**Mechanisms of Infectious Disease • Fall 2008**

**Lecture 1**

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## Genetic Basis of Variation in Bacteria

I. Organization of genetic material in bacteria
- a. chromosomes
- b. plasmids

II. Genetic variation: Source
- a. point mutation
- b. DNA rearrangements

III. Genetic variation: Transmission
- a. transformation
- b. transduction
- c. conjugation

IV. Genetic variation: Implications for pathogenesis

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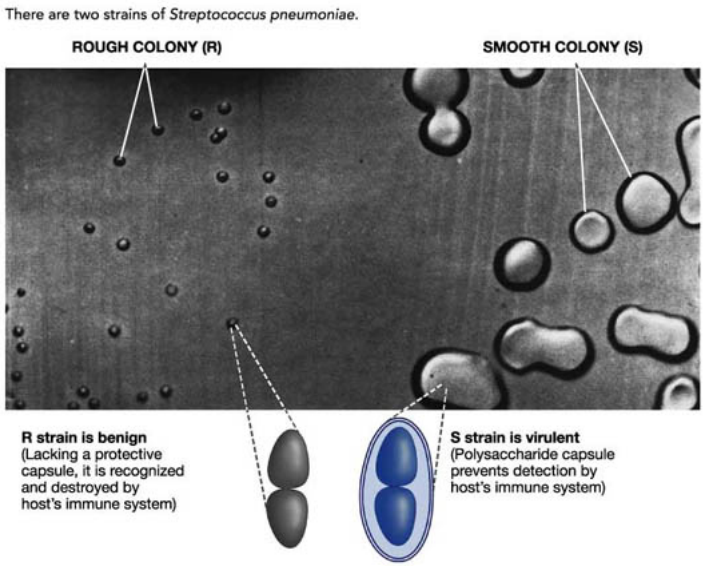
## Genetic basis of variation: Griffiths (1928)

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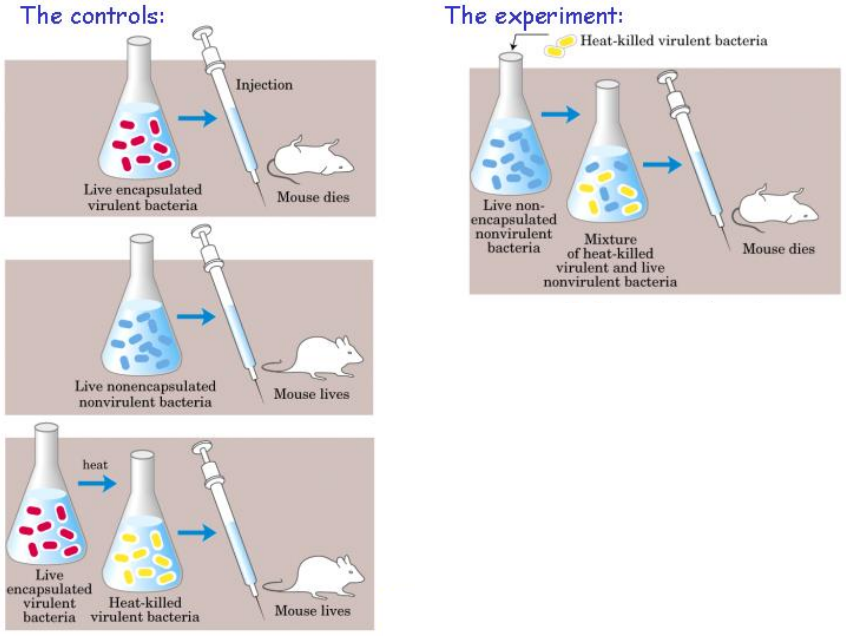
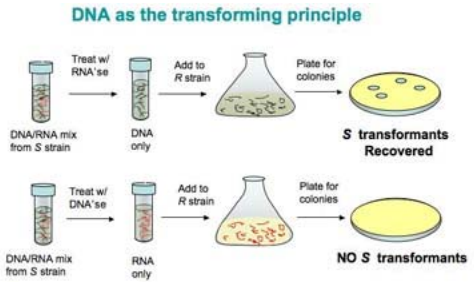
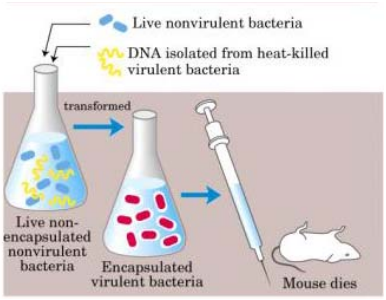
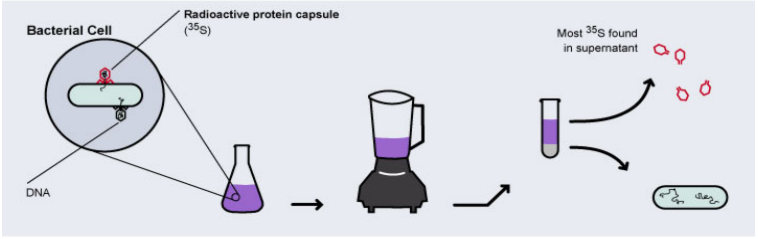


