Plasmids

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What are Plasmids?

- Plasmids are small circular DNA fragments, double-stranded, self-replicating extra chromosomal structures found in many microorganisms.
- The term Plasmid was coined by Joshua Lederberg in 1952.
- Plasmids are important genetic tools, used to introduce, manipulate or delete certain genes from the host cell.

Properties of Plasmids

- They are extrachromosomal DNA fragments present in the cell.
- They are double-stranded structures.
- They can replicate independently.
- The absence of a plasmid in the cell does not affect cell functioning, but the presence of a plasmid in the cell is usually beneficial.
- Plasmids are also known as sex factors, conjugants, extrachromosomal replicons, or transfer factors.
- The copy number refers to the number of copies of plasmid present in the bacterial cell. Usually, small plasmids are present in high numbers and large plasmids are present in few numbers.
- Compatibility of plasmids refers to the ability of two different plasmids to coexist in the same bacterial cell.

Types of Plasmids

- Based on the presence of the transfer genes, plasmids can be classified into two types:

1. **Conjugative plasmids:** contain a set of transfer genes which promote sexual conjugation between different cells (commonly seen in bacteria).
2. **Non-conjugative plasmids:** these types of plasmids lack the transfer genes.

Based on functions the plasmids can be classified into the following types:

1. F Plasmids (Fertility plasmids)

- They contain the TRA genes and hence can be transferred from one cell to another.
- They can replicate inside the bacterial cell.
- They cause the synthesis of a pilus, which is a long protein-rich structure that helps in cell-cell interaction.
- It also contains a sequence responsible for incompatibility.

2. R plasmid (Resistance plasmids)

- These plasmids contain and transmit genes for Antibiotic resistance from one cell to another.
- The antibiotic resistance gene protects the bacteria from antibiotics in human medicines and antibiotics naturally present in the soil.

3. Col Plasmids (Colicin plasmids)

- These are known as bacteriocinogenic plasmids because they produce bacteriocins.
- These proteins have the ability to kill the closely related bacterial cells which lack Col plasmids.
- These plasmids are observed in *E. coli*.

4. Degradative Plasmids

- These types of plasmids have the ability to digest unusual substances such as toluene, salicylic acid, etc.
- The presence of these plasmids in the organism enables the breakdown of various chemicals and substances.

5. Virulence Plasmids

- These plasmids produce virulence factors that enable the bacteria to infect other cells.
- Bactria containing virulence plasmids are able to infect the plant, animal, and human cells.
- Example / Ti plasmid is the virulence plasmid present in *Agrobacterium tumefaciens* which causes crown gall disease in plants.

Functions and Applications of Plasmids

- The important use of plasmids is that they can be used as vectors to insert a specific gene into other organisms due to their capacity to incorporate a gene and replicate inside the cell.
- They are an important factor in bacteria as they carry antibiotic-resistance genes.
- Degradative plasmids can be used to degrade industrial chemicals which are a threat to the environment.
- As plasmids are easy to manipulate, they are being used in gene therapy.
- Because plasmids are good vectors (a vehicle/factor which is used to transfer a gene from one organism to another) they are used in drug delivery and for hormone production in other cells.
- Plasmids are an important source of horizontal gene transfer.

