### **Bacteria**

- Ehrenberg (1838) first of all coined the word Bacteria
- Very complex behavior
- Most extensive metabolic diversity
- **Dialister pneumosintes** in nasopharunx of man in influenza 1.5 μ

#### Size

- 3 to 5 μm
- A huge bacterium, *Epulopscium fishelsoni* which was discovered in the intestine of the brown surgeon fish, is as large as **600** μm and as wide as **80** μm.

#### **Robert Hooke**

- In 1665, Hooke built a microscope and examined thin slices of cork cut with a penknife.
- He saw a network of tiny boxlike compartments that reminded him of a honeycomb and called these little compartments cells, from the Latin word cellula, meaning "little room."

### **Antonie van Leeuwenhoek 1683**

- First to observe living cells, including blood cells, sperm cells, bacteria, and single celled organisms (algae and protozoa) found in pond water.
- 'Animalcules'
- Father of bacteriology

### **Robert Brown (1831)**

- Observed presence of a small sphere within the cells of orchid roots.
- English botanist Robert Brown found that every plant cell he looked at contained a rounded structure, which he called a nucleus, a term derived from the Latin word for "kernel."

Alfonso Corti (1772) observed living substances in the cells.

**Hugo von Mohl** (1838-1846) and **Johannes Purkinje** (1839) called the jelly-like substance **Protoplast**.

### Cell Theory

- In 1838, Malthias Schleiden, a German botanist, examined a large number of plants and observed that all plants are composed of different kinds of cells which form the tissues of the plant.
- At about the same time, Theodore Schwann (1839), a British Zoologist, studied different types of animal cells and reported that cells had a thin outer layer which is today known as the plasma membrane'. He also concluded, based on his studies on plant tissues, that the presence of cell wall is a unique character of the plant cells.
- On the basis of this, Schwann proposed the hypothesis that the bodies of animals and plants are composed of cells and products of cells. Schleiden and Schwann together formulated the cell theory

As originally postulated by Schwann in 1839, the cell theory had two basic principles:

- **1.** All organisms consist of one or more cells.
- 2. The cell is the basic unit of structure for all organisms.

### **Rudolf Virchow (1855)**

- a German physiologist
- First explained that cells divided and new cells are formed from pre-existing cells (Omnis cellula-e cellula).
- He modified the hypothesis of Schleiden and Schwann to give the cell theory a final shape.

Less than 20 years later, a third principle was added.

## **3.** All cells arise only from preexisting cells.

# **Shape**

- Bacillus -
  - The most common shape is rod-like.
  - The bacilli differ considerably in their shape of rod as it may be flat or round or cigarshaped.
- Coccus or Spherical
- Vibrios :
  - A few rod-shaped bacteria may be slightly curved, or comma-shaped.
- Spirilla
  - Many bacteria are shaped like a long, twisted spiral; these may resemble a comma.
  - Other bacteria may be highly coiled and resemble a cork screw (Spirochetes)

### **Glycocalyx / Sugarcoat:**

- Glycocalyx is the outermost layer comprising a **coating of macromolecules**, which protects cells and also helps in adhesion.
- This layer differs in thickness and chemical composition in different bacteria.
- Capsules
  - thick and tough
  - capsule is closely associated with cells and does not wash off easily
  - responsible for giving gummy and sticky character to the cell.
- Slime layers
  - slime layer is more diffuse and is easily washed away.
  - loose sheath which protects the cells from loss of water and nutrients

## Cell Wall

- provides a strong structural support
- prevent the bacterium from bursting or collapsing
- Made up of special macromolecule called peptidoglycan (murein or mucopeptide) a heteropolymer.
- It is composed of repeating framework of long glycan strands (N-acetyl muramic acid and N-acetyl glucosamine)
- Lysozyme, a naturally occurring enzyme in **saliva and tears**, also provides defense against certain bacteria by hydrolysing the peptidoglycan hydrolyzes the NAM-NAG bond
- The outer face of the outer membrane of Gram negative bacteria contains **lipopolysaccharides**, a part of which is integrated into the membrane lipids.

