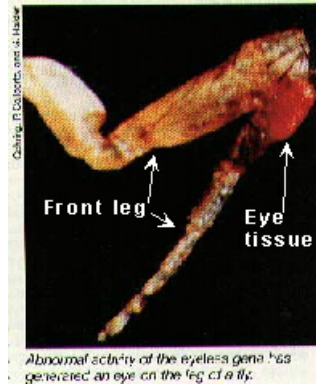


Different Fly Regions

Fly Leg Switch + Mouse Eye Gene



What Does This Experiment Tell Us About Mouse & Fly Eye Genes?



DNA Genetic Code of Life

Entire Genetic Code of a Bacteria

DNA Fingerprinting

Cloning: Ethical Issues and Future Consequences

Plants of Tomorrow

## What About Inserting Bacterial Genes Into Plants To Produce a Result With Significant Agricultural Application?

GARDEN GUIDE SUNSET

WHAT TO DO IN YOUR GARDEN IN SEPTEMBER

### Southern California Checklist

PROTECT CABBAGE CROPS. The minute you plant a brassica, squadrons of cabbage white butterflies seem to descend on it to lay their eggs. The easiest way to thwart them is to cover your cabbage crops with row covers right from the start. The next best option is spraying with *Bacillus thuringiensis* to kill the young caterpillar larvae. ♦

  
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 Plants of Tomorrow

## How to Use Bt Pesticide as an Organic Pest Control


Learn how to use Bt pesticide to kill cabbage worms, tomato hornworms and other pests in your organic vegetable garden.


By Barbara Pleasant  
 April 24, 2013



Bt is one of the safest natural pesticides you can use to control caterpillar pests without harming beneficial insects.

Photo Courtesy Safe Brand

 **OMRI**  
 LISTED

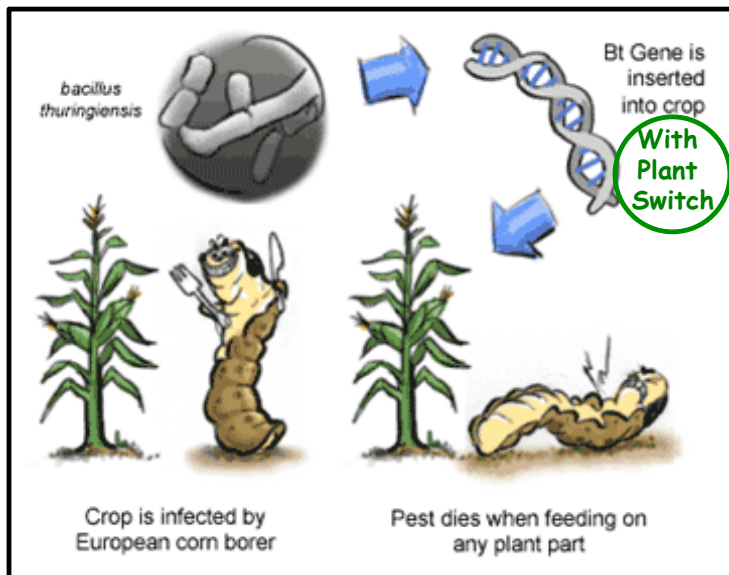
 **Monterey**

FOR ORGANIC GARDENING

Active Ingredient:  
*Bacillus thuringiensis* subspecies *kurstaki* strain SA-12 solids,  
 spores and Lepidopteran active toxins (At least 6 million  
 viable spores per mg) . . . . . 98.35%  
 Other ingredients: . . . . . 1.65%  
 Total: . . . . . 100.00%

\*The percent active ingredient does not indicate product performance and potency measurements are not federally standardized.

