Bacterial Chromosomes

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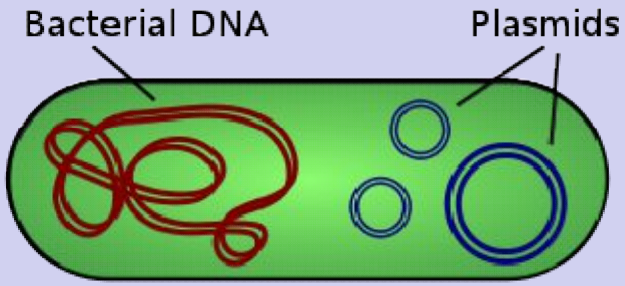
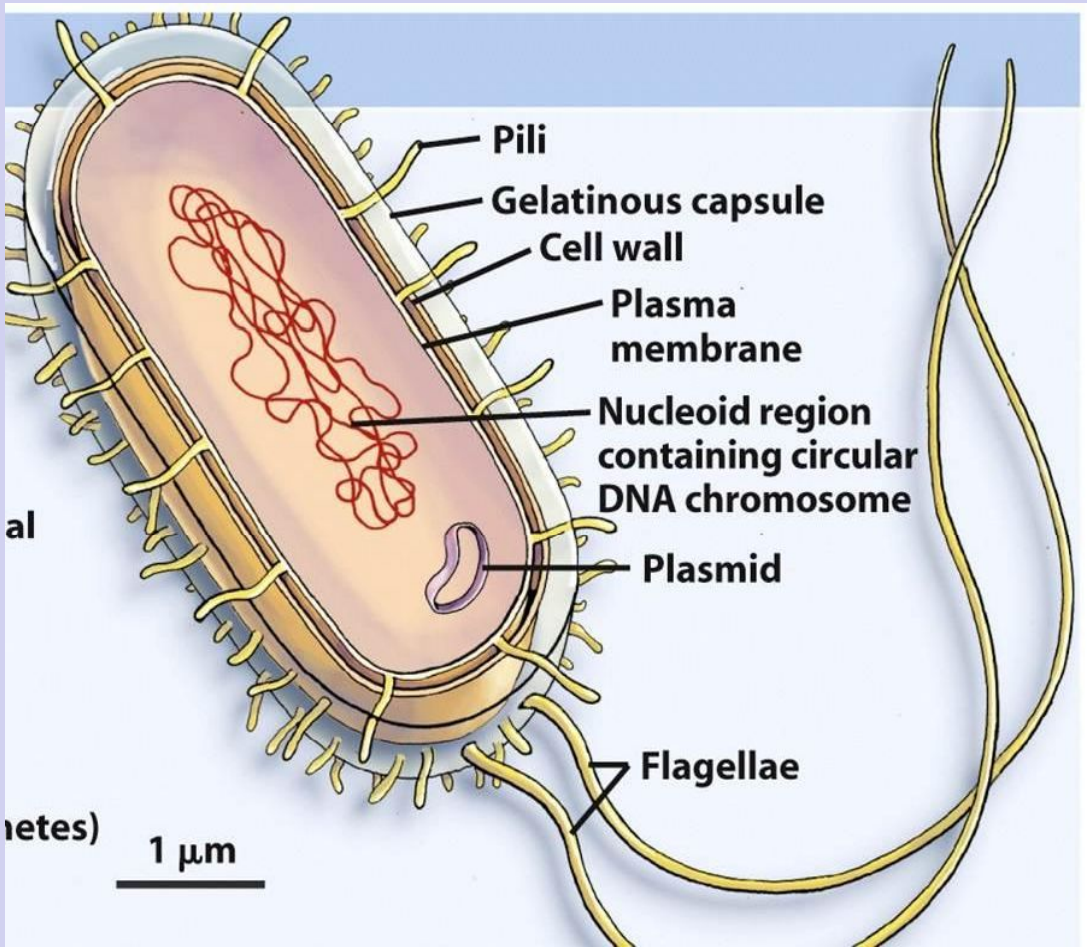