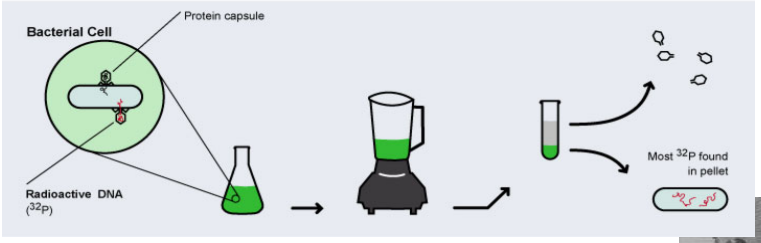
## Genetic basis of variation: Avery et al. (1944)




## Genetic basis of variation: Hershey and Chase (1952)

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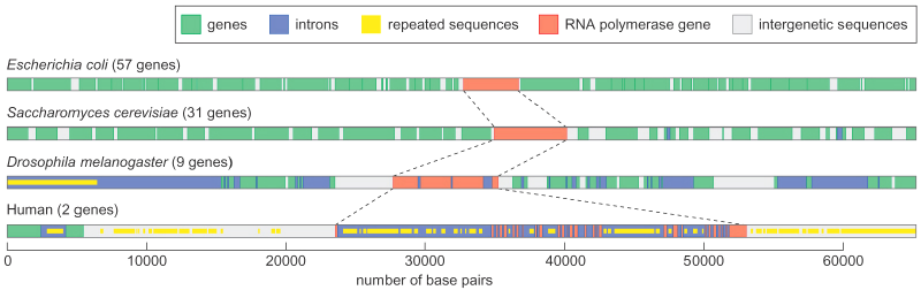
## Organization of genetic material in bacteria: chromosomes

- **Most** bacteria contain a single chromosome (+ extrachromosomal elements)
- **Some** bacteria have been found also to contain 2-3 replicons which can be considered either megaplasmids or minichromosomes e.g. 3.0 Mb and 0.9 Mb replicons in *Rhodobacter sphaeroides*
- A **few** bacterial genera contain >1 chromosome e.g. 2.1 Mb and 1.2 Mb chromosomes in *Brucella*
- **Some** bacteria harbour large replicons essential for survival in a specific ecological niche but not under laboratory conditions e.g. 1.4 Mb and 1.7 Mb replicons in *Rhizobium meliloti* are required for plant symbiosis

## Organization of genetic material in bacteria: chromosomes

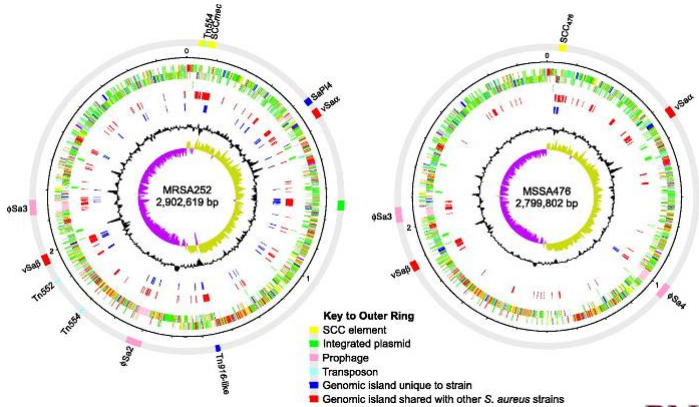





## Organization of genetic material in bacteria: chromosomes

**Complete genomes of two clinical *Staphylococcus aureus* strains: Evidence for the rapid evolution of virulence and drug resistance**

Matthew T. G. Holden*, Edward J. Feil*, Jodi A. Lindsay*, Sharon J. Peacock[§¶], Nicholas P. J. Day[§¶], Mark C. Enright*, Tim J. Foster[‖], Catrin E. Moore[§], Laurence Hurst*, Rebecca Atkin*, Andrew Barron*, Nathalie Bason*, Stephen D. Bentley*, Carol Chillingworth*, Tracey Chillingworth*, Carol Churcher*, Louise Clark*, Craig Corton*, Ann Cronin*, Jon Doggett*, Linda Dowd*, Theresa Feltwell*, Zahra Hance*, Barbara Harris*, Heidi Hauser*, Simon Holroyd*, Kay Jagels*, Keith D. James*, Nicola Lennard*, Alexandra Line*, Rebecca Mayes*, Sharon Moule*, Karen Mungall*, Douglas Ormond*, Michael A. Quail*, Ester Rabbinowitsch*, Kim Rutherford*, Mandy Sanders*, Sarah Sharp*, Mark Simmonds*, Kim Stevens*, Sally Whitehead*, Bart G. Barrell*, Brian G. Spratt**, and Julian Parkhill*[††]