## Crops Can Be Engineered With Bt For Insect Resistance



*Bacillus thuringiensis*



**Bt Toxin in Spores**



Spore Crystal

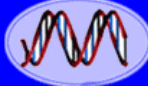
*Hornworm*






## Genetic Engineering a Plant to Resist Worms! Implications For Agriculture

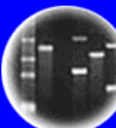





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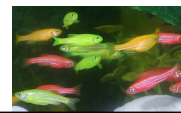
Plants of Tomorrow

### What Can We Infer FROM These Genetic Engineering Experiments About How Genes “Work” and Genetic Processes in All Living Organisms?

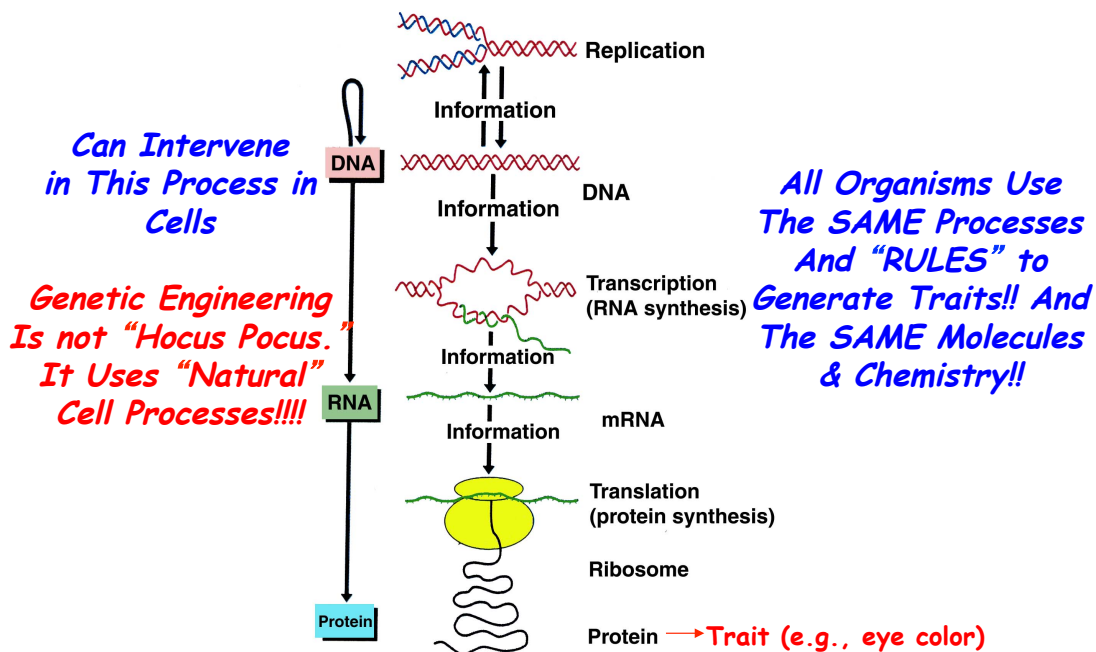
The diagram illustrates the central dogma of molecular biology within a cell. It shows DNA (labeled 'Genes') being transcribed into mRNA (RNA synthesis). This mRNA is then translated into a polypeptide chain (protein synthesis) by a ribosome, with tRNAs acting as adaptors. The final product is a polypeptide, which is linked to the concept of 'Traits!'.

# Observations and Inferences From Genetic Engineering Experiments

1. **Genes Can Work Independently of Each Other**  
*E.G. - The Jellyfish Fluorescence Gene Works Perfectly in a Variety of Organisms*
2. **Basic Genetic Processes Are Universal (Replication & DNA to RNA to Protein)**  
*E. G. - The Bt Gene Directs the Production of BT Protein in Crops.*
3. **Basic Genetic Processes Can Be Used to Engineer or Transfer Genes From One Organism to Another and Transfer Them Stably Generation After Generation**  
*E.G. - The Chimeric GloFish & Bt Genes Are Inherited Generation After Generation.*



## Translating The Genetic Code Into Proteins is a Conserved Process





DNA  
Genetic Code of Life



Entire Genetic Code  
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Cloning: Ethical Issues  
and Future Consequences