Prokaryotes

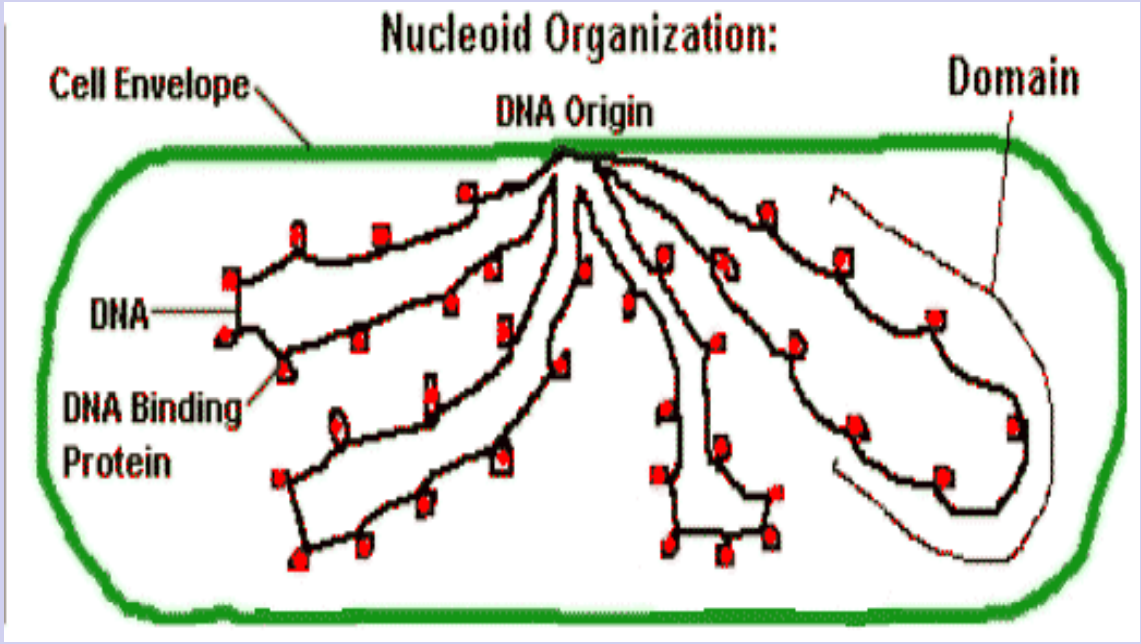
- **The genomes of prokaryotes are having single chromosomes**
- **This single chromosome bears little morphological resemblance to eukaryotic chromosomes.**
- Bacterial chromosomes are generally ~1000 times longer than the cells in which they reside
- The bacterial chromosome is found in a region called the nucleoid
- The nucleoid is not membrane-bounded -So the DNA is in direct contact with the cytoplasm
- Bacteria may have 1 - 4 identical copies of the same chromosome - The number depends on the species and growth conditions