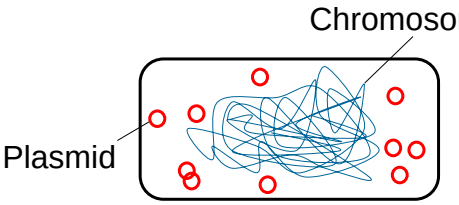


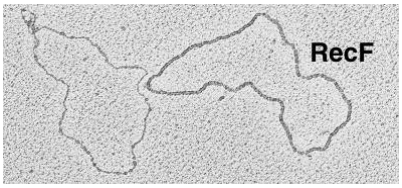



PNAS

## Organization of genetic material in bacteria: plasmids

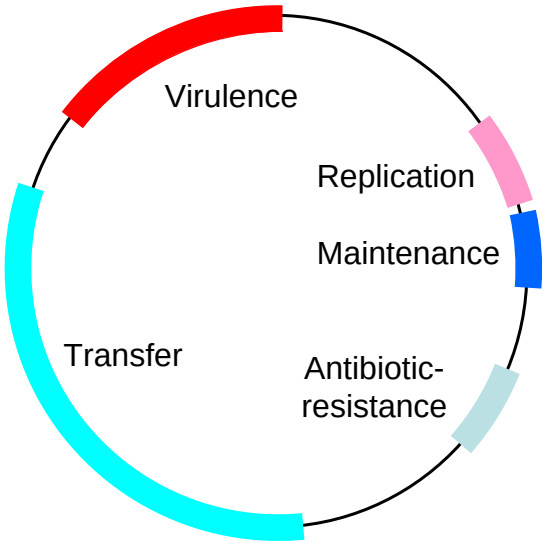
- Extrachromosomal
- Circular or linear
- 2 kb to hundreds of kb in size
- Non-essential
- May carry ‘supplemental’ genetic information or may be cryptic
- Employ host functions for most of DNA metabolism



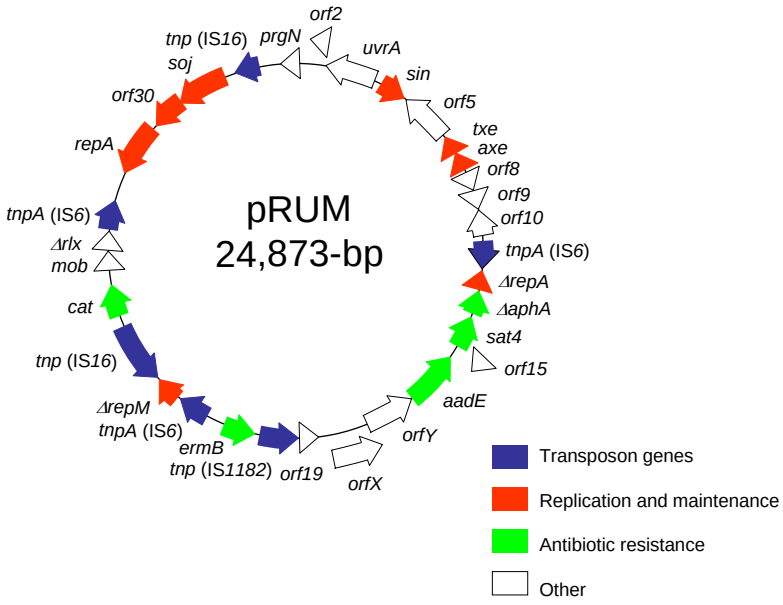




http://www.biochem.wisc.edu/inman/empics/dna-prot.htm

## Organization of genetic material in bacteria: plasmids



## Organization of genetic material in bacteria: plasmids




## Organization of genetic material in bacteria: plasmids

**Examples of naturally-occuring plasmids and relevant features**

| Plasmid | Host | Plasmid size (kb) | Relevant feature |
|---|---|---|---|
| pT181 | *Staphylococcus aureus* | 4.4 | Tetracycline resistance |
| ColE1 | *Escherichia coli* | 6.6 | Colicin production and immunity |
| pGKL2 | *Kluyveromyces lactis*[b] | 13.5 | Killer plasmid |
| pAMβ1 | *Enterococcus faecalis* | 26.0 | Erythromycin resistance |
| pSK41 | *Staphylococcus aureus* | 46.4 | Multidrug resistance |
| pBM4000 | *Bacillus megaterium* | 53.0 | rRNA operon |
| pI258 | *Staphylococcus aureus* | 28.0 | Metal ion resistance |
| pSLT | *Salmonella enterica* subsp. *typhimurium* | 93.9 | Virulence determinants |
| pMT1 | *Yersinia pestis* | 101.0 | Virulence determinants |
| pADP-1 | *Pseudomonas* sp. | 108.8 | Atrazine (herbicide) catabolism |
| pWW0 | *Pseudomonas putida* | 117.0 | Aromatic hydrocarbon degradation |
| pBtoxis | *Bacillus thuringiensis* subsp. *israelensis* | 137.0 | Mosquito larval toxicity |
| pX01 | *Bacillus anthracis* | 181.7 | Exotoxin production |
| pSOL1 | *Clostridium acetobutylicum* | 192.0 | Solvent production |
| pSymB | *Sinorhizobium meliloti* | 1683.3 | Functions associated with plant symbiosis |

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## Sources of genetic variation: point mutations

