

This week in 206

From 1 week ago:

Any live chickens?

Fungi reproductive structures.

- I. Escherichia coli plasmid transformation and gene expression.
antibiotic selection, bioluminescence, melanins
- II. Arabidopsis thaliana molecular genetic analysis.
mutants, transgenics, gene expression

Model genetic organisms

Used to study processes in development, physiology, behavior, reproduction, disease, gene regulation, signal transduction, etc. etc.

Traits of models.

EASY + CHEAP

Limitations on space, time, funds.

Model genetic organisms

Used to study processes in development, physiology, behavior, reproduction, disease, gene regulation, signal transduction, etc. etc.

Traits of models.

EASY + CHEAP

Small

Lots of progeny

Fast generation

Inexpensive culture

Simple

Good genetics (diploid or haploid, mutagenesis, crossing)

Relevant-normal or does something interesting or useful

No “societal” problems

Small genome-(sequenced)

Transformable

Examples of model genetic organisms.

Examples of model genetic organisms.

Escherichia coli (and lambda)

Saccharomyces -Baker's Yeast

Drosophila -Fruit fly

Mus -Mouse

Danio -Zebra fish

Xenopus -African Clawed frog

Caenorhabditis -Nematode

Dictyostelium -Slime mold

Arabidopsis

Genetics has had a huge resurgence in the past several years.

Biochemistry and molecular biology can't answer all the questions. Functional genomics.

Examples of model genetic organisms.

Escherichia coli (and lambda)

Arabidopsis thaliana

Very small

lots of progeny

short generation (30 days)

Smallest genome (125 million bases)

Easy to grow under many conditions

Easy to mutate and do genetics

Easy to transform.

Has roots, leaves, stems, flowers, fruits, seeds, etc.

Organism	estimated size	estimated gene number	average gene density	chromosome #
<i>Homo sapiens</i> (human)	2900 million bases	~30,000	1 gene per 100,000 bases	46
<i>Rattus norvegicus</i> (rat)	2,750 million bases	~30,000	1 gene per 100,000 bases	42
<i>Mus musculus</i> (mouse)	2500 million bases	~30,000	1 gene per 100,000 bases	40
<i>Drosophila melanogaster</i> (fruit fly)	180 million bases	13,600	1 gene per 9,000 bases	8
<i>Arabidopsis thaliana</i> (plant)	125 million bases	25,500	1 gene per 4000 bases	5
<i>Zea mays</i> (corn)	5000 million bases	~25,000	1 gene per 200,000 bases	10
<i>Oryza sativa</i> (rice)	565	~25,000	1 gene per 23000 bases	12
<i>Caenorhabditis elegans</i> (roundworm)	97 million bases	19,100	1 gene per 5000 bases	6
<i>Saccharomyces cerevisiae</i> (yeast)	12 million bases	6300	1 gene per 2000 bases	16
<i>Escherichia coli</i> (bacteria)	4.7 million bases	3200	1 gene per 1400 bases	1
<i>H. influenzae</i> (bacteria)	1.8 million bases	1700	1 gene per 1000 bases	1



Macdonald



Sisson



Stein



Iyer



Chan



Appling



Jayarrum



Fischer



Atkinson



Paull



Johnson



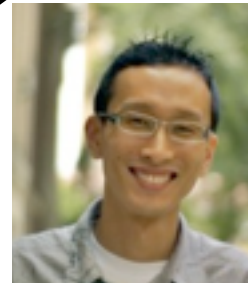
Stevens

Saccharomyces

Drosophila



Gottlieb



Sung



Roux



Brown



Lloyd

Arabidopsis



Browning



Juenger



Linder



Huq



Chen



Mehdy

Mutation-change in DNA sequence. Can lead to a change (usually loss) in gene expression.