• In Mycobacterium and Nocardia, the wall is that of Gram-positive type but a part of their cell wall is made up of a very long chain of the fatty acid, called mycoic acid.

### The Gram-negative's cell wall

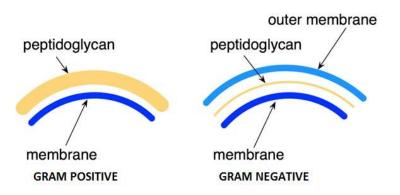
- Gram-negative bacteria have two membranes—an outer membrane and an inner (cytoplasmic) membrane.
- Their peptidoglycan layer is located between the two membranes in the periplasmic space.
- The outer membrane of Gram-negative bacteria invariably contains a unique component, lipopolysaccharide (LPS) in addition to proteins and phospholipids.

### Teichoic acid (wall acid ):

- only in gram-positive species
- stabilises the cell wall and makes it stronger

### **Porins**

- Proteins that form pores in the outer membrane
- Allow passage of most small hydrophilic molecules.



Cell envelope in bacteria

Property	Gram-positive	Gram-negative
Thickness of wall	20-80 nm	8-12 nm
Wall	smooth	Wavy and comes in contact with PM inly at some loci
Peptidoglycan content	70- 80%	10-20%
Teichoic acid in wall	+	_
Endospore	Common	Less common

#### Periplasm

• Space in between plasma membranes and cell wall

#### **Plasma Membrane**

- Similar structurally to that of the eukaryotes.
- Lack sterols such as cholesterol except PPLO
- It also serves as a selective permeability barrier allowing particular ions and molecules to pass either into or out of the cell while preventing others

- Additionally, the bacterial plasma membrane is the location of critical metabolic processes like respiration, photosynthesis, synthesis of lipids and cell wall constituents
- The plasma membrane can be invaginated to form internal membrane structures.

#### Mesosomes

- A special membranous structure is the mesosome which is formed by the extensions of plasma membrane into the cell.
- These extensions are in the form of vesicles, tubules and lamellae.
- These are commonly seen in **Gram-positive bacteria**.

### They help in

- cell wall formation,
- DNA replication and distribution to daughter cells.
- help in respiration, secretion processes
- increase the surface area of the plasma membrane and enzymatic content.

### **Chromatophores**

- In some prokaryotes like cyanobacteria, there are other membranous extensions into the cytoplasm called **chromatophores** which contain pigments
- Chromatophores are internal membrane systems in prokaryotic cells that may become extensive and complex in photosynthetic forms like cyanobacteria and purple-bacteria.

### **Ribosomes:**

- The cytoplasmic matrix of the prokaryotic cell is often packed with ribosomes.
- Ribosomes are also associated with plasma membrane of the cell.
- They are about 15 nm by 20 nm in size
- The ribosome is the site of protein synthesis.

# **Inclusion bodies: Inclusions or Storage Granules**

- Reserve material in prokaryotic cells are stored in the cytoplasm in the form of inclusion bodies. or storage granules.
- Inclusion bodies are randomly scattered in the cytoplasmic matrix.
- Dense aggregates of specific chemical compounds

### 1. Phosphate granules Polymetaphosphate / Volutin Granules:

Phosphate polymers and function as a storage reservoir for phosphate.

### 2. Cyanophycean granules Polyglucan granules

Starch

### 3. Glycogen granules

### 4. Poly-β-hydroxybutyrate granule

Carbon and energy storage product

### 5.Sulfur (S<sub>0</sub>) granules

• Some bacteria also store sulphur temporarily as sulphur granules.

• These may be formed when bacteria use hydrogen sulphide as electron donor during photosynthesis.