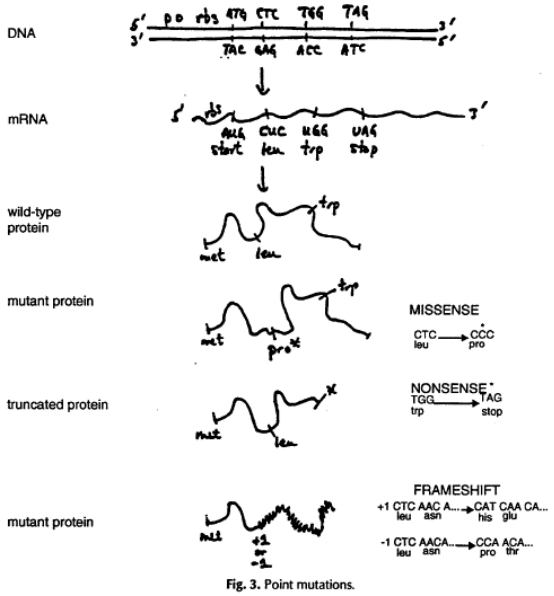
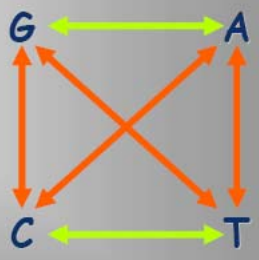



Fig. 3. Point mutations.

Sources of genetic variation: point mutations

## Point Mutations

- Transitions
  - Purine ⟺ Purine
  - Pyrimide ⟺ Pyrimide
    - A ⟺ G
    - C ⟺ T
- Transversions
  - Purine Pyrimide
    - A ⟺ C
    - A ⟺ T
    - G ⟺ C
    - G ⟺ T




There are four types of transitions.
There are eight types of transversions

Sources of genetic variation: point mutations

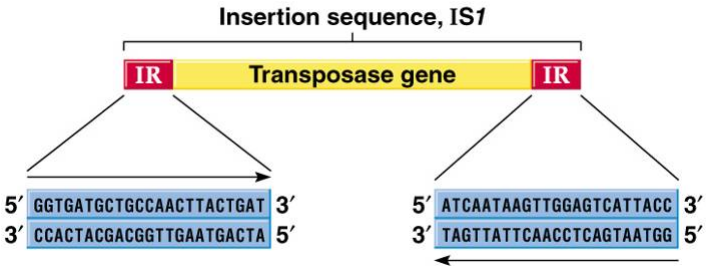
## Mutation phenotypes

- Silent mutation (synonymous), no change in amino acid
  AGG > AGA , both codons specify Arginine
- Missense mutation (replacement; nonsynonymous), change in amino acid
  - Nonsynonymous missense (or radical replacement)
    UUU (Phe) > UCU (Ser); Phe is hydrophobic and Ser is polar
- Nonsence mutation, premature termination of translation
  CAG (Gln) > UAG (Stop)
- Frameshift, addition or deletion of base pairs, not in a multiple of three, within the coding region of a gene.

## Sources of genetic variation: DNA rearrangements

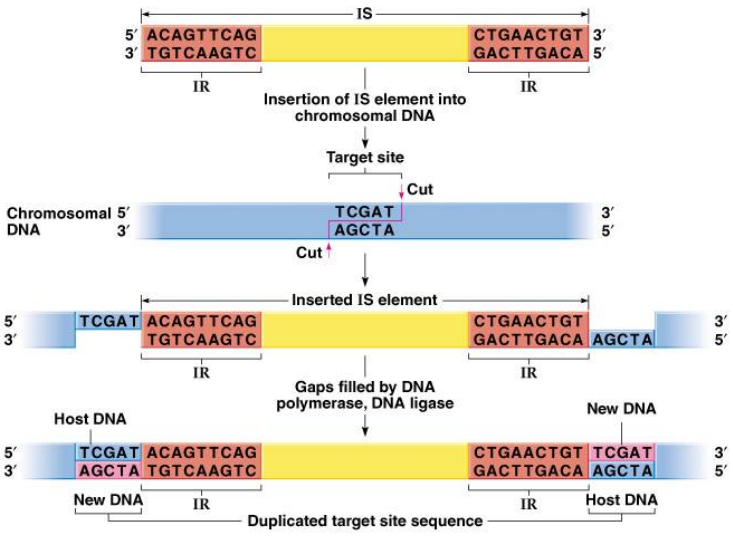
**Insertion sequence (IS) elements:**

1. **Simplest type of transposable element found in bacterial chromosomes and plasmids.**
2. **Encode only genes for mobilization and insertion.**
3. **Range in size from 768 bp to 5 kb.**
4. **IS1 first identified in *E. coli*'s glactose operon is 768 bp long and is present with 4-19 copies in the *E. coli* chromosome.**
5. **Ends of all known IS elements show inverted terminal repeats (ITRs).**




## Sources of genetic variation: DNA rearrangements

**Integration of IS element in chromosomal DNA.**



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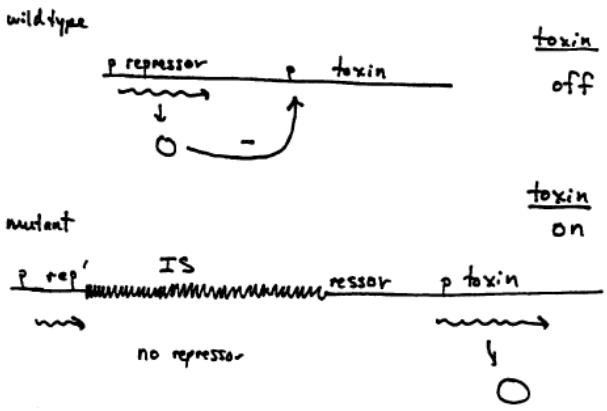