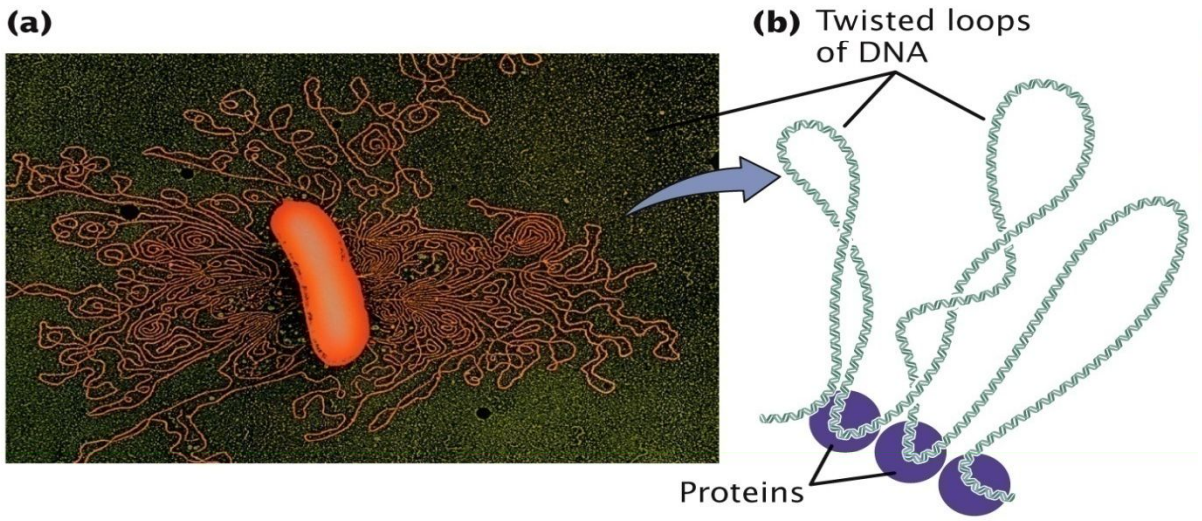
Conti…

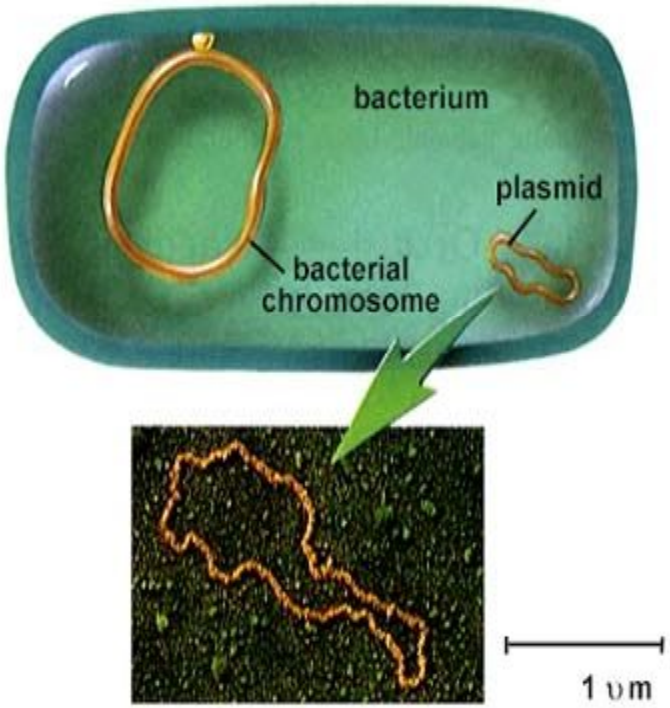
- Most of the Prokaryotic cells including bacteria are having circular DNA that is a few million nucleotides in length
  - *Escherichia coli* → ~ 4.6 million base pairs
  - *Haemophilus influenzae* → ~ 1.8 million base pairs
- **Usually the entire genome is a single circle, but often there are extra circles called plasmids.**
- **The DNA is packaged by DNA-binding proteins.**
- **Single, circular DNA molecule located in the nucleoid region of cell**

Some examples of bacterial genome organization

Bacteria	Chromosome(s)	Plasmid(s)
Agrobacterium tumefaciens	one linear (2.1 Mb) + one circular (3.0 Mb)	two circular (450 + 200 Kb)
Bacillus subtilis	one circular (4.2 Mb)	
Bacillus thuringiensis	one circular (5.7 Mb)	six (each >50 Kb)
Borrella	one linear (0.91 Mb)	multiple circular + linear (5-200 Kb)
Bradyrhizobium japonicum	one circular (8.7 Mb)	
Brucella melitensis	two circular (2.1 + 1.2 Mb)	