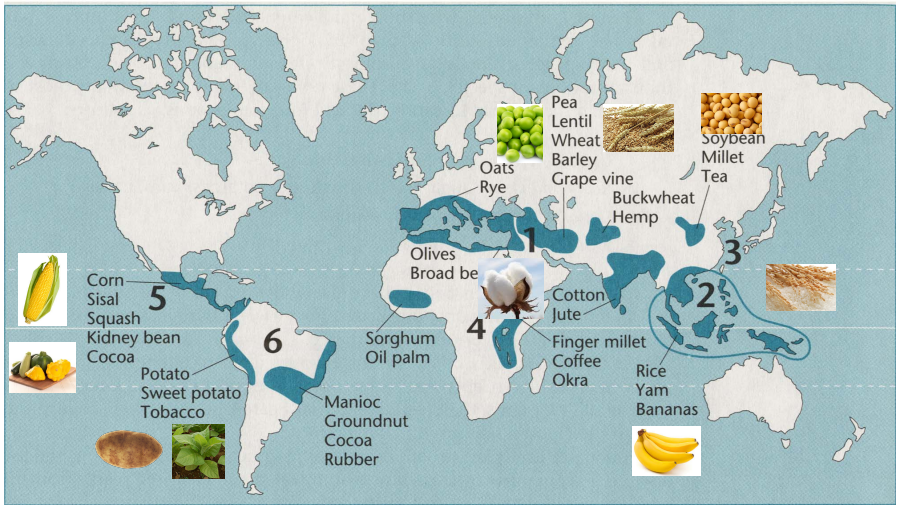
Plants of Tomorrow

# There is Nothing New About Genetic Engineering!

Manipulating Genes IS  
 Manipulating Genes No Matter  
 What Technology or Techniques  
 Are Used!!

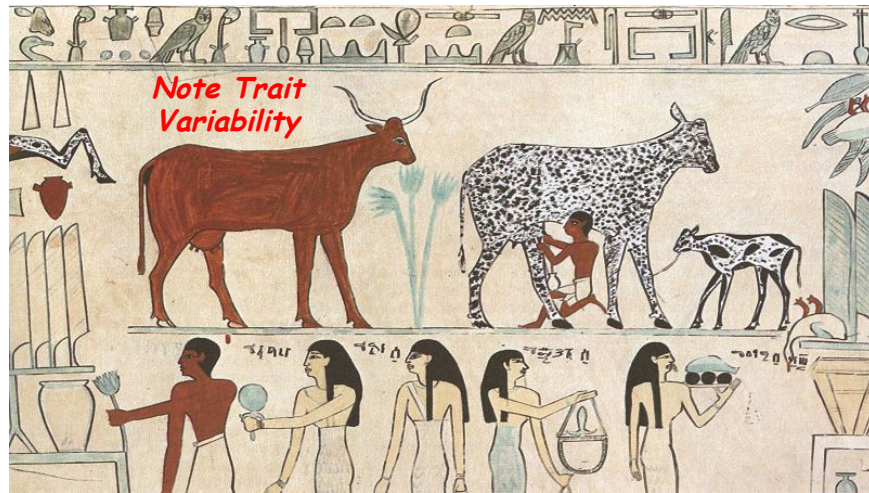
*All Major Crops Were Engineered From Wild Relatives  
 by Early "Bioengineers" Over 10,000 Years Ago!!*