Ser Glu Asp Tyr Val Tyr
AGC GAG GAC TAT GTT TAC G

Base change T → G

Ser Glu Asp Stop
AGC GAG GAC TAG GTT TAC G

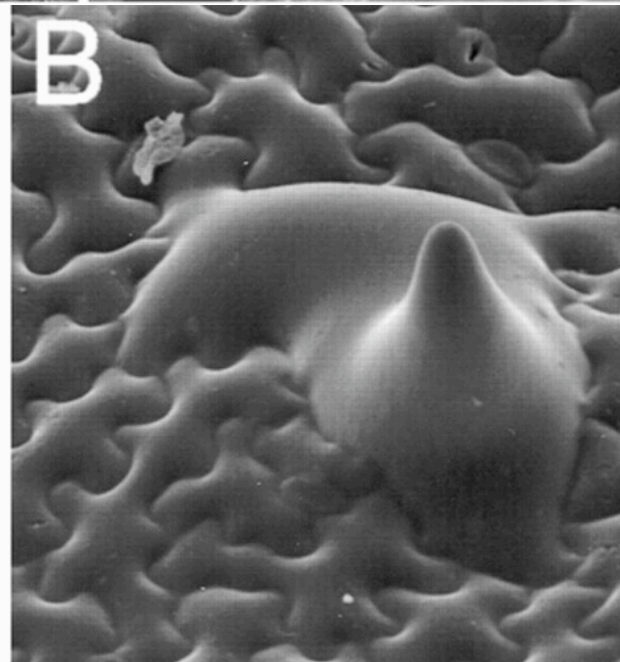
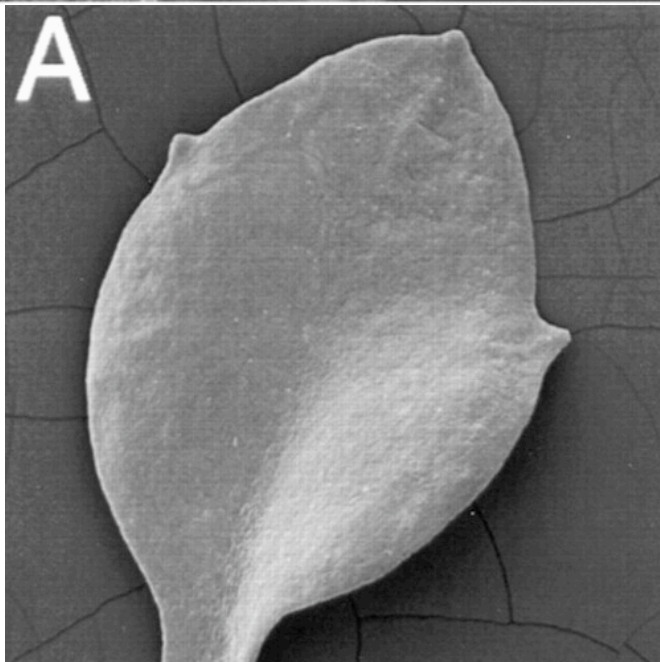
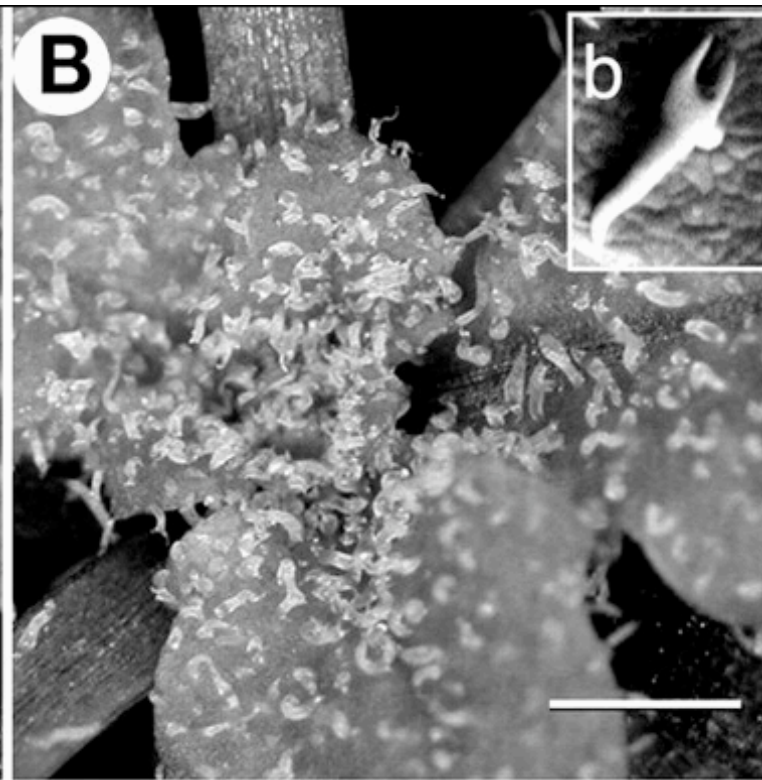
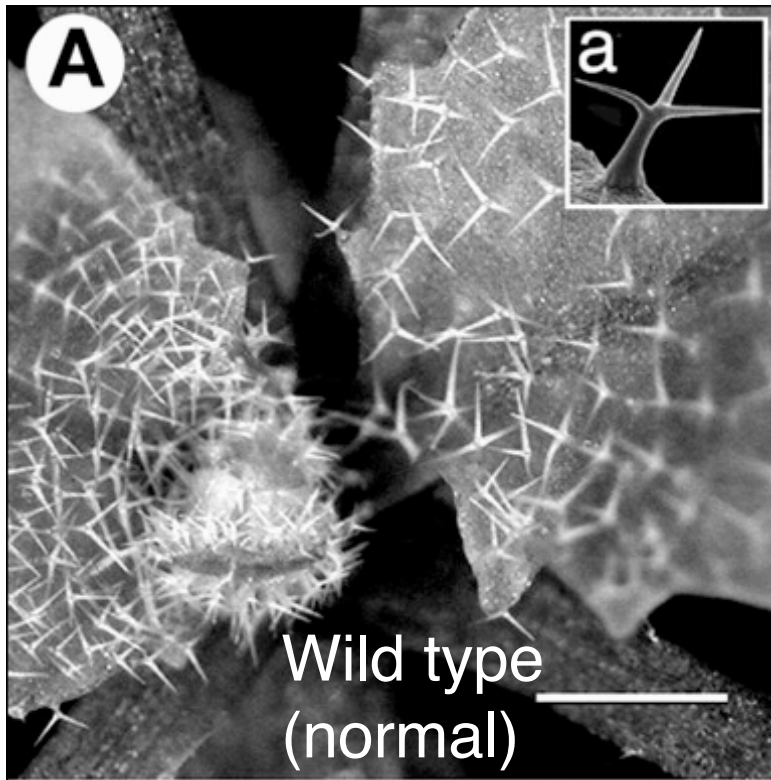
Truncation of protein.