Fig. 5 *(at left).* Disruption of a gene by IS element transposi-

## Sources of genetic variation: DNA rearrangements

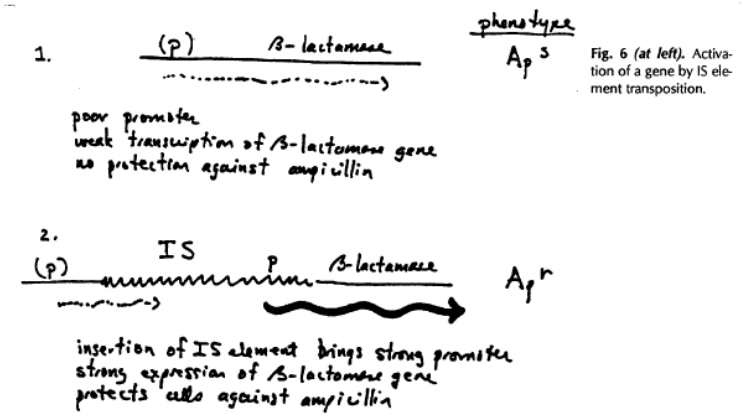
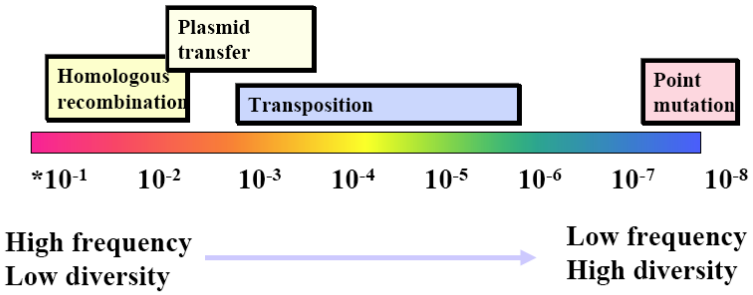



Fig. 6 *(at left).* Activation of a gene by IS element transposition.

Sources of genetic variation: frequency of occurrence




High frequency
Low diversity

Low frequency
High diversity

* As frequency per cell per generation

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## Transmission of genetic variation

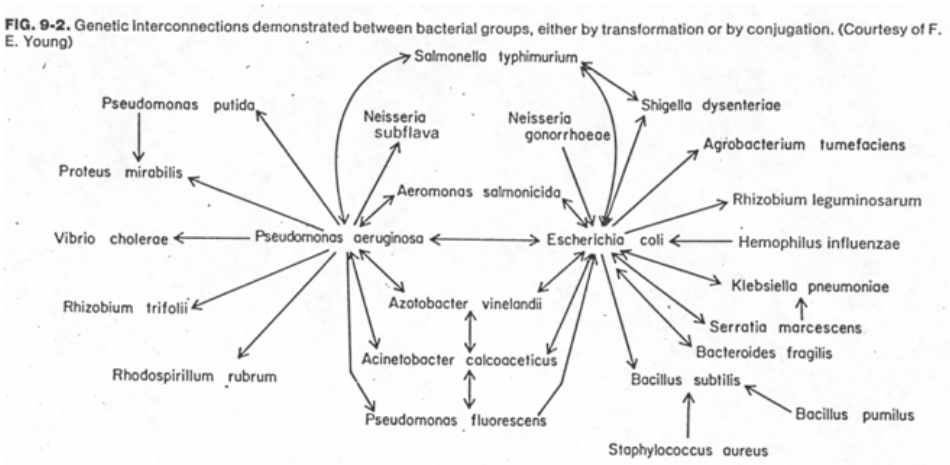
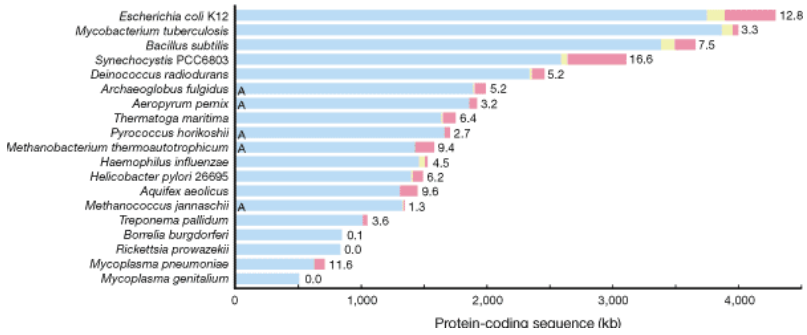
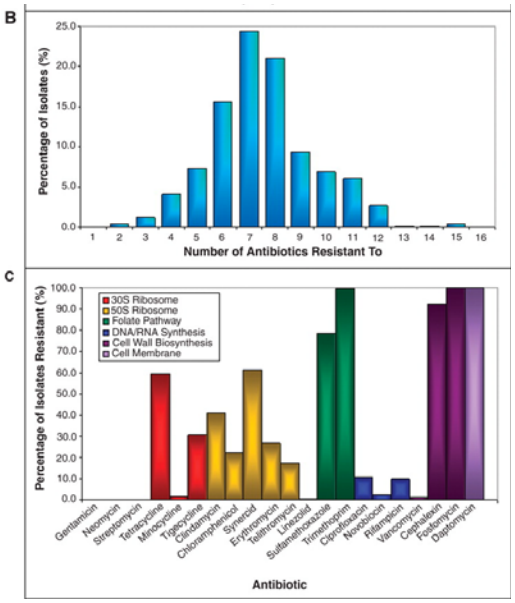