**Regions Where Major Crops Were Established**



*Breeding Involves Gene Manipulation Using EXISTING Genetic Variability!*

**Farm Animals Were Also “Engineered” By  
Breeding Wild Relatives  
Cattle Breeding in Egypt 4,000 Years Ago!**



**Manipulating Existing Genetic Variability  
Brought About By Chance Mutations!**

**Breeding Uses Natural Genetic Variability of Genes  
As Raw Material - Variability Generated by Mutations**



**Mutations in a Gene That Change Its Chemical Sequence  
& Slightly Alters Its Function (e.g., fruit size, color)**

**DNA**  
Genetic Code of Life

Entire Genetic Code of a Bacteria

DNA Fingerprinting

Cloning: Ethical Issues and Future Consequences

Plants of Tomorrow

**Populations of All Organisms Contain Genetic Variability**

***Tomatoes Were Engineered From Small Wild Relatives Because of Mutations in Fruit Size Genes!***



***The Early Tomato "Bioengineers" Selected For Large Fruit Size Because it Provided More Food!***

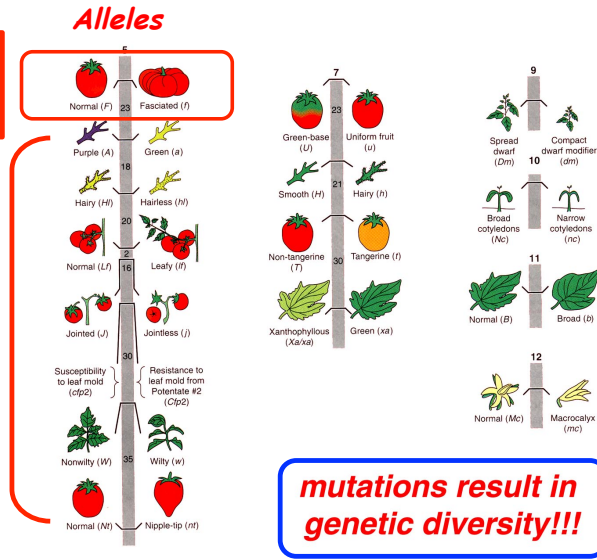
***What They Were Selecting Was a Different Form (Allele) of a Fruit Size Gene.***

*al·lele e'lēl/ Noun GENETICS plural noun: alleles  
one of two or more alternative forms of a gene that arise by mutation and are found at the same place on a chromosome.*

## Alleles Reside at the Same Position on a Chromosome

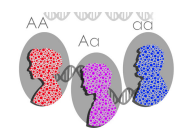
Allele Phenotypes Specify Markers For Each Gene Location!

Different Genes

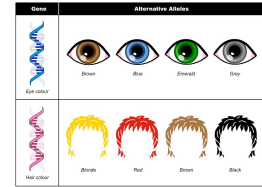


Gene Engineering Can Generate New Forms of Alleles of a Gene and, therefore, Results in More Genetic Diversity

mutations result in genetic diversity!!!

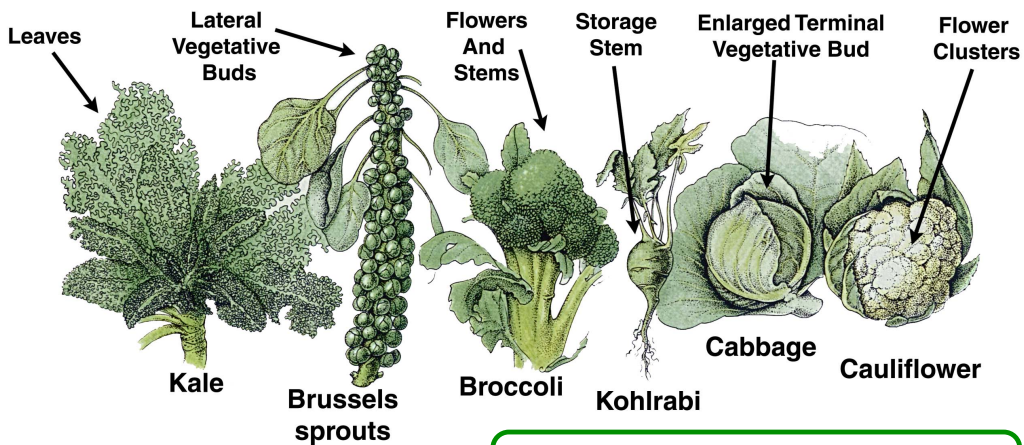


Alleles Are Different Forms of the Same Gene That Arise By Mutation & Can be Made in a Laboratory By Modern Genetic Engineering!