Deletion of A

Ser Gly Thr Met Phe Thr
AGC GGG ACT ATG TTT ACG

Codon Frameshift changes aa sequence.

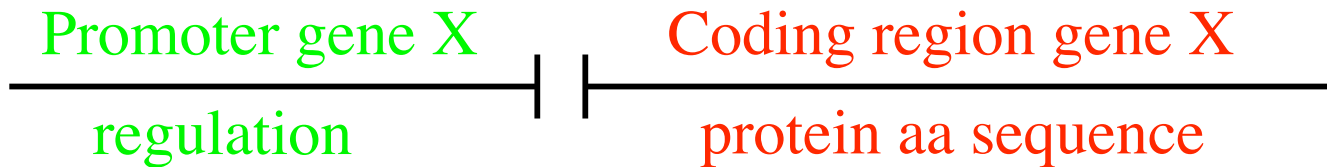
Other mutations include insertions, inversions, translocations.
Mutagens- chemical, x-ray, UV, sloppy polymerase.



Arabidopsis
trichome mutants

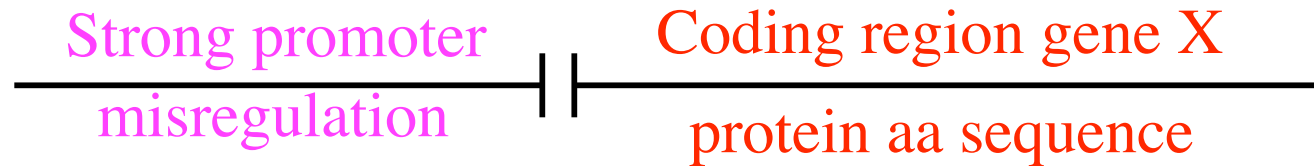
Gene organization and expression.

DNA ↔ RNA ↔ Protein “Central Dogma”