**FIG. 9-2.** Genetic interconnections demonstrated between bacterial groups, either by transformation or by conjugation. (Courtesy of F. E. Young)




**Lengths of bars denote the amount of protein-coding DNA. For each bar, the native DNA is blue; foreign DNA identifiable as mobile elements, including transposons and bacteriophages, is yellow, and other foreign DNA is red. The percentage of foreign DNA is noted to the right of each bar. 'A' denotes an Archaeal genome.**

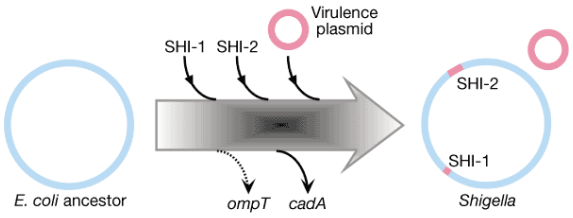
## Transmission of genetic variation: antibiotic resistance




V. M. D'Costa et al. Science (2006)

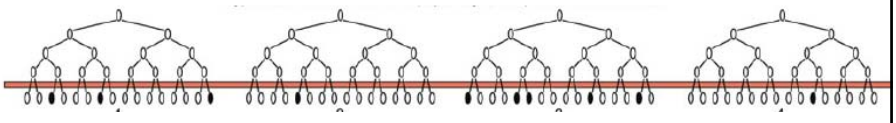
**Fig. 1. Antibiotic resistance profiling of 480 soil-derived bacterial isolates**

## Transmission of genetic variation: pathogenesis



## Transmission of genetic variation: Luria-Delbruck test

1. Resistance by mutation is a physiological response



Time of exposure to selective agent

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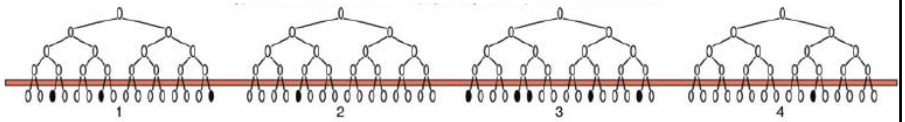
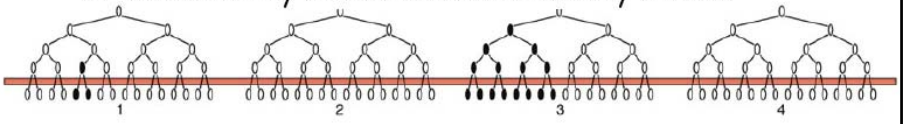



2. Resistance by mutation arises randomly in time

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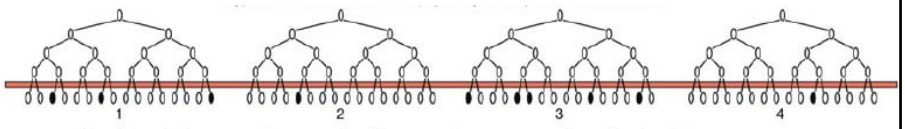


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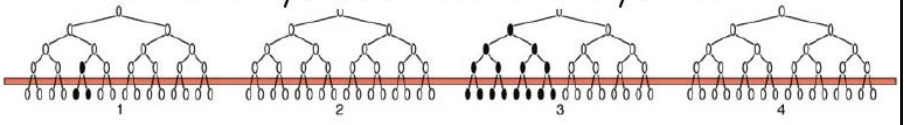
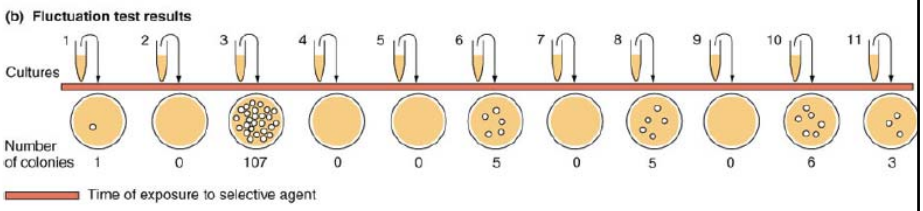



Results fit with expectations if random mutation occur at random.

## Linear transmission of genetic variation

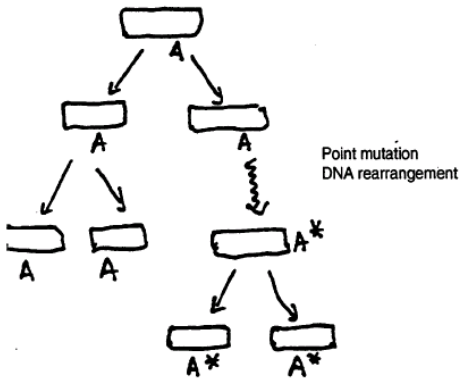



Fig. 1. Clonal variation.