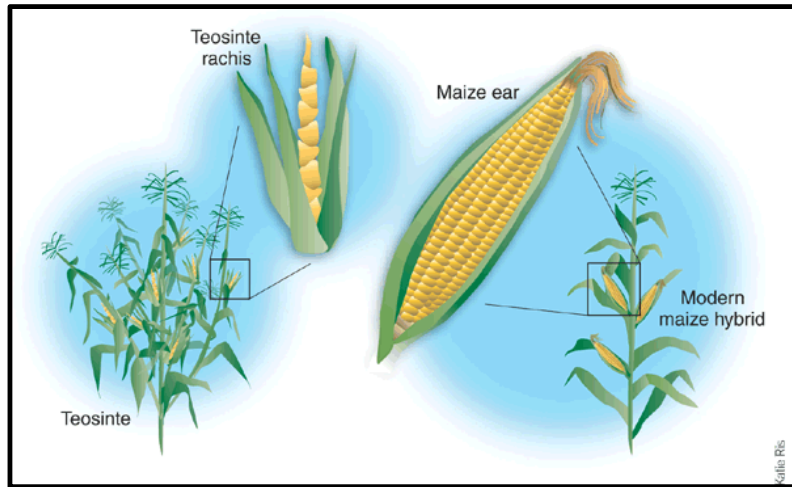
## Engineering Brassica Vegetables From a Wild Mustard

Breeders Have Selected For Variability In Plant Control Genes To Generate Novel Crops



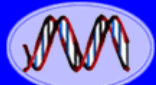
How Are These Plants Related?

## Engineering Corn From the Wild Grass Teosinte



**Note:** *Architecture and Fruit (cob) Size*

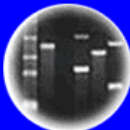
*Only Five Genes Cause These Plants to Differ  
& We Now Know What They Are*



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Genetic Code of Life



Entire Genetic Code  
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DNA Fingerprinting



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Plants of Tomorrow

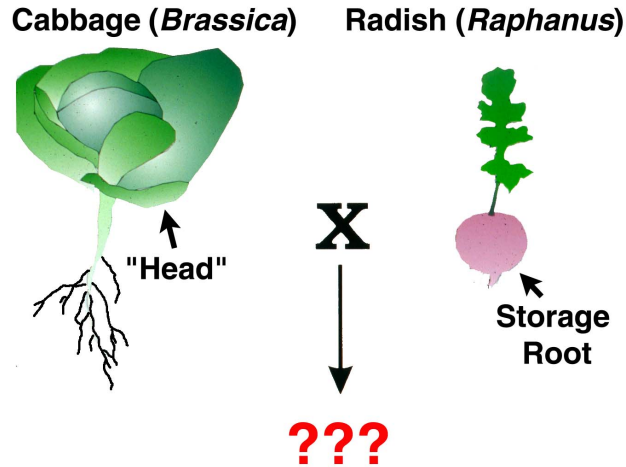
## The Problem With Breeding the "Old Fashioned Way"

**Cannot Predict Results!**



# The Problem With Breeding the "Old Fashioned Way"

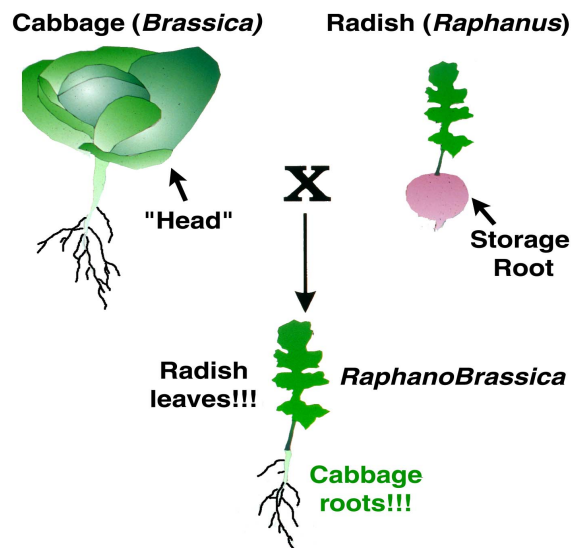
## Engineering A Novel Crop By "Wide" Breeding



Karpechenko, G.D., 1928. *Polyploid hybrids of Raphanus sativus L. X Brassica oleracea L.*  
*Zeitschrift für induktive Abstammungs- und Vererbungslehre* 48, 1-85.