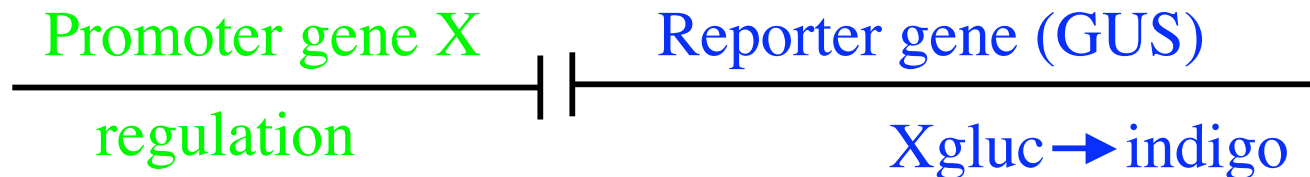


Transcriptional fusions

Change expression pattern of protein X by changing promoter

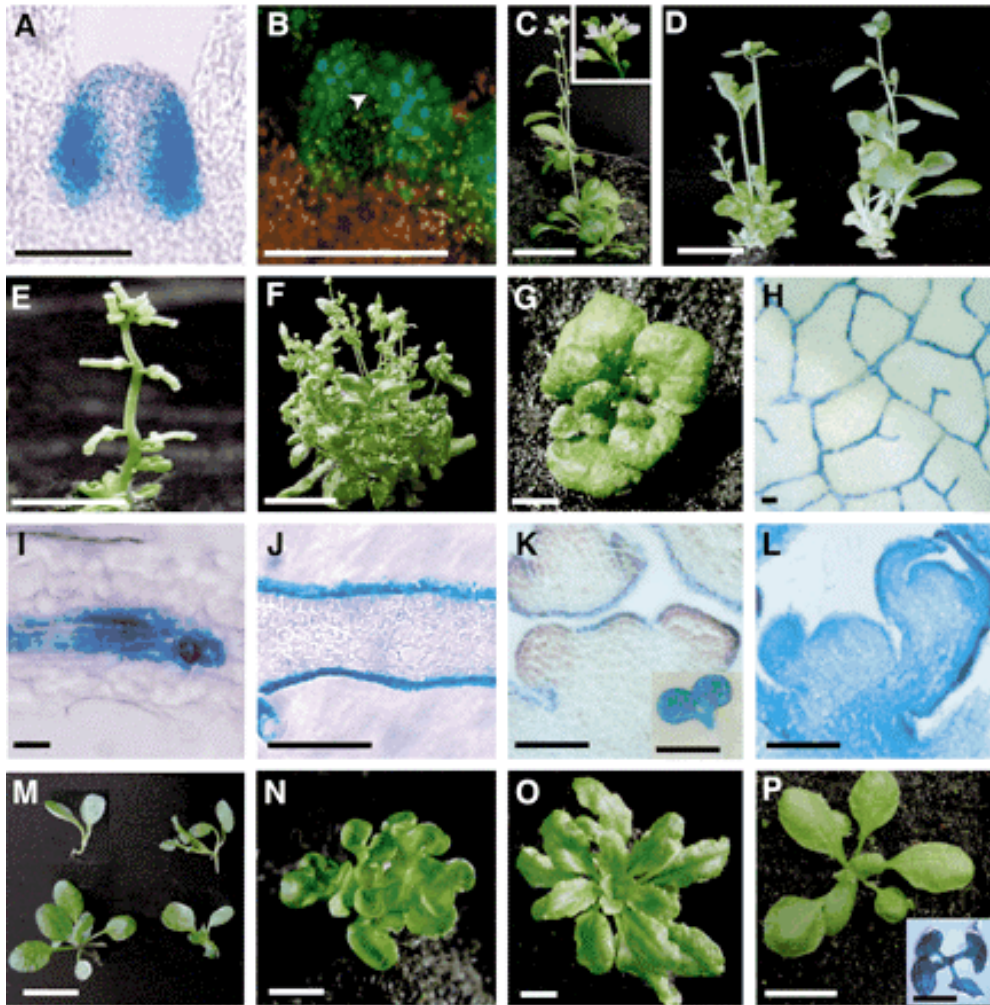


Observe expression pattern of promoter X by fusing to reporter gene

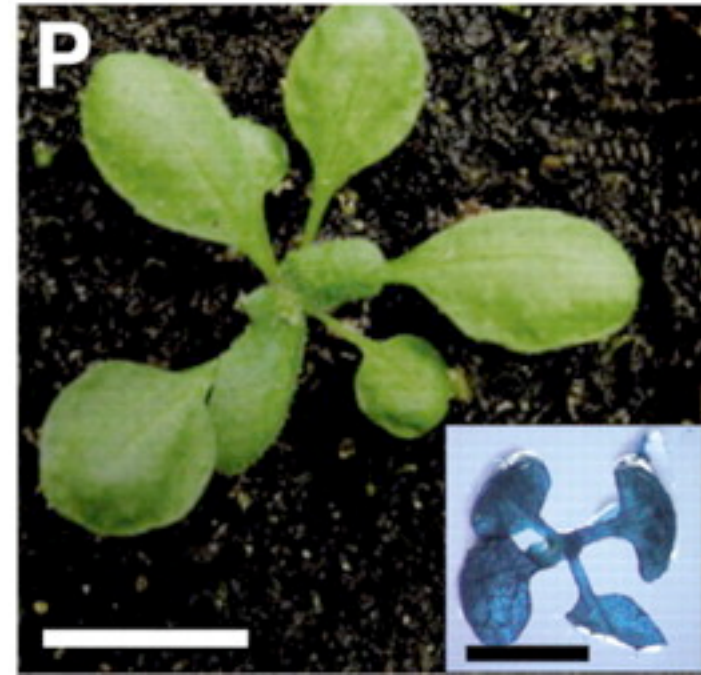


Basic tools to study gene function and expression in “model genetic organisms”

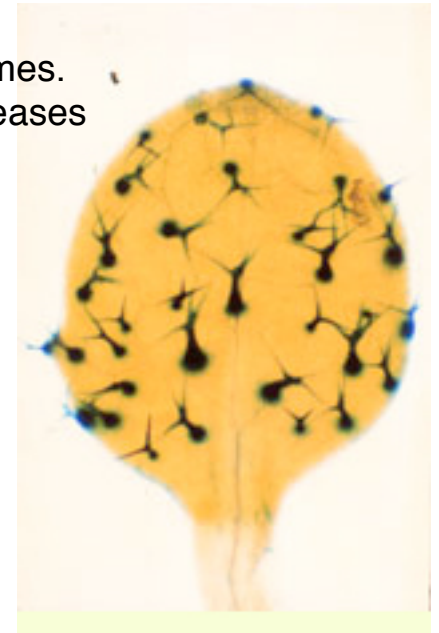
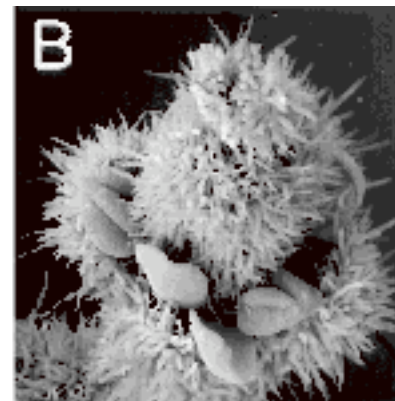
Where is a gene expressed?
 What does the gene control?