## Horizontal transmission of genetic variation

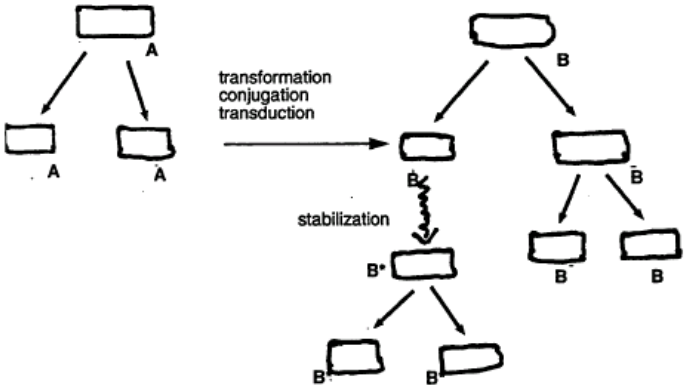
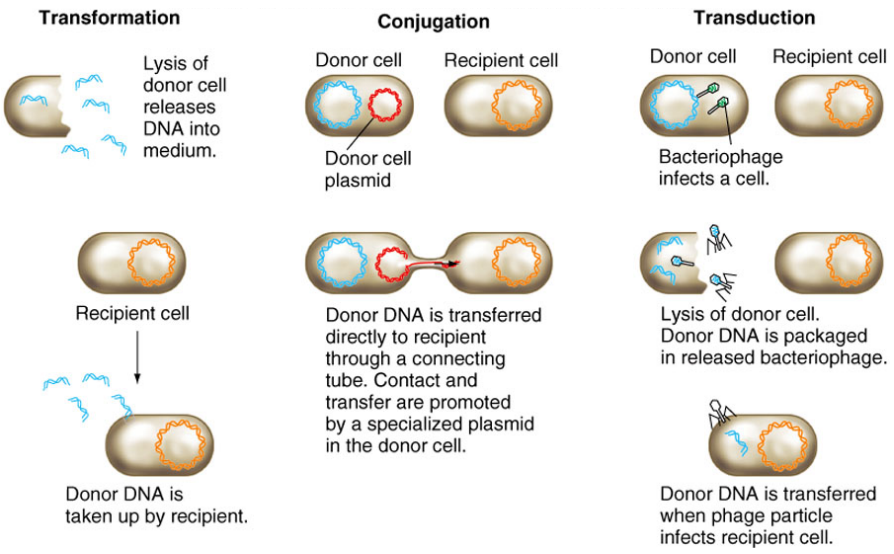
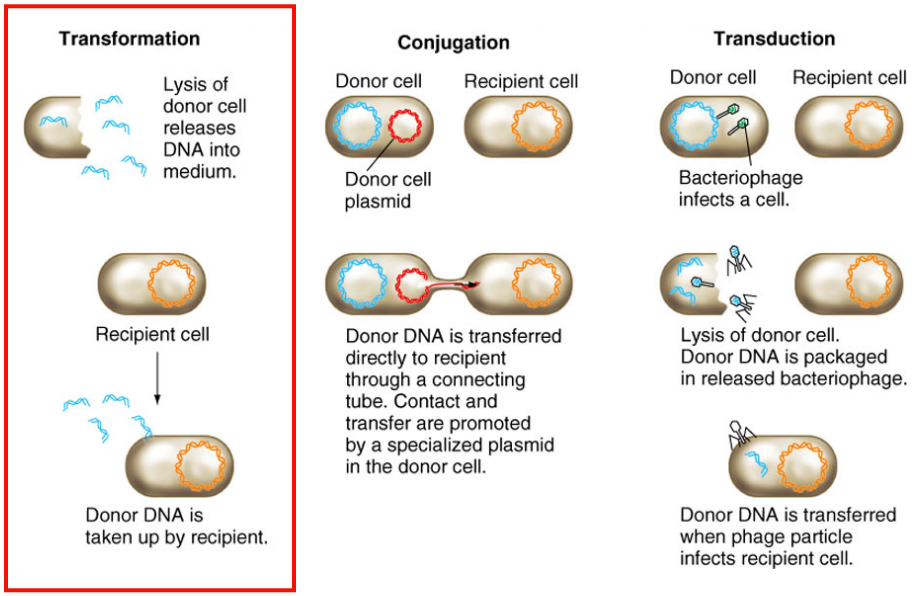



**Fig. 2 *(at left).*** Variation by transfer of genetic information.

## Transmission of genetic variation: mechanisms



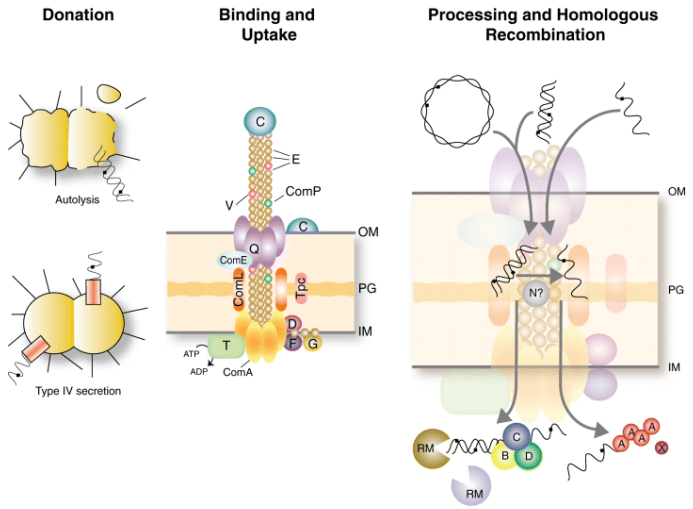
## Transmission of genetic variation: transformation