Brucella suis biovars 1, 2, 4	two circular (1.0 + 2.0 Mb)	
Brucella suis biovar 3	one circular (3.1 Mb)	
Buchnera sp. strain APS	one circular (640 Kb)	two circular (< 7.8 Kb each)
Deinococcus radiodurans	two circular (2.6 + 0.4 Mb)	two circular (177 + 45 Kb)
Escherichia coli K-12	one circular (4.6 Mb)	
Leptospira interrogans	two circular (4.7 + 0.35 Mb)	
Paracoccus denitrificans	three circular (2.0 + 1.1 + 0.64 Mb)	
Pseudomonas aeruginosa	single circular (6.3 Mb)	
Rhizobacterium meliloti	two circular (3.4 + 1.7 Mb	one circular megaplasmid (1,400 Kb)
Rhodobacter sphaeroides	two circular (3.0 + 0.3 Mb)	
Ureaplasma urealyticum	one circular (0.75 Mb)	
Vibrio cholerae	two circular (2.9 + 1.1 Mb)	
Vibrio parahaemolyticus	two circular (3.2 + 1.9 Mb)	
Xylella fastidiosa	one circular (2.7 Mb)	two circular (51 + 1.3 Kb)







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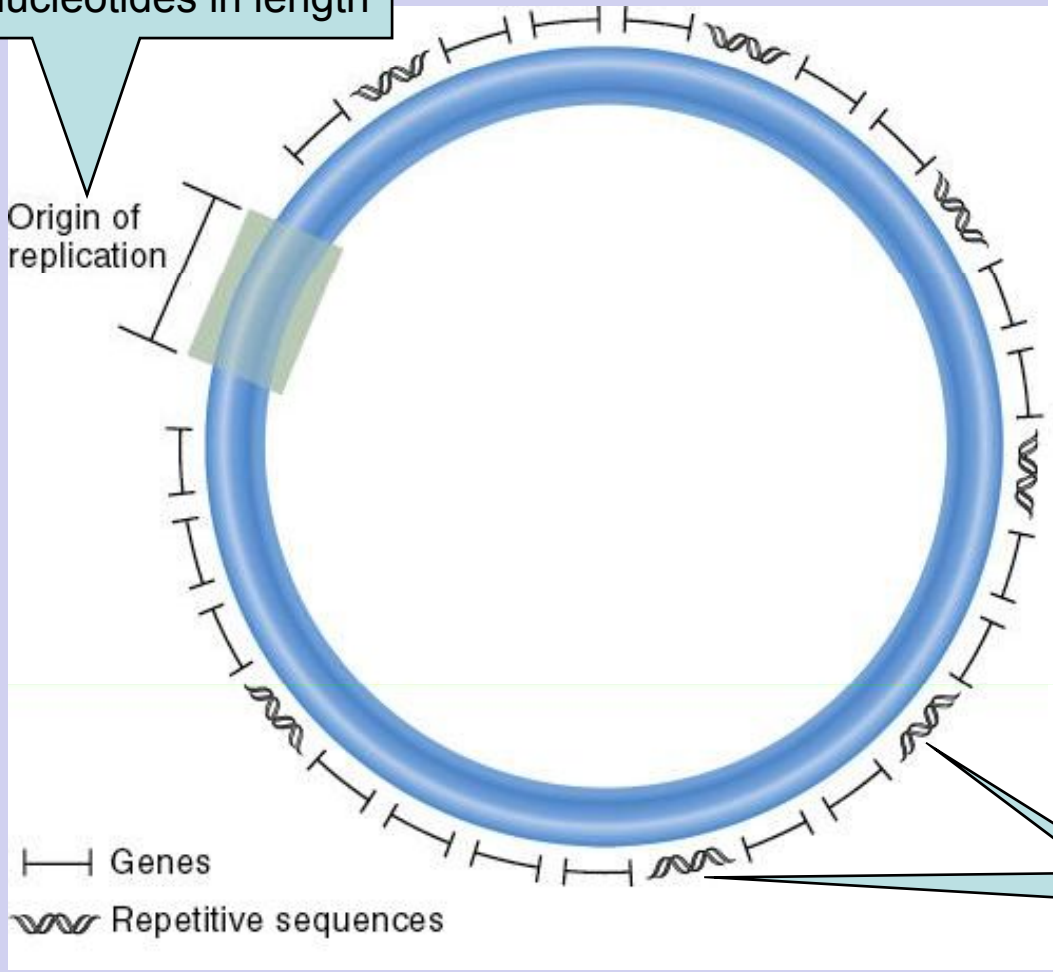
A few hundred nucleotides in length



