Mutations

Spontaneous mutations are those that occur without a known cause. They may truly be spontaneous, resulting from a low level of inherent metabolic errors, or they may actually be caused by unknown agents present in the environment. Induced mutations, as already discussed, are those resulting from exposure of organisms to physical and chemical agents that cause changes in DNA (or RNA in some viruses). Such agents are called mutagens; they include ionizing irradiation, ultraviolet light, and a wide variety of chemicals.

Measurements of spontaneous mutation frequencies for various genes of phage and bacteria range from about 10^{-8} to 10^{-10} detectable mutations per nucleotide pair per generation. For eukaryotes, estimates of mutation rates range from about 10^{-7} to 10^{-9} detectable mutations per nucleotide pair per generation. The mutation rate per gene varies from about 10^{-4} to 10^{-7} per generation.

Replica-plating technique

Consider a population of bacteria such as E. coli growing in a streptomycin-free environment. When exposed to streptomycin, most of the bacteria will be killed by the antibiotic. However, if the population is large enough, it will soon give rise to a streptomycin-resistant culture in which all the cells are resistant to the antibiotic. Does streptomycin simply select rare, randomly occurring mutants that preexist in the population, or do all of the cells have some low probability of developing resistance in response to the presence of streptomycin? How can geneticists distinguish between these two possibilities? Resistance to streptomycin can only be detected by treating the culture with the antibiotic. How, then, can a geneticist determine whether resistant bacteria are present prior to exposure to streptomycin, or are induced by the presence of the antibiotic? In 1952,Joshua and Esther Lederberg developed an important new technique called replica plating. This technique allowed them to demonstrate the presence of antibiotic-resistant mutants in bacterial cultures prior to exposure to the antibiotic (Fig 1). The Lederbergs first diluted the bacterial cultures, spread the bacteria on the surface of semisolid nutrient agar medium in petri dishes, and incubated the plates until each bacterium had produced a visible colony on the surface of the agar. They next inverted each plate and pressed it onto sterile velvet placed over a wood block. Some of the cells from each colony stuck to the velvet. They then

gently pressed a sterile plate of nutrient agar medium containing streptomycin onto the velvet. They repeated this replica-plating procedure with many plates, each containing about 200 bacterial colonies. After they incubated the selective plates (those containing streptomycin) overnight, rare streptomycin-resistant colonies had formed. The Lederbergs subsequently tested the colonies on the nonselective plates (those not containing streptomycin) for their ability to grow on medium containing streptomycin. Their results were definitive. The colonies that grew on the selective replica plates almost always contained streptomycin-resistant cells, whereas colonies that failed to grow on the selective medium seldom contained any resistant cells. If a mutation that makes a bacterium resistant to streptomycin occurs at an early stage in the growth of a colony, the resistant cell will divide and produce two, then four, then eight, and eventually a large number of resistant bacteria. Thus, if mutation is a randomly occurring, nonadaptive process, many of the colonies that form on the nonselective plates will contain more than one antibiotic-resistant bacterium and will give rise to resistant cultures when tested for growth on selective media. However, if mutation is adaptive and the mutations to streptomycin resistance occur only after exposure to the antibiotic, then the colonies on the nonselective plates that gave rise to resistant colonies on the selective plates after replica plating would be no more likely to contain streptomycin-resistant cells than the other colonies on the nonselective plates.Thus, by using their replica-plating technique, the Lederbergs demonstrated the existence of streptomycin-resistant mutants in a population of bacteria prior to their exposure to the antibiotic.