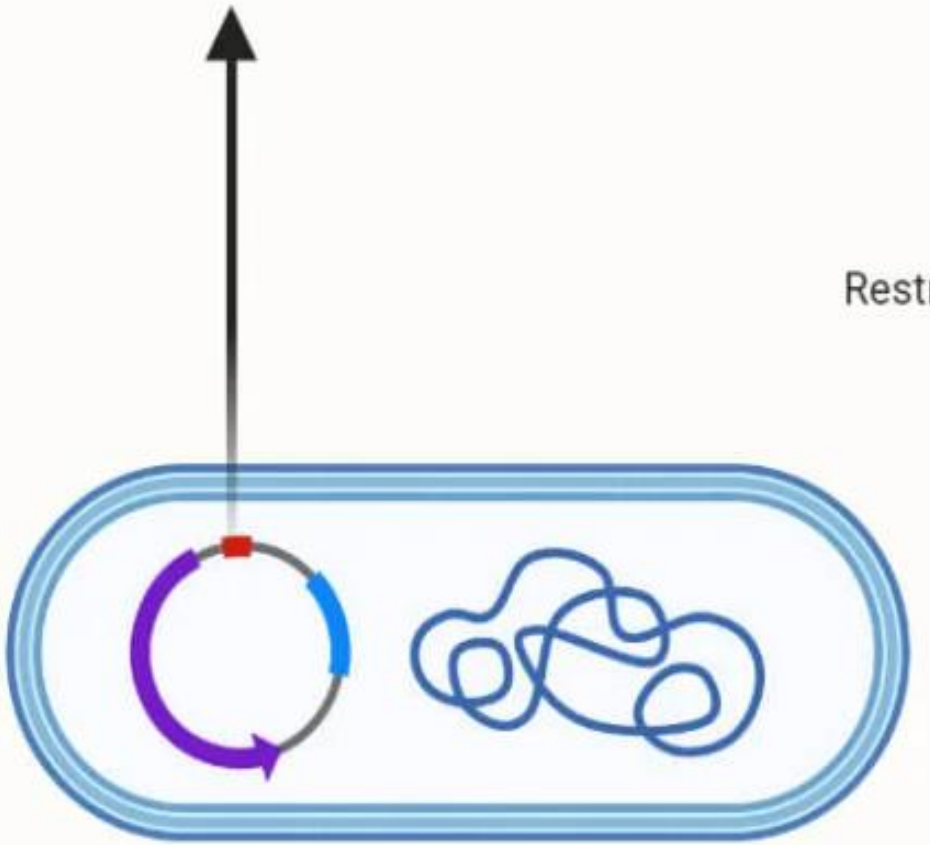
Structure of Plasmids

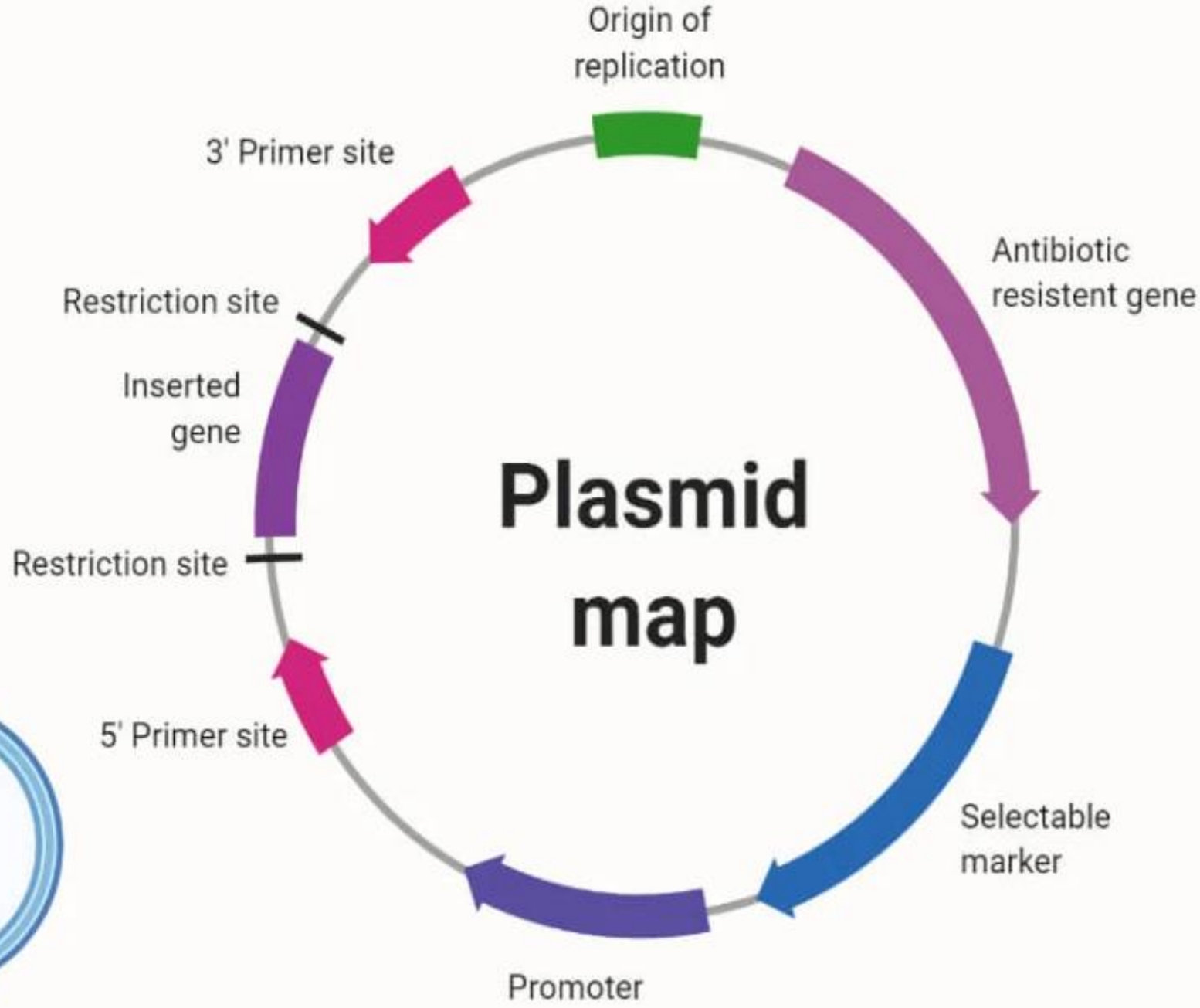
- **Origin of replication (OR):** Refers to a specific location in the strand where the replication process begins. In plasmids, this region is A=T rich region as it is easier to separate the strands during replication.
- **Selectable marker site:** A region consists of Antibiotic resistance genes which are useful in the identification and selection of bacteria that contain plasmids.
- **Promoter and terminator regions:** are regions where the transcriptional machinery is started and ended, respectively.
- **Primer binding site:** A short sequence where primers can anneal on the plasmid, which is useful in DNA amplification and DNA sequencing.
- **Multiple cloning sites (MCS)** This site contains various sequences where the restriction enzymes can bind and cleave the double-stranded structure. Which is used for cloning

Origen of the plasmid

- Wild type plasmid: plasmid already exist
- Synthetic plasmid: plasmid made in a laboratory for different uses

Plasmid




Origin of replication
3' Primer site
Antibiotic resistent gene
Restriction site
Inserted gene
Plasmid map
Restriction site
5' Primer site
Selectable marker
Promoter

- 2. The size of the plasmid varies from 2 kb to 200 kb.
- 3. It is the extrachromosomal element of the cell which is not required for the growth and development of the cell.
- 4. Most of the plasmids contain the TRA gene, which is the transferred gene and is essential in transferring the plasmid from one cell to another.