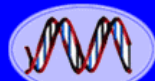
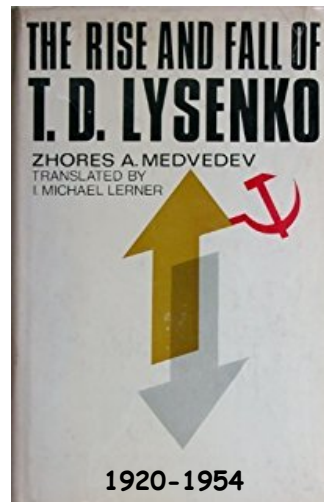
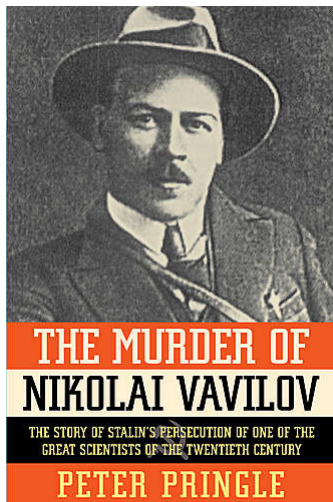
## Engineering A Novel Crop By "Wide" Breeding



*Results Show the Unpredictability of Classical Breeding Approaches!!  
Compare With the Modern Genetic Engineering Examples Shown Previously*



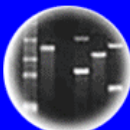
## How Ideology Destroys Science & Leads to Horrific Tragedy



DNA  
Genetic Code of Life



Entire Genetic Code  
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DNA Fingerprinting



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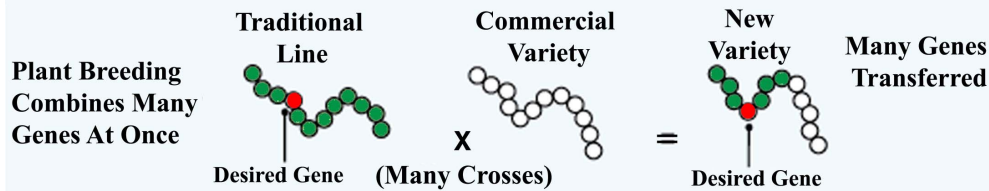
Plants of Tomorrow

**Genetic Engineering is a TECHNIQUE!**

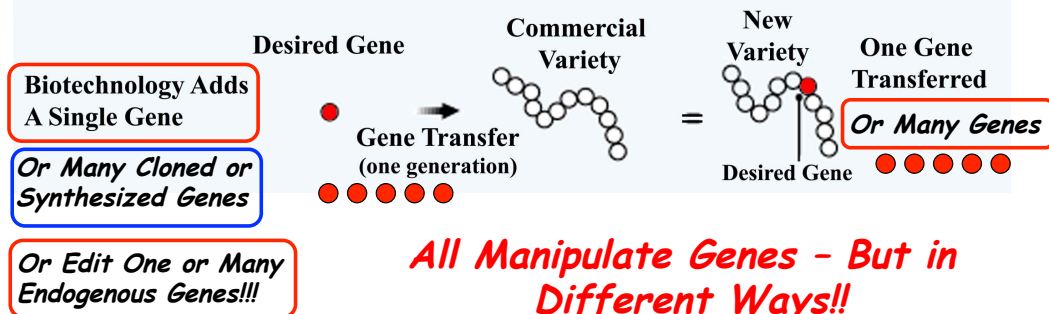
*How Do Classical Genetic Engineering  
Methods Differ From Those Using  
DNA and 21st Century Technologies?*