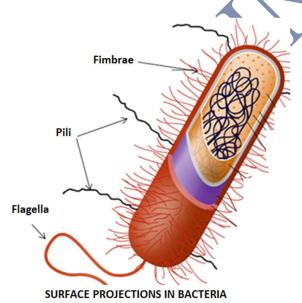
#### **Gas vacuoles**

- Gas vacuoles are found in blue green and purple and green photosynthetic bacteria and a few other aquatic forms that are free-floating (planktonic).
- These are basically aggregates of a number of small, hollow cylindrical gas vesicles.
- Gas vesicles are not permeable to water but are permeable to atmospheric gases.
- Because of gas vacuoles, bacteria keep floating on or near the surface of water.

### Carboxysomes

- Found in many autotrophic bacteria, Cyanobacteria, Thiobacillus Nitrobacteria
- Contain enzymes like Rubisco and carbonic anhydrase
- Involved in carbon fixation

# **Surface Projections**



## Flagella

Bacterial flagellum is composed of three parts – filament, hook and basal body.

### The filament

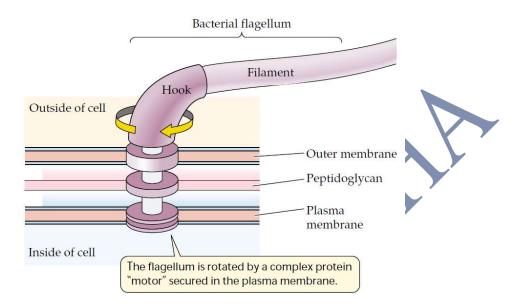
- longest portion
- hollow rigid cylindrical structure made of the protein called flagellin.
- It is inserted into a curved tubular hook which is anchored to the cell by basal body.

The hook is quite different from the filament and is made up of different protein subunits.

### The basal body

- most complex part of the flagellum
- consists of rings connected to a central rod in Gram-negative bacteria.

The filament, hook and basal body are arranged in such a manner that the structure permits the filament to rotate by 360° rather than undulating back and forth like a whip.



### **Distribution of Flagella**

Atrichous - Lactobacillus

Monotrichous - ThiobacillusAmphitrichous - Nitrosomonas

Cephalotrichous - Pseudomonas

• Lophotrichous - Spirillum

• **Peritrichous** - E.coli, Clostridium

## Pili and Fimbriae

These two terms have been used interchangeably to indicate any bacterial surface appendage not involved with motility.

## Pili

- Elongated tubular structures
- Longer than fimbriae and there are only a few per cell
- Made up of a special protein called pilin

### **Two basic functions**

- Gene transfer (Sex pili) in mating
- Attachment to surfaces

### **Fimbriae**

- Small bristle like fibres sprouting out of the cell.
- Protein of fimbriae has been called fimbrilin
- Some type of fimbriae are known to **attach bacteria** to solid surfaces such as rocks in streams and to host tissues.

• These are also responsible for mutual clinging of cells

### **Nucleoid:**

- DNA is not scattered throughout the cell.
- DNA (being negatively charged) is held with some proteins (that have positive charges) in a region termed as 'nucleoid'.
- The genetic material in prokaryotes is composed of a single circular DNA, and is packed in a nucleoid.
- In some cases RNA and a small amount of protein are present.
- The nucleoid is usually associated with mesosomes.
- DNA tighly coiled circular single DNA molecule, called the bacterial chromosome or Nucleoid or Incipient nucleus which is attached to Mesosome

#### **Plasmid**

- This extrachromosomal DNA fragment
- Self replicating and stably inherited
- The plasmid DNA confers certain unique phenotypic characters to such bacteria. One such character is resistance to antibiotics
- **Episomes** are the plasmid which get integrated into the bacterial chromosomes.

## **Types**

## **Fertility F- plasmids**

They are involved in sex determination

### Resistance (R) plasmids

• contain genes that can build a resistance against antibiotics or poisons.

#### Col plasmids

• contain genes that code for **bacteriocins**, proteins that can kill other bacteria.