Transgene examples

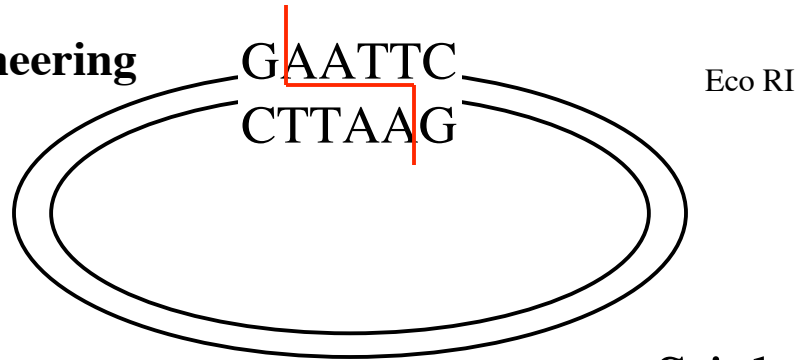


Gene expressed in trichomes.
 Overexpressed gene increases
 trichome number.

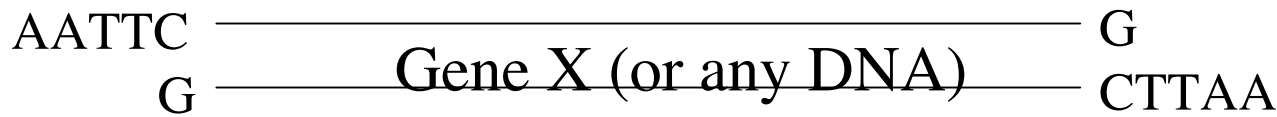


Molecular Biology and Genetic engineering With Plasmids

Restriction enzymes or endonucleases
Cut 4 to 6 bases mostly palindromes (dyad symmetry).