Key features:

- Most, but not all, bacterial species contain circular chromosomal DNA.
- A typical chromosome is a few million base pairs in length.
- Most bacterial species contain a single type of chromosome, but it may be present in multiple copies.
- Several thousand different genes are interspersed throughout the chromosome.
- One origin of replication is required to initiate DNA replication.
- Short repetitive sequences may be interspersed throughout the chromosome.

These play roles in DNA folding, DNA replication, and gene expression

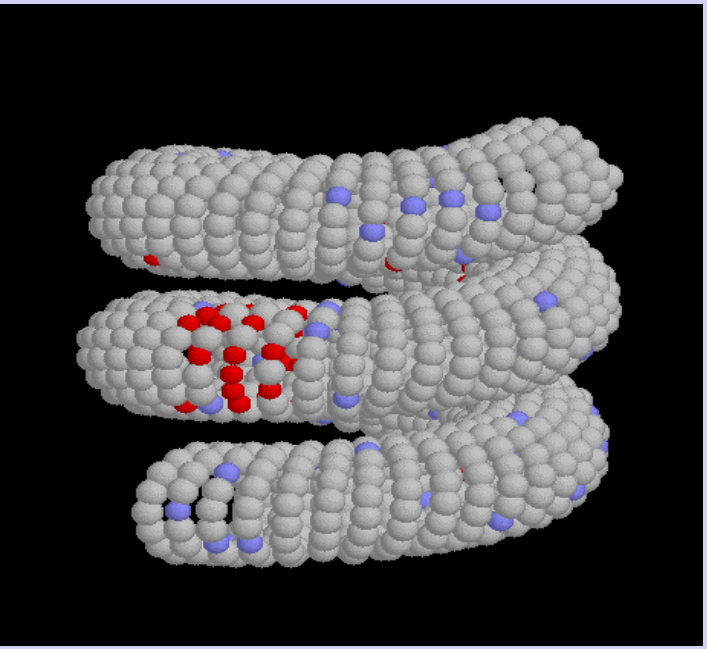
- The number .of gene may be as high as 150 in some larger bacteriophage genome.
- In ***E.coli***, about **3000 to 4000 genes** are organized into its one circular chromosome.
- In contrast to the linear chromosomes found in eukaryotic cells, the strains of bacteria initially studied were found to have single, covalently closed, circular chromosomes.
- The circularity of the bacterial chromosome was elegantly demonstrated using electron microscopy in both G+ (*Escherichia coli*) and G- (*Bacillus subtilis*).
- Bacterial plasmids were also shown to be circular.

The chromosome exists as a highly folded and coiled structure dispersed throughout the cell.

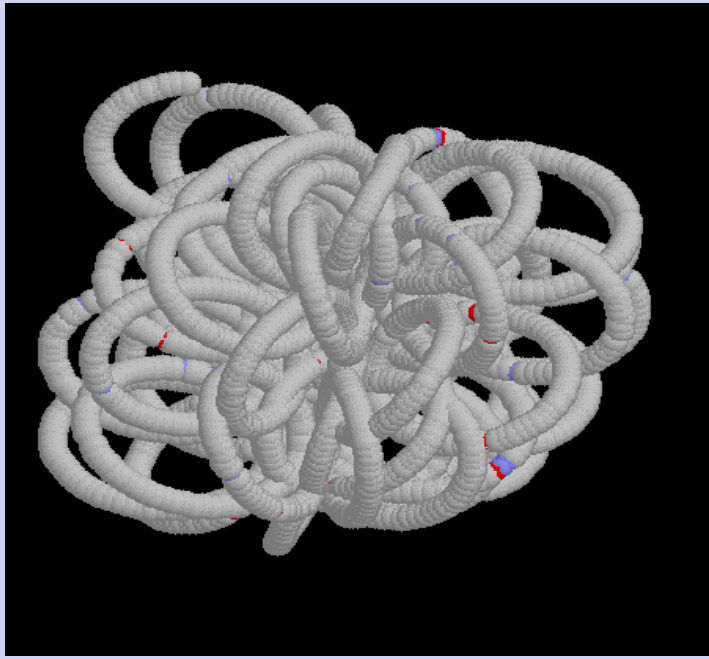
The folded nature of chromosome is due to the incorporation of RNA with DNA.