### Virulence plasmids

- turn the bacterium nto a pathogen.
- Ti plasmids

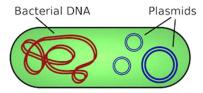


Fig.: Plasmid

# **Gram's staining**

 Bacteria can be classified into two groups on the basis of the differences in the cell envelopes and the manner in which they respond to the staining procedure developed by Gram

- Method is named after the Danish bacteriologist Hans Christian Gram in 1882
- It is a **differential staining method** of differentiating bacterial species into two large groups (Gram-positive and Gram-negative) based on the **chemical and physical properties of their cell walls.**

# **Gram staining consists of four components:**

- Primary stain (weakly alkaline solution of Crystal violet, methyl violet or Gentian violet)
- Mordant Gram's lodine
- Decolouriser -(ethyl alcohol, acetone
- Counterstain (safranin)

### **Steps**

- The primary stain renders all the bacteria uniformly violet.
- The smear is treated with a few drops of Gram's lodine and allowed to act for a minute
- This is followed by washing with water and then with alcohol or acetone
- The process of decolorisation is fairly quick seconds
- Those bacteria that hold on to primary dyeiodine complex and remain violet are called Gram positive and those which get decolorized and subsequently take up counterstain (pink/red) are called Gram negative.

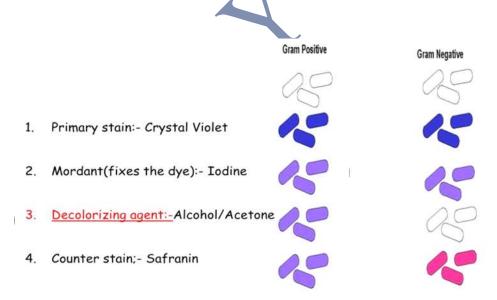


Fig. : Gram staining

# Reproduction in bacteria

### **Vegetative reproduction:**

- Binary fission
- Endospores

### **Bacterial Cell Division:**

- Bacteria reproduce by binary fission.
- When conditions are favourable such as the right temperature and nutrients are available, some bacteria like *Escherichia coli* can divide **every 20 minutes.**

# **Endospores**

- Endospore formation occurs in bacteria to tide over unfavourable environmental conditions.
- Dormant, tough, and non-reproductive structure, refractile
- Usually occurs in Gram-positive bacteria- Bacillus and Clostridium.
- Resistant to ultraviolet radiation, desiccation, high temperature, extreme freezing and chemical <u>disinfectants</u> acids, bases, alcohol, chloroform boiling at 100°C for hours

### Structure:

### Core / protoplast

- consists of the cytoplasm, DNA, ribosomes
- core is dehydrated (10-25 %)
- contain calcium dipicolinate

### **Cortex**

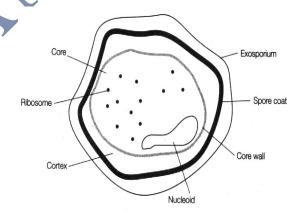
contains large amounts of dipicolinic acid and calcium ions

### Spore coat

containing several protein layers that are impermeable

### Exosporium-

polysaccharide protein membrane



# Sexual reproduction in bacteria

- Genetic recombination) / Parasexuality
- No meiosis
- No syngamy

## **Transfer of Genetic Material in Bacteria:**

Bacteria have mechanisms by which they can 'obtain' **extra DNA**, which creates opportunities for recombination to occur. The three main mechanisms by which bacteria acquire new DNA are

### Transformation:

- Transformation involves acquisition of DNA from the environment
- F Griffith
- Streptococcus pneumoniae

# **Conjugation:**

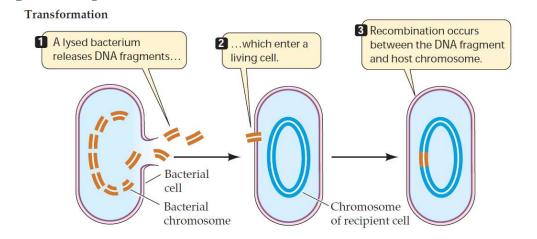
- Conjugation involves acquisition of DNA directly from another bacterium
- Lederberg and Tatum 1946
- E coli

## Transduction:

- Transduction involves acquisition of bacterial DNA via a bacteriophage intermediate
- Zinder and Lederberg 1952
- Salmonella typhimurium

## **Transformation**

• Transformation is the process by which bacteria pick up DNA from their environment.