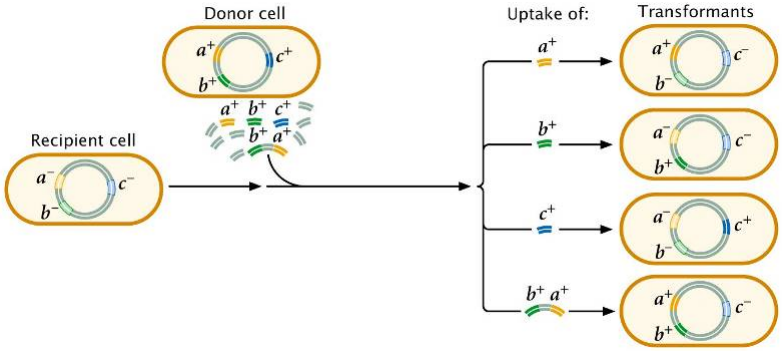

## Transmission of genetic variation: transformation

- Gene transfer resulting from the uptake of DNA from a donor.
- Factors affecting transformation
  - DNA size and state
    - Sensitive to nucleases
  - Competence of the recipient (*Bacillus, Haemophilus, Neisseria, Streptococcus*)
    - Competence factor
    - Induced competence

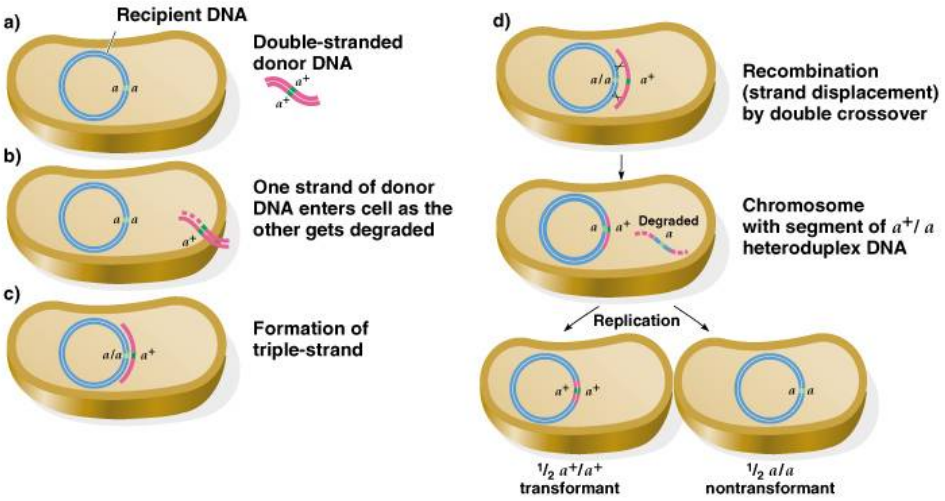
## Transmission of genetic variation: transformation



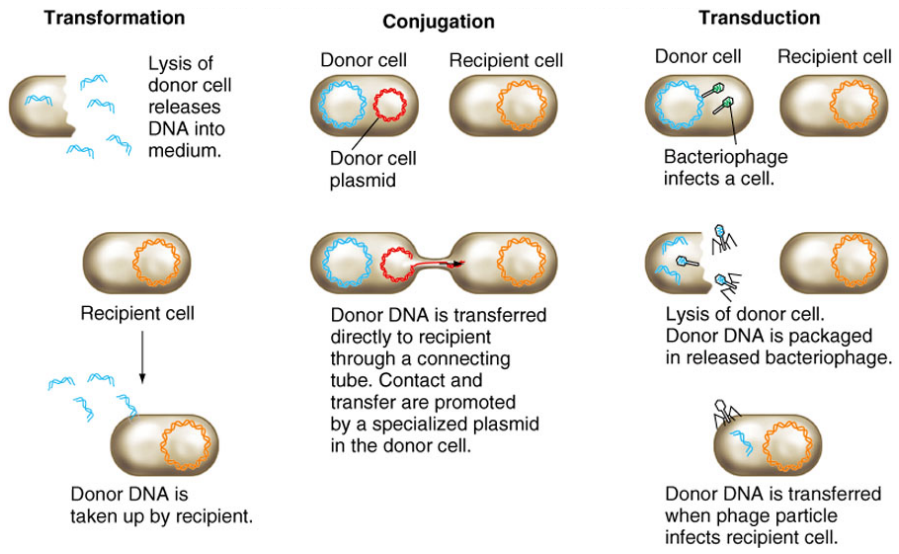

## Transmission of genetic variation: transformation



## Transmission of genetic variation: transformation




## Transmission of genetic variation: mechanisms



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