There are about 50 loops in the chromosome of *E. coli*.

These loops are highly twisted or supercoiled structure with about four million nucleotide pairs. Its molecular weight is about 2.8 X109

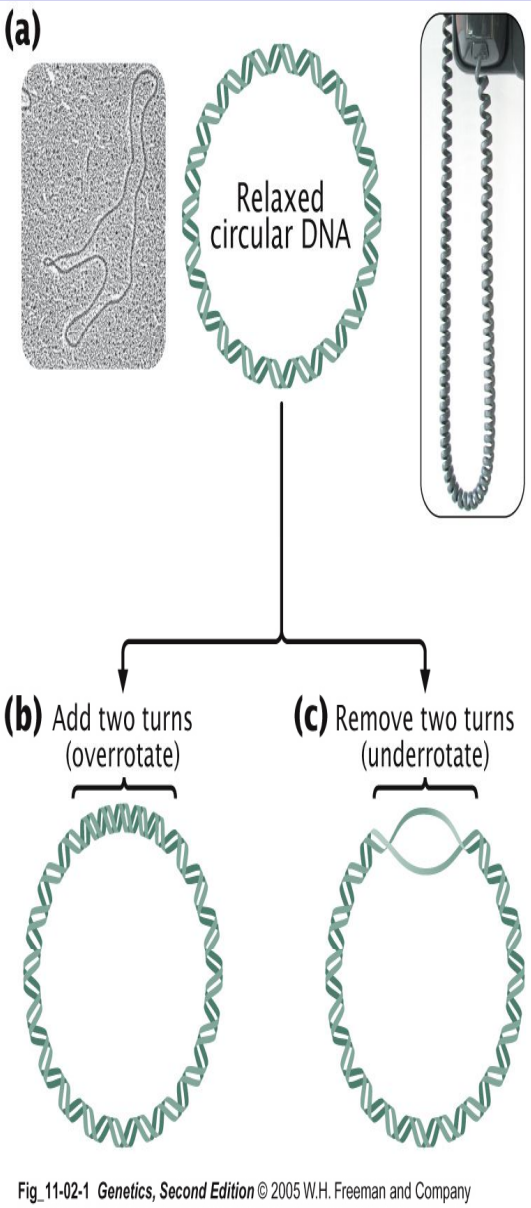


Simple supercoil



Complex supercoil

Supercoiling



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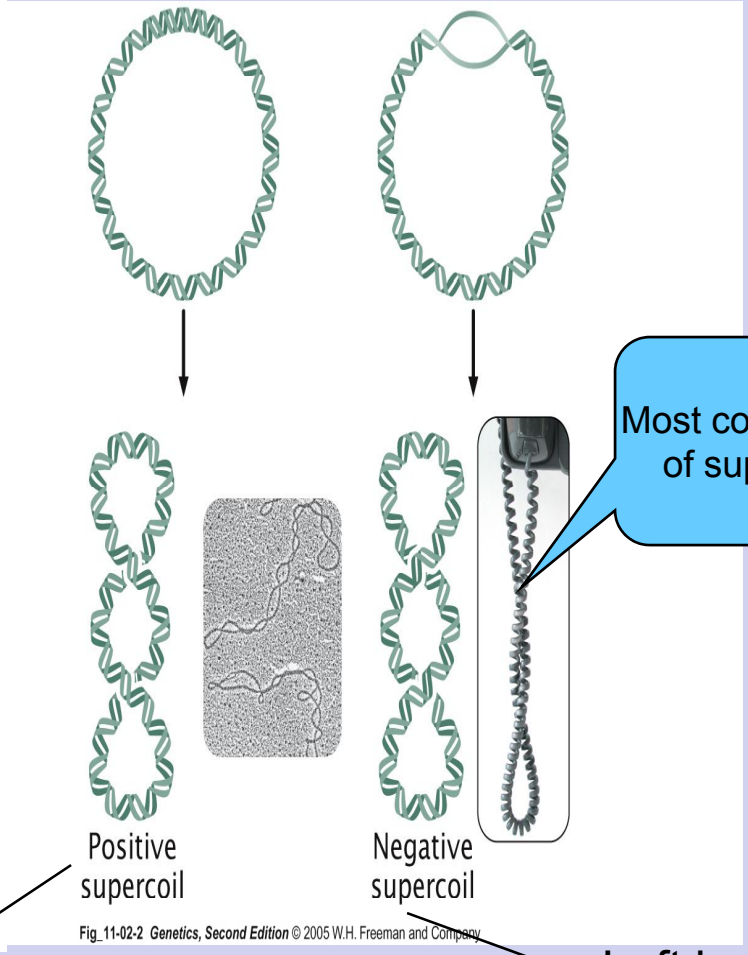