**Griffith's experiment,** conducted in 1928 by <u>Frederick Griffith</u>, was one of the first experiments suggesting that bacteria are capable of transferring genetic information through a process known as <u>transformation</u>.

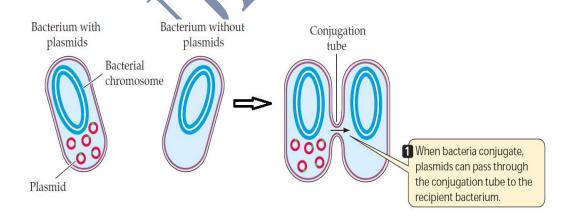
Griffith used two <u>strains</u> of *Pneumococcus* (<u>Streptococcus pneumoniae</u>) bacteria which infect <u>mice</u> – a type III-S (smooth) and type II-R (rough) strain. The III-S strain covers itself with a <u>polysaccharide</u> capsule that protects it from the host's <u>immune system</u>, resulting in the death of the host, while the II-R strain doesn't have that protective capsule and is defeated by the host's immune system.

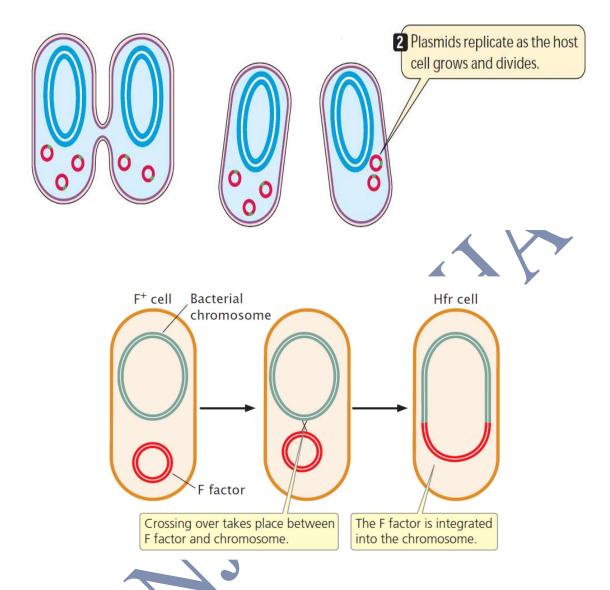
In this experiment, <u>bacteria</u> from the III-S strain were killed by heat, and their remains were added to II-R strain bacteria. While neither alone harmed the mice, the combination was able to kill its host.

Griffith was also able to isolate both live II-R and live III-S strains of pneumococcus from the blood of these dead mice. Griffith concluded that the type II-R had been "transformed" into the lethal III-S strain by a "transforming principle" that was somehow part of the dead III-S strain bacteria. Today, we know that the "transforming principle" Griffith observed was the **DNA** of the III-S strain bacteria.

# Conjugation

- Conjugation is a mating process involving bacteria. It involves transfer of genetic information from one bacterial cell to another, and requires physical contact between the two bacteria involved.
- The contact between the cells is via a protein tube called an **F** or **sex pilus**, which is also the conduit for the transfer of the genetic material.