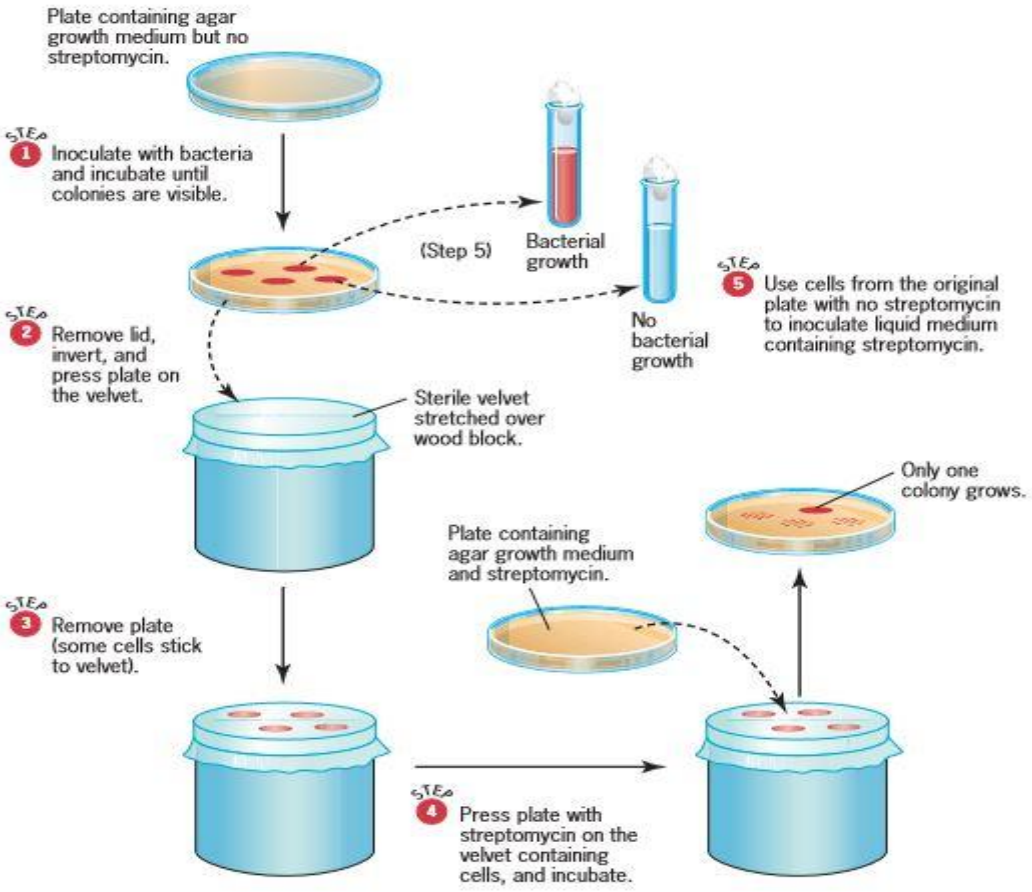

Fig 1.Replica-plating technique.

Is mutation reversible process?

The mutation of a wild-type gene to a form that results in a mutant phenotype is referred to as forward mutation.When a second mutationrestores the original phenotype lost because of an earlier mutation, the process is called reversion or reverse mutation. Reversion may occur in two different ways: (1) by back mutation, a second mutationat the same site in the gene as the original mutation, restoring the wild-type nucleotide sequence, or (2) by suppressor mutation, a second mutation at a different location in the genome, which compensates for the effects of the first mutation (Figure 2). Back mutation restores the original wild-type nucleotide sequence of the gene, whereas a suppressor mutation does not. Suppressor mutations may occur at distinct sites in the same gene as the original mutation or in different genes, even on different chromosomes. Some mutations revert primarily by back mutation, whereas others do so almost exclusively through the occurrence of suppressor mutations. Thus, in genetic studies, researchers

often must distinguish between these two possibilities by backcrossing the phenotypic revertant with the original wild-type organism. If the wild-type phenotype is restored by a suppressor mutation, the original mutation will still be present and can be separated from the suppressor mutation by recombination. If the wild-type phenotype is restored by back mutation, all of the progeny of the backcross will be wild-type.

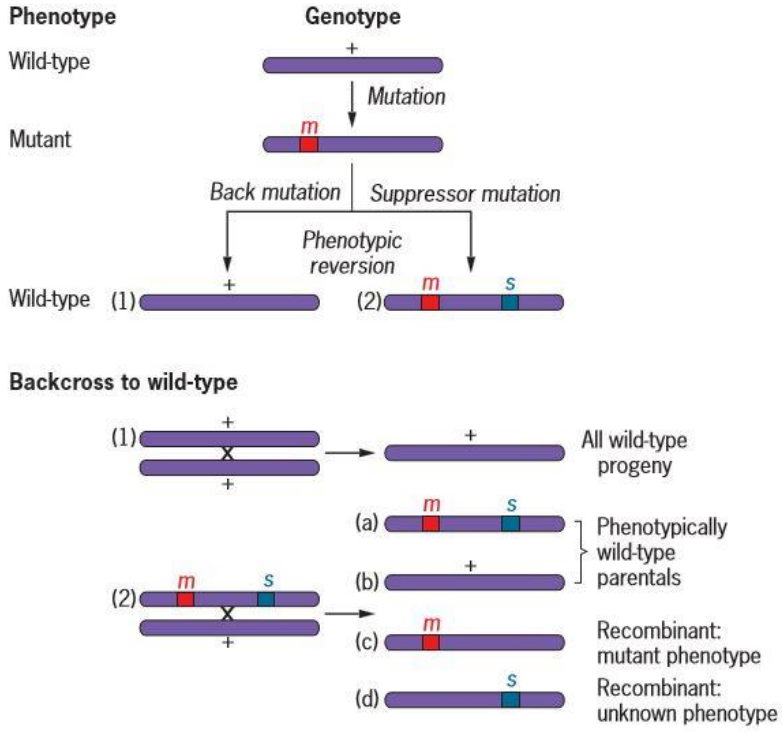

Fig 2.Restoration of the original wild-type phenotype of an organism may occur by (1) back mutation or (2) suppressor mutation