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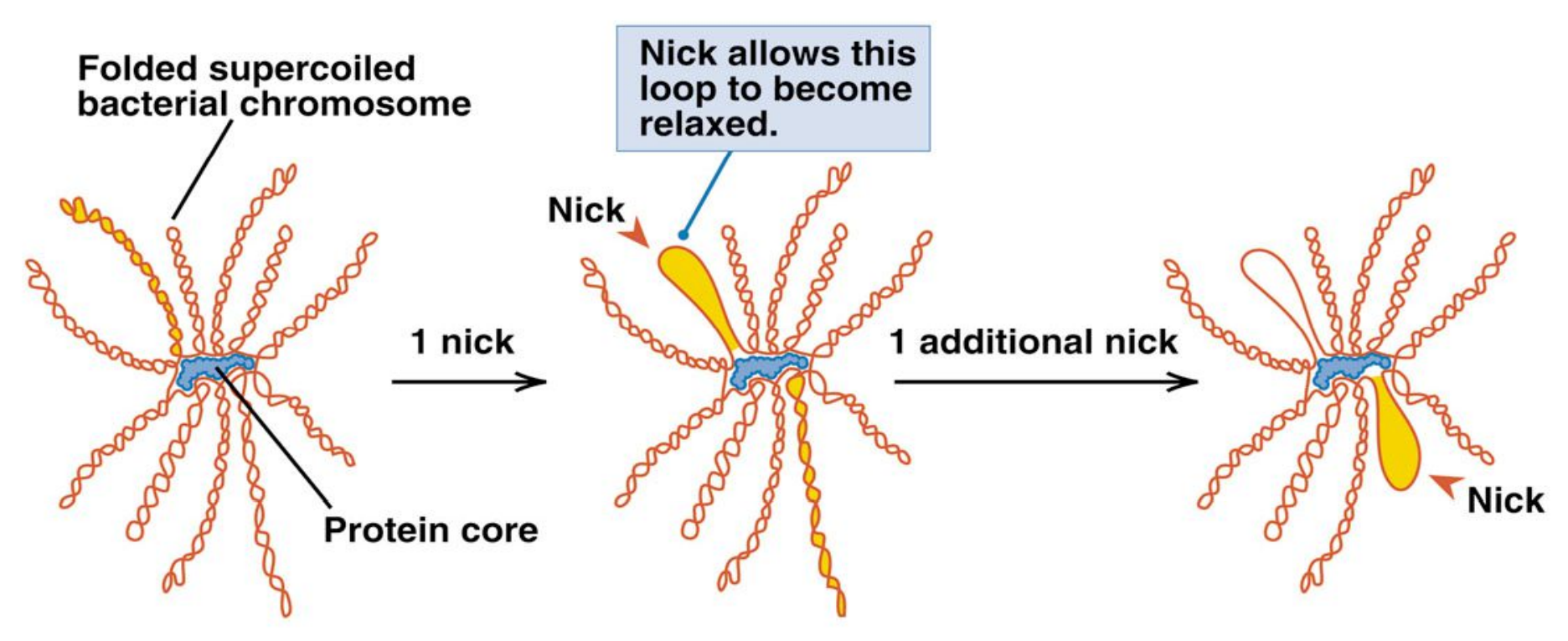
Right handed

Left handed



There are many supercoiled loops (~100 in *E. coli*) attached to a central core protein. Each loop can be independently relaxed or condensed.

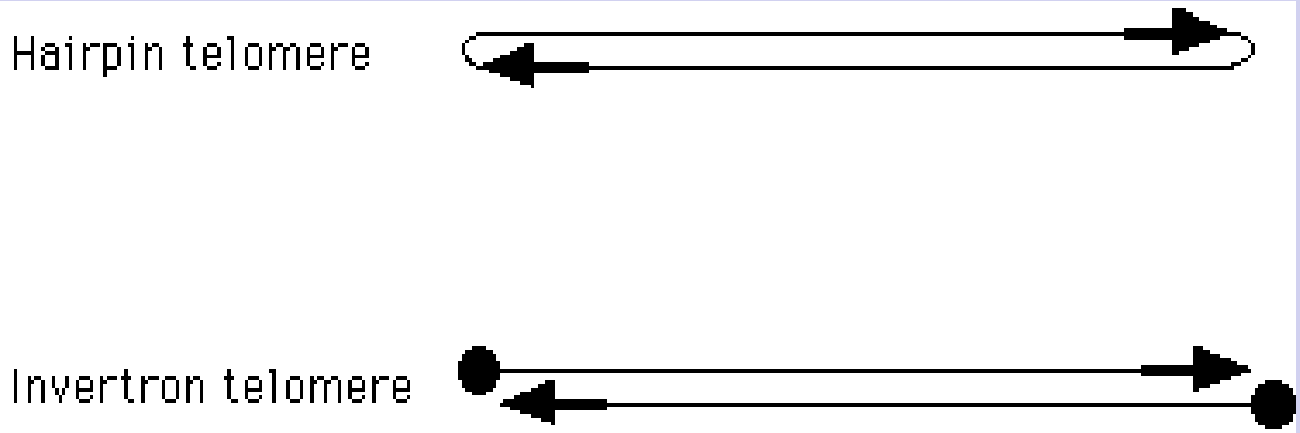
Topoisomerase enzyme – (Type I and II) that introduce or remove supercoiling.