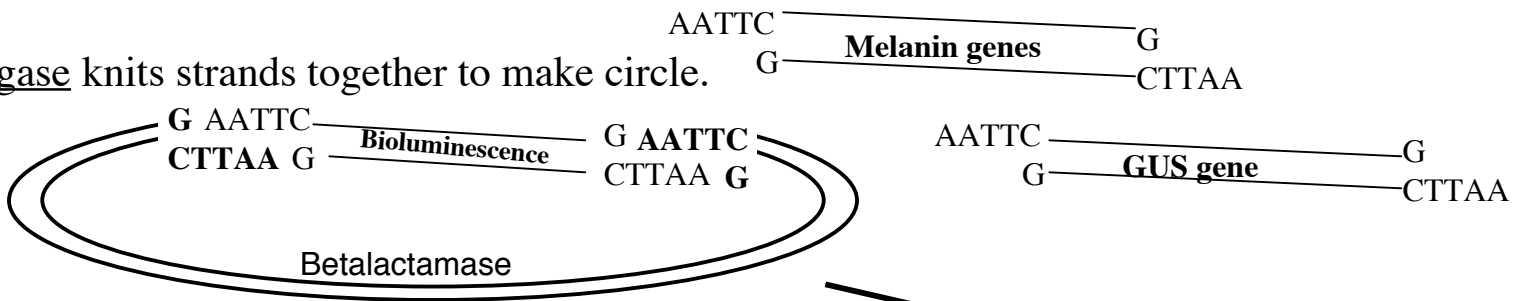
Sticky ends



Complementary bases

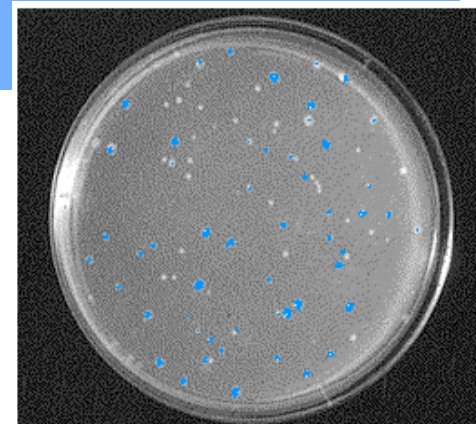
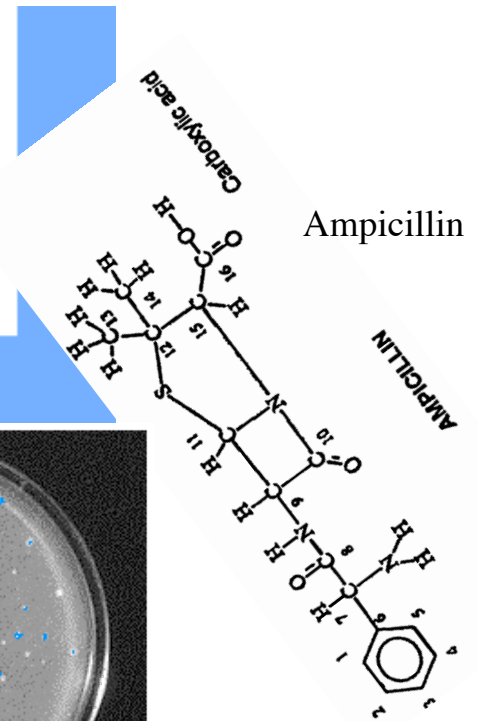
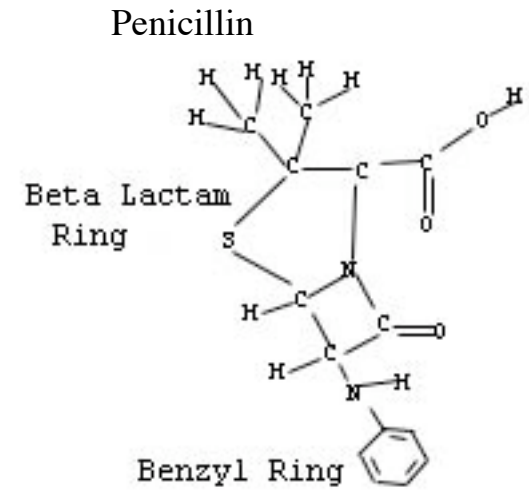
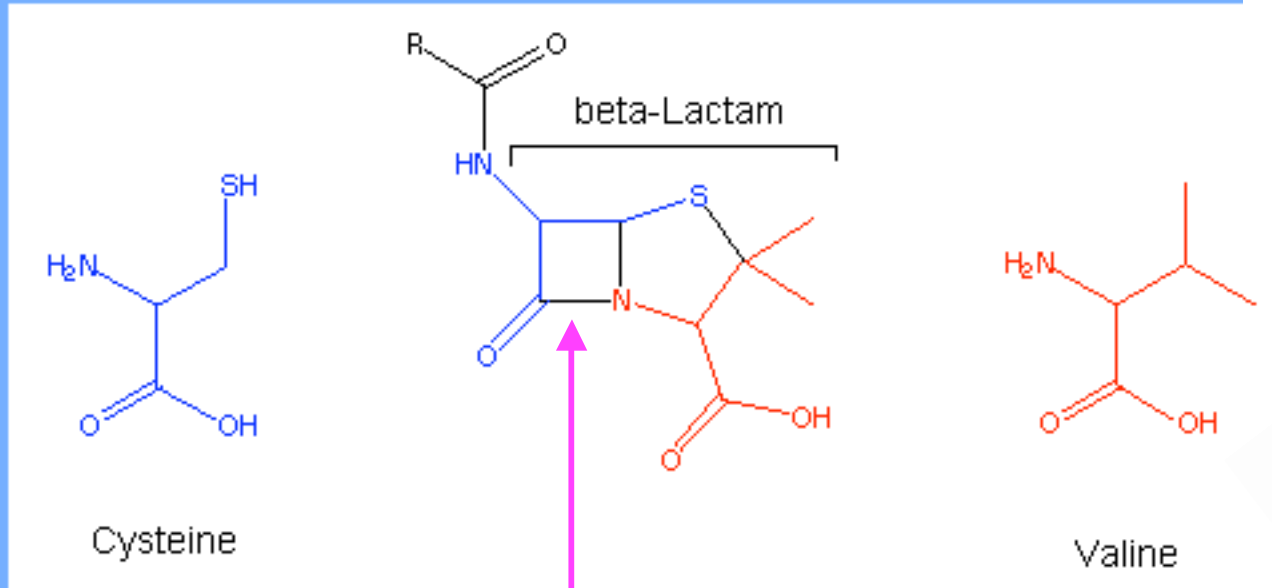


DNA ligase knits strands together to make circle.