Transfer of Plasmid

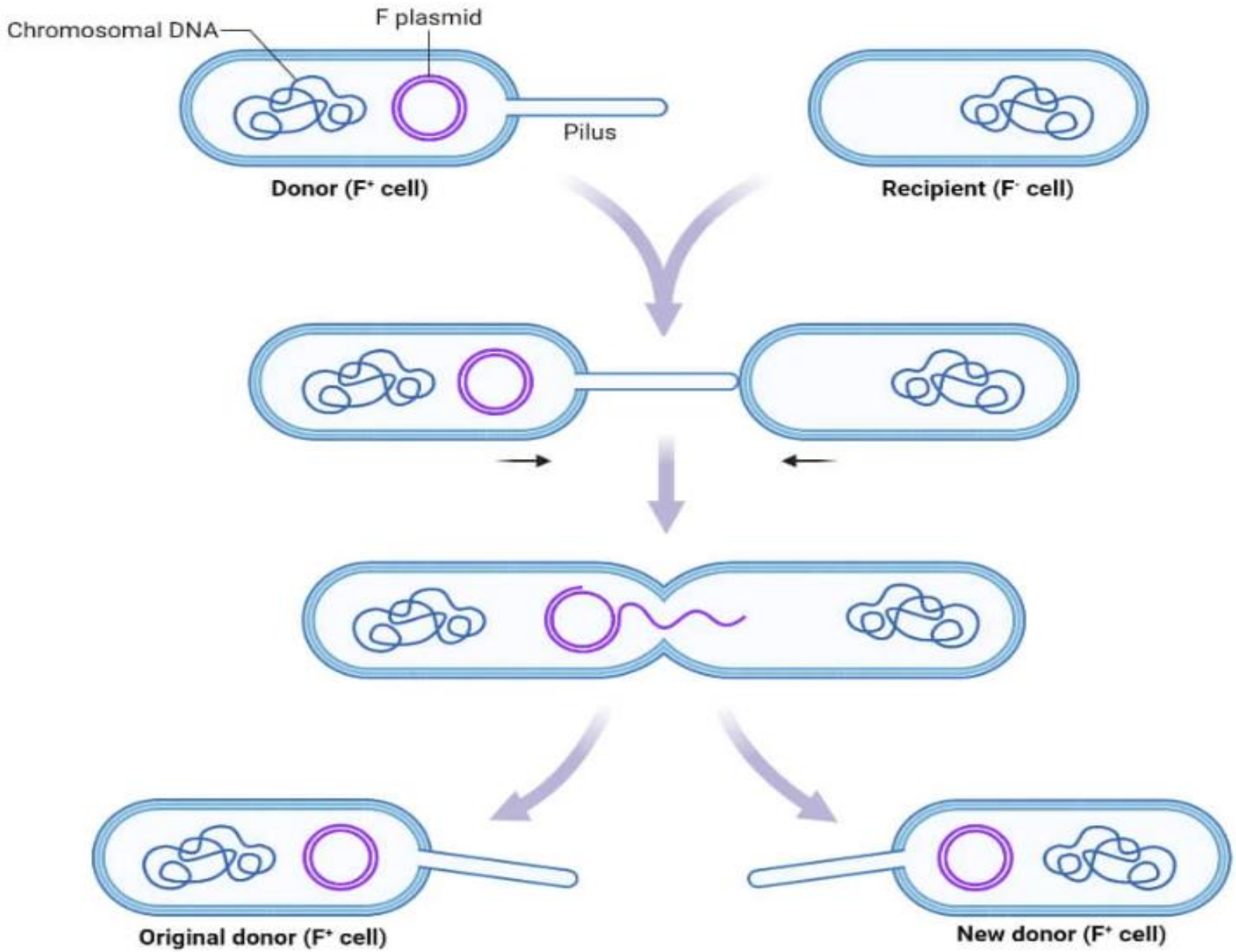
1. Naturally

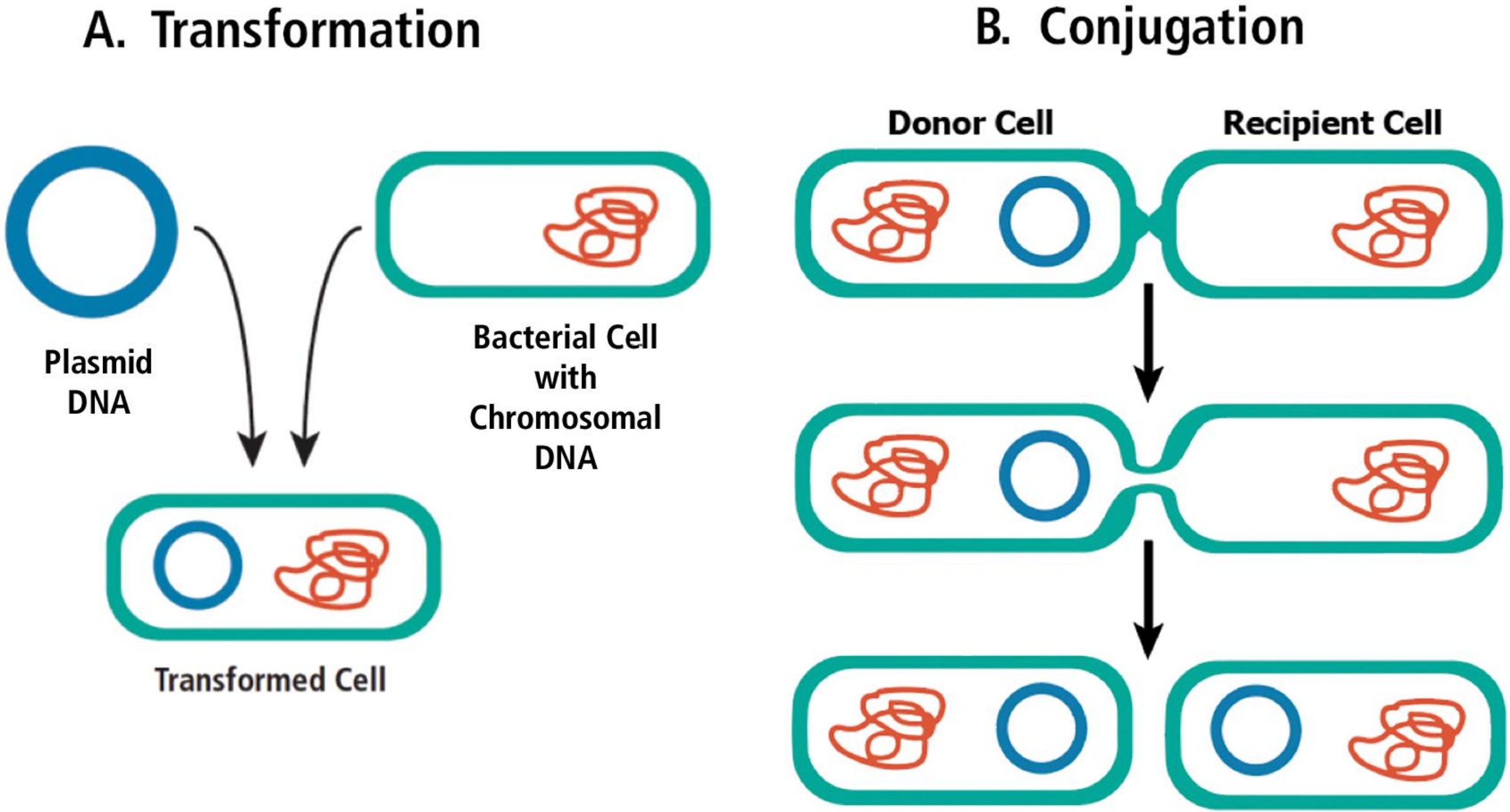
- Plasmids are transferred by the process of Conjugation:
- The process of conjugation involves two cells: a donor cell and a recipient cell.
- The donor cells form a conjugation bridge now as pilus and attach to the recipient cell.
- One copy of the plasmid is transferred from the donor to the recipient cell.

2. Artificially (plasmid transformation)

- Plasmids can be inserted into bacterial cells by using laboratory methods such as heat or electric shock competent cell transformation

Conjugation




A. Transformation
Plasmid DNA
Bacterial Cell with Chromosomal DNA
Transformed Cell
B. Conjugation
Donor Cell
Recipient Cell

Plasmid Examples

S.N.	Plasmids	Organism
1.	pBR322	*E. coli*
2.	pUC19	*E. coli*
3.	ColE1	*E. coli*
4.	RP4	*Pseudomonas*
5.	TOL	*Pseudomonas putida*

Functions Encoded by Plasmids

- Plasmids can encode a few or hundreds of different proteins, resulting in vast differences in their sizes. However, as mentioned above, plasmids rarely encode gene products that are always essential for growth, such as RNA polymerase, ribosomal subunits, or enzymes of the tricarboxylic acid cycle. Instead, plasmid genes usually give bacteria a selective advantage under only some conditions.