Conti…

- Not all bacteria have a single circular chromosome:
  1. some bacteria have multiple circular chromosomes, and
  2. many bacteria have linear chromosomes and linear plasmids.
- The circular nature of the chromosome is proved using Pulsed field gel electrophoresis (PFGE) .
- Eg: *Rhodobacter sphaeroides* has two large circular chromosomes.
- One of the chromosomes is 3.0 Mb and the other is 0.9 Mb.
- Genes encoding rRNAs and tRNAs required for translation, and metabolic enzymes are distributed between the two chromosomes.

Conti…

- Multiple chromosomes have also been found in many other bacteria, including *Agrobacterium tumefaciens*, *Vibrio cholerae*
- There are examples of linear DNA molecules in bacteria that are protected by both types of telomeres: palindromic hairpin loops are protected by the lack of free double-stranded ends, and invertron telomeres are protected by proteins that bind to the 5'-ends.
- Both of these mechanisms are also used by some phage, eukaryotic viruses, and eukaryotic plasmids

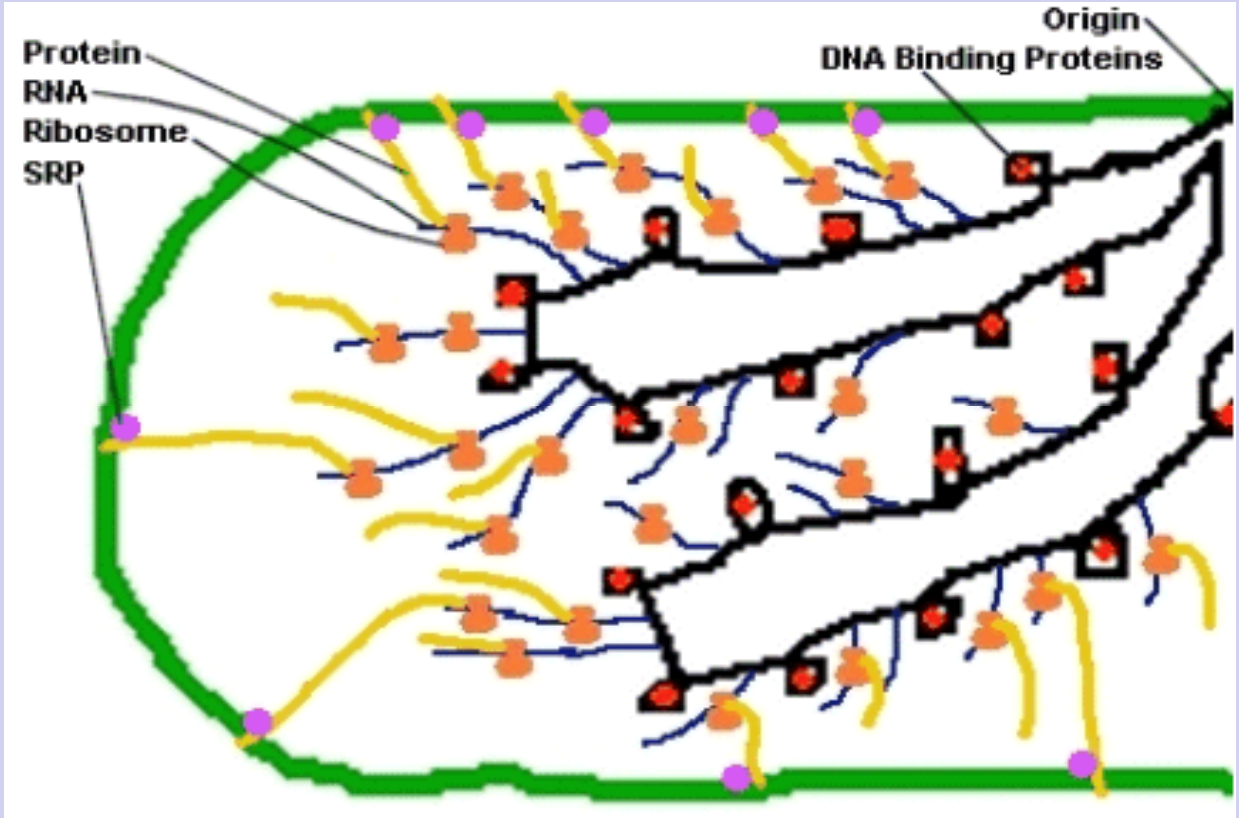


Mycoplasma pneumoniae

- It is nearly a minimal cell with a genome that is 816 kbp long and only 688 genes.
- It has limited metabolism, no known regulation, and very few DNA binding proteins
- The bacterial DNA is packaged in loops back and forth.
- The bundled DNA is called the **nucleoid**.
- It concentrates the DNA in part of the cell, but it is not separated by a nuclear membrane (as in eukaryotes.)
- The DNA does form loops back and forth to a **protein core, attached to the cell wall.**

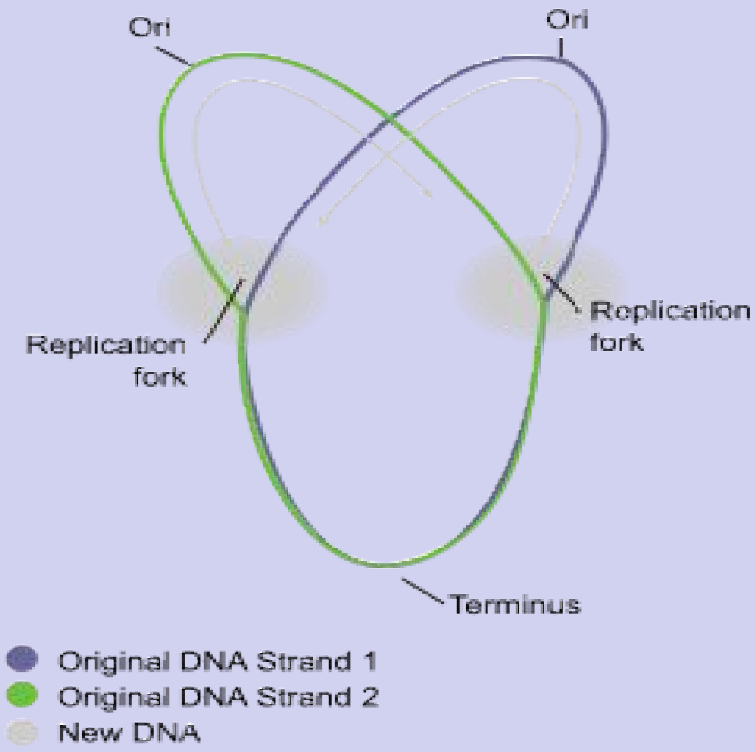
Conti..

- The DNA is accessible to enzymes that make RNA and protein.
- In the bacterial cell, the DNA gets transcribed to RNA, and the RNA gets translated to protein before it is even completed.



Circular bacterial chromosome

- **Circular bacterial chromosome** are the bacterial chromosomes contained in a circular DNA molecule.
- Unlike the linear DNA of vertebrates, typical bacterial chromosomes contain circular DNA.
- Most bacterial chromosomes contain a circular DNA molecule - there are no free ends to the DNA.
- Free ends would otherwise create significant challenges to cells with respect to DNA replication and stability.
- Cells that do contain chromosomes with DNA ends, or telomeres (most eukaryotes), have acquired elaborate mechanisms to overcome these challenges.
- However, a circular chromosome can provide other challenges for cells
- After replication, the two progeny circular chromosomes can sometimes remain interlinked or tangled, and they must be resolved so that each cell inherits one complete copy of the chromosome during cell division.



A circular bacterial chromosome, showing DNA replication proceeding bidirectionally, with two replication forks generated at the "origin".
Each half of the chromosome replicated by one replication fork is called a "replichore". (Graphic computer art by Daniel Yuen)

Replication of a circular bacterial chromosome

Bacterial chromosome replication is best understood in the well-studied bacteria *Escherichia coli* and *Bacillus subtilis*.

Chromosome replication proceeds in three major stages:

1. initiation,
2. elongation and
3. termination.

Initiation

- The *E. coli* bacterial replication origin, called *oriC* consists of DNA sequence which recognised by the DnaA protein
- DnaA protein plays a crucial role in the initiation of chromosomal DNA replication
- This region also contains four “GATC” sequences

Elongation

- When the replication fork moves around the circle, a structure shaped like the Greek letter theta Θ is formed.
- Replication occurs in a bidirectional manner
- DNA polymerase III holoenzyme, DNA polymerase I, exonuclease, DNA ligase are essential for this activity

Termination

- Termination is the process of fusion of replication forks and disassembly of the resplisomes to yield two separate and complete DNA molecules.
- It occurs in the terminus region, approximately opposite oriC on the chromosome
- The terminus region contains several DNA replication terminator sites, or "Ter" sites.