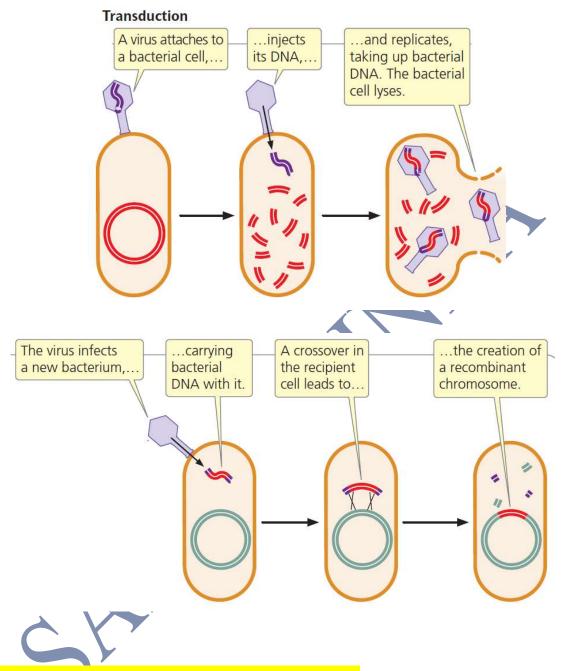




# **Transduction**

Transduction is a type of recombination that involves the exchanging of bacterial DNA through bacteriophages.

Bacteriophages are viruses that infect bacteria.



# Cyanobacteria / BGA blue green algae / oxyphotobacteria

- Anabaena
- Tolypothrix
- Aulosira
- Nostoc
- Microcystis
- Spirulina
- Trichodesmium erythrium causes red sea

- One-celled or colonial or filamentous
- Flagella are absent
- Cell wall similar to Gram negative bacteria.
- PM infoldings mesosome
- Phycobilins are water-soluble pigments- absorb light 470 to 600 nm
- Oldest oxygen producing organisms.
- Cyanophycean starch
- Volutin granules
- Cyanophycin granules
- Gas vacuoles
- Rep by Fission Budding Fragmentation

## **Heterocysts**

- Differentiated cells that specialize in nitrogen fixation.
- Cysts like Rounded structures distributed at regular intervals
- Develop a surface that is impermeable to gasses
- Begin synthesising large amounts of nitrogenase
- Heterocysts maintain photosystem I
- Heterocysts lack Photosystem II
- Heterocysts are the site of nitrogen fixation in filamentous algae.

# Archebacteria: A different type of microbe

- Discovery of the extreme thermophile *Thermus aquaticus* in the hot springs of Yellowstone National Park was made by Tom Brock and Carl Woese.
- Inhabitants of some of the most extreme environments
  - thermal vents in the deep sea
  - hot springs
  - extremely alkaline or acid waters.
  - inside the digestive tracts of cows, termites-- produce methane.
  - in the anoxic muds of marshes

Prokaryotic characters	Eukaryotic characters	Unique characters
Binary fission 70 S ribosomes N2 fixation Circular DNA Plasmids present Gas vacuoles present Reproduction	Transcription Translation Introns in DNA Have histones	Cell Wall  PM  Phospholipids are glycerol ether of fatty acids

## **Cell walls**

- Non cellulosic polysaccharides
- Lack peptidoglycan, but some contain pseudomurein.

Methanogens	Halophiles	Thermoacidophiles
Obligate anaerobe	Aerobic	High temp > 80 c
Chemoautotroph	Heterotroph	Low pH –2
Methanococcus Metahnobacterium	High salt conc.  In highlight intensity they	Hot S springs
	produce	Aerobic / anaerobic
	bacteriorhodopsin in	Auto / heterotrophs
	PM and <b>produce ATP</b> and form bright red colonies and survive until O2 increases  Halobacterium	Thermoplasma Thermoproteus

# **Actinomycetes:**

- Ray fungi / mycelial bacteria / higher bacteria
- Intermediate between bacteria and fungi
- Have mycelium
- Cell walls contain peptidoglycon contain Mycolic acid
- Gram positive bacteria
- Primary decomposers of tough plant tissues
- Source of antibiotics
- Reproduction: by spores fragmentation and conidia