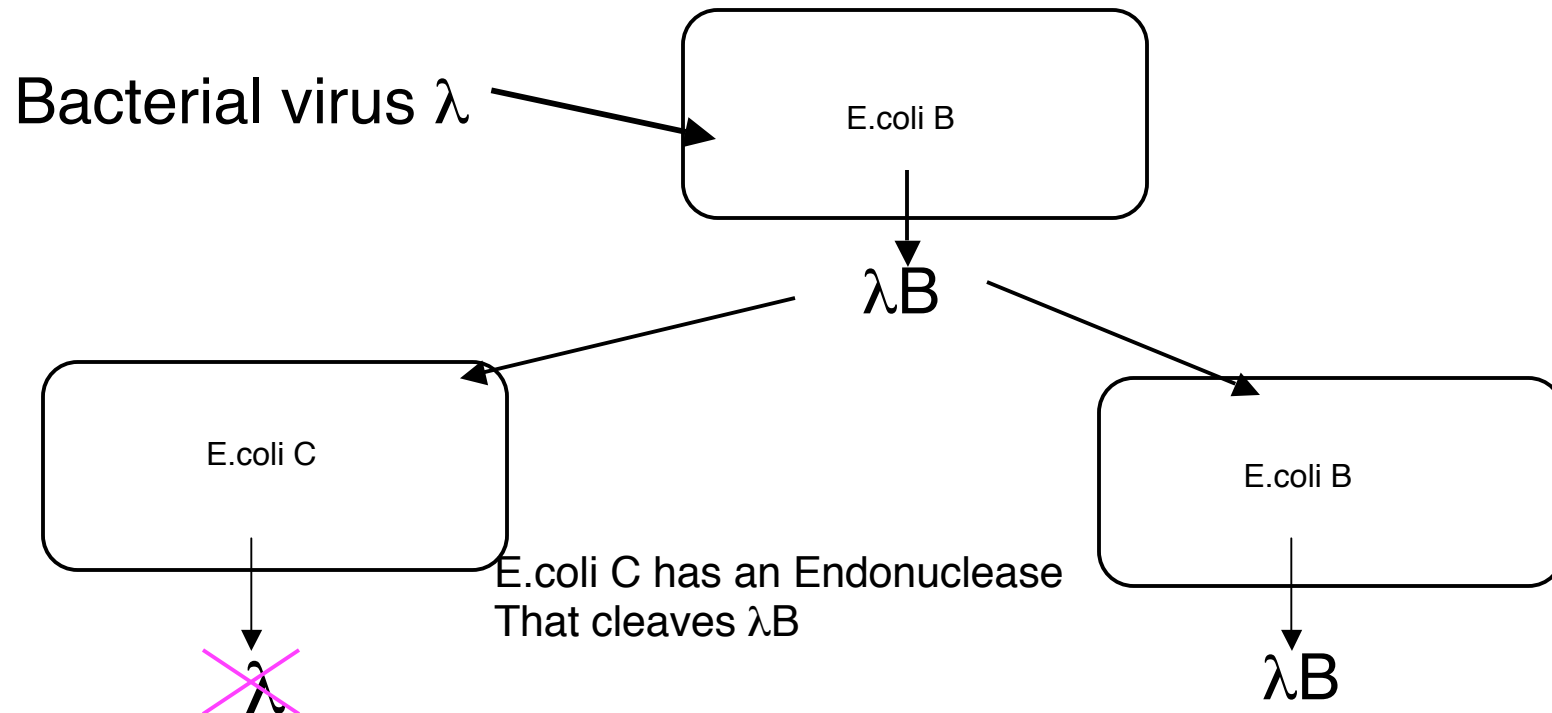
Transform E. coli

Penicillin - Beta Lactam Structure



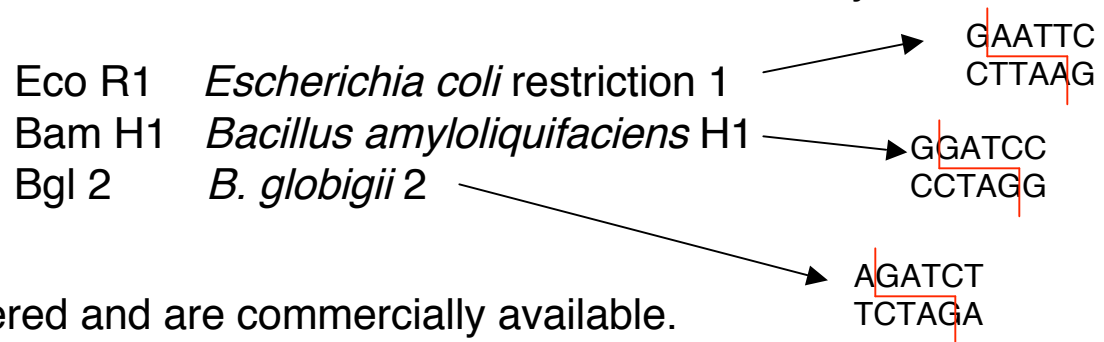
Beta lactamase cleaves lactam ring.

Restriction Endonucleases part of Restriction System Of most bacteria.



E.coli C is said to have a Restriction System which Restricts the growth of λB .