A gene is a sequence of nucleotide pairs that usually encodes a specific polypeptide. Any mutation occurring within a given gene will thus produce a new allele of that gene.

Isoalleles:Genes containing mutations with no effect on phenotype or small effects that can be recognized only by special techniques.

Null alleles:Genes containing mutations that result in no gene product or totally nonfunctional gene products.

Recessive lethals: Ifmutations resulting in non-functional gene products occur in genes that are required for the growth of the organism, individuals that are homozygous for the mutation will not survive.

Neutral mutations: Mutations resulting inno effect on the phenotype of the organism because of the degeneracy and order in the genetic code.

Mutations can be either recessive or dominant.

- In monoploid organisms such as viruses and bacteria, both recessive and dominant mutations can be recognized by their effect on the phenotype of the organism in which they occur.
- In diploid organisms such as fruit flies and humans, recessive mutations will alter the phenotype only when present in the homozygous condition. Thus, in diploids, most recessive mutations will not be recognized at the time of their occurrence because they will be present in the heterozygous state. X-linked recessive mutations are an exception; they will be expressed in the hemizygous state in the heterogametic sex (for example, males in humans and fruit flies; females in birds). X-linked recessive lethal mutations will alter the sex ratio of offspring because hemizygous individuals that carry the lethal will not survive.

Thus mutations, which cause random changes in the highly adapted amino acid sequences, usually will produce less active or totally inactive products (Fig 3).

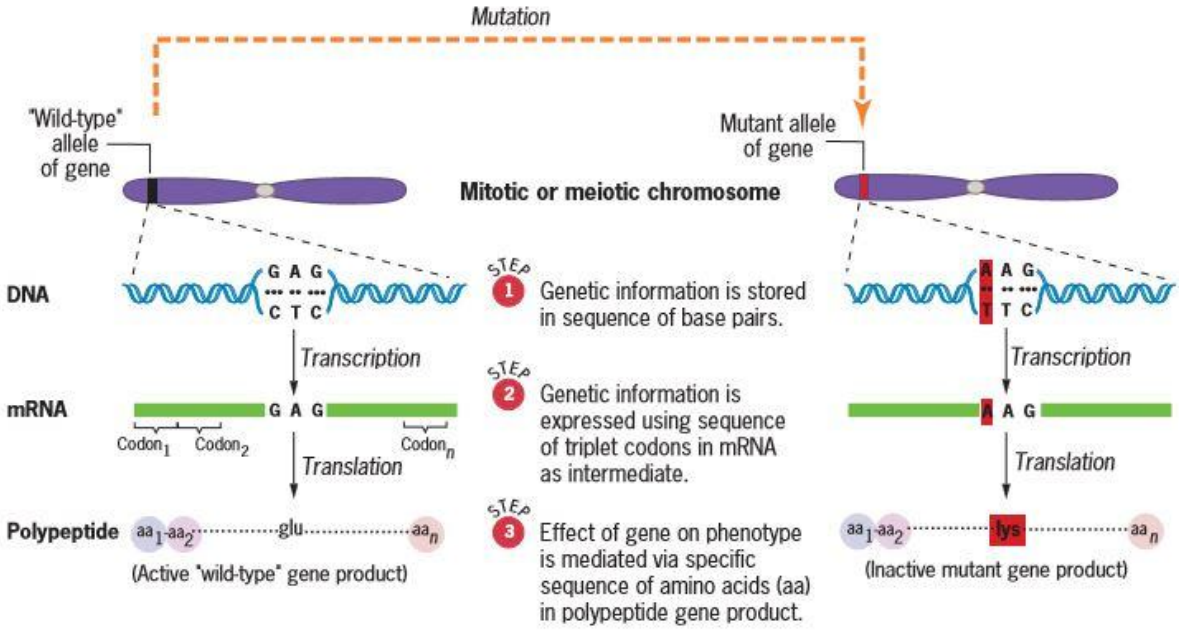

Fig 3.Process of mutation and expression of wild-type and mutant alleles.