# Classical vs. DNA or Molecular Genetic Engineering Techniques

## TRADITIONAL PLANT BREEDING



## PLANT BIOTECHNOLOGY

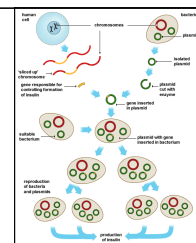
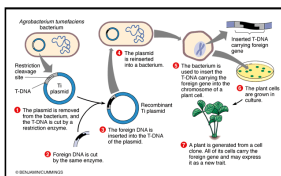


## What Are The Limitations of Classical Breeding/Genetic Engineering?

1. Limited To Genes of Interbreeding Organisms and, Clearly, Severe Ethical Issues With Humans (Eugenics)
2. Only Can Make New Combinations of EXISTING Genes - Genes Created By "Natural" Mutations
3. Can't Make Existing Genes "Better" - Just Better or More Useful Combinations of Existing Genes and/or Alleles
4. Takes Time - Limited To Generation Time of Organism - Decades For Some Crop Plants
5. Only Useful For "Obvious" Traits - One's That Can Be Observed or Followed
6. Unpredictable Outcomes (Bringing in Thousands of Genes at Once - Some With Deleterious Consequences)

# What Are The Advantages of Using 21st Century Genetic Engineering Methods?

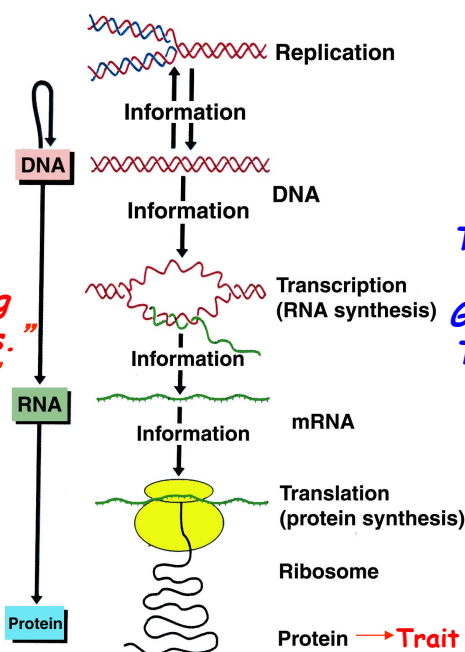
1. Any Gene From Any Organism Can Be Used In Any Organism - There Are No Breeding Barriers (e.g., genes of all sequenced genomes)
2. New Genes Can Be Engineered - Genes That Work Better and/or Produce New Proteins (i.e., create new genetic variability and/or alleles)
3. Existing Genes Can Be Engineered to be Switched On in "Places" That They Are Normally Off - Gene Control or Regulation Altered (e.g., fly eye on leg)
4. Speed - Can Engineer a New Organism in a Generation
5. Can Change, Alter, Manipulate, Synthesize and/or Control the Genetic Blueprint of Any Organism
6. Very Precise (Working With Known Genes & Proteins)



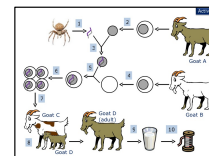
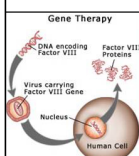
## THE FACTS ARE:

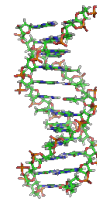
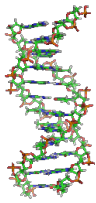
*Can Intervene in This Process in Cells*

**Genetic Engineering Is Not "Hocus Pocus." It Uses "Natural" Cell Processes!!!!**



*All Organisms Use The SAME Processes And "RULES" to Generate Traits!! And The SAME Molecules & Chemistry!!*





## **We Live in The Age of DNA & Genetic Engineering!**

***Understanding Genetic Engineering  
Requires a Basic Understanding of Genes  
And How They Work***