## **Examples:**

- Actinomyces Nocardia Mycobacterium Streptomyces
- Corynbacterium Propionibacterium Bifidibacterium

**Frankia** fixes atmospheric nitrogen forming root nodule by symbiotic association in **non-leguminous plants** like **Casuarina**, **Ginkgo** 

Corynbacterium diptheriae	diptheria	
Corynbacterium tritici	Tundu disease of wheat	
Mycobacterium leprae	Leprosy	
Mycobacterium tuberculosis	TB	
Streptomyces scabies	Potato scab	

# **Mycoplasmas**

- Originally called the pleuropneumonia organism (PPLO).
- Bacteria lacking a rigid cell wall
- They are also called 'Jokers of plant kingdom'
- Can survive without O2
- Smallest known free-living organisms 0.33 micron size
- The cells are bounded by a single triple-layered membrane contains sterols
- Form minute, transparent colonies. fried egg / mulberry colonies
- No flagella
- Insensitive to penicillin and cephalosporins
- Killed by chloramphenicol tetracycline streptomycin erythromycin
- ds linear DNA

#### Diseases

- Primary atypical pneumonia (PAP) Walking pneumonia
- Urethritis
- Stillbirth Infertility
- Cattle pneumonia
- Witches broom of Legumes
- Little leaf disease of Brinjal

## Rickettsiae

- Named after H.T. Ricketts
- Obligate intracellular parasites
- Link between bacteria and virus

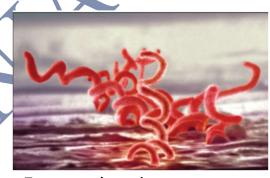
- Occur in the gut of arthropods
- Diseases caused:
  - Rocky Mountain Spotted Fever RMSF
  - Scrub typhus
  - Q-fever
  - Typhus fever
  - Trench fever

# **Chlamydia**

- Obligate intracellular parasites humans, animals and birds
- Although they synthesise most of their metabolic intermediates, they are unable to make their own ATP and thus are energy parasites.
- Chlamydia is called the "silent STD"
- Chlamydia can cause **pelvic inflammatory disease (PID)**, which can lead to sterility and sometimes death.
- Also causes Trachoma Conjunctivitis Urethritis

# **Spirochetes**

- Spirochetes are very thin, flexible, spiral-shaped procaryotes
- Tightly coiled, and so look like miniature springs or telephone cords.
- Borrelia burgdorferi causes Lyme disease
- Leptospira causes Leptospirosis
- Treponema pallidum causes Syphilis



Treponema bacteria

# **Nutrition in bacteria**

- Most bacteria are saprophytes and parasites
- Some are photosynthetic and chemosynthetic

# **Photoautotrophs**

Photosynthetic Eubacteria					
<b>Types</b>	<b>Carbon Source</b>	e <sup>-</sup> Source	<b>Pigment</b>		
Green Sulphur Chlorobium	CO <sub>2</sub>	H <sub>2</sub> S	Bacterioviridin		
Purple Sulphur	CO <sub>2</sub>	Na <sub>2</sub> SO <sub>3</sub>	Bacteriopurpurin		

# **Lithotrophic Types of Metabolism**

• Lithotrophy is the use of an inorganic compound as a source of energy.

# **Sulphur Bacteria:**

- These bacteria utilizes the energy released by the oxidation of sulphur and its compounds.
- Found in environments rich in H₂S, such as volcanic hot springs
- Thiobacillus

## Iron bacteria:

- These bacteria generally **oxidize the ferrous ion** into ferric ion and releases energy.
- Leptothrix, Ferrobacillus.

# Nitrifying bacteria:

- These bacteria utilize the energy released from nitrogen compound.
- CO<sub>2</sub> fixation utilizes RUBP carboxylase and the Calvin Cycle.
- Nitrosomonas Nitrobacter