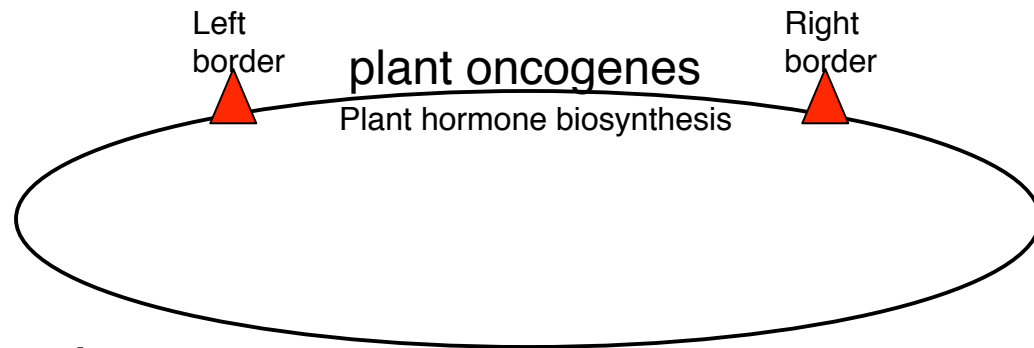
Most bacteria have some sort of restriction system.



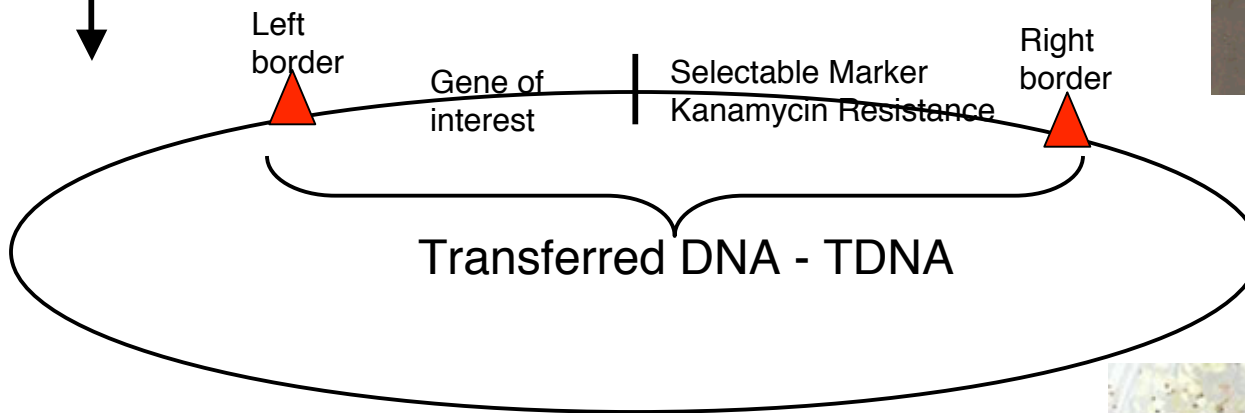
Hundreds have been discovered and are commercially available.

Agrobacterium tumefaciens crown gall disease

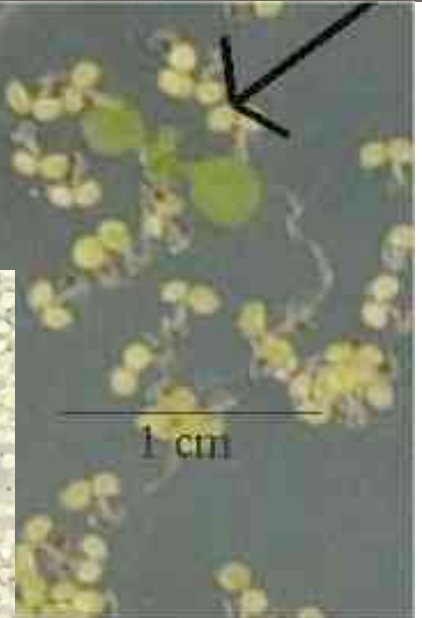
TI plasmid tumor inducing plasmid



Engineer oncogenes out--desired genes in.



Transform plant.



Genetically engineered products.

Flavr-savr tomato--altered senescence gene.

turn red on vine but don't soften. 1st commercial transgenic crop product.

Insect resistance--*Bacillus thuringiensis* insect toxin gene.

Bt cotton, corn, soybean, etc. **Bollgard®** **YieldGard®**

Herbicide resistance--EPSP synthase-aromatic amino acid.

Roundup-ready cotton, etc.

Companies rejecting GMO--Frito Lay, Gerber, Heinz,
Seagram, McDonald's

Global Status of Biotech Crops in 2003



Increase over 2002



■ 18 countries have adopted biotech crops

In 2003, global area of biotech crops was 167.2 million acres, representing an increase of 15% acres over 2002.

Source: Clive James, 2003, *International Service for the Acquisition of Agri-biotech Applications*

125,000 acres or more

USA:	105.7 million
Argentina:	34.3 million
Canada:	10.9 million
Brazil:	7.4 million
China:	6.9 million
South Africa:	0.9 million
Australia:	0.25 million
India:	0.25 million
Romania:	<0.25 million
Uruguay:	<0.25 million

125,000 acres or less

Spain	Bulgaria
Mexico	Honduras
Philippines	Germany
Colombia	Indonesia

Human single gene traits.

1. A. Hair between 1st and 2nd knuckle.
B. No hair ----

2. A. Widow's peak.
B. No widow's peak.

3. A. Detached ear lobe.
B. Attached ear lobe.