FACTORS AFFECTING GROWTH

In addition to their chemical needs, microorganisms are affected by the physical nature of their environment. Environmental factors such as temperature, pH, osmotic pressure and radiation, influence the growth of microorganisms.

1- Temperature

Temperature plays a very important role in the cell cycle. The optimum temperature for enzyme activity in microorganisms is depending on the **ecological site** of the species and the **enzyme's location**. Certain temperatures are lethal to some microorganisms but other microorganisms are capable of growth at these temperatures.

Microorganisms can grow over a temperature range of 30°C or more but the optimum growth temperature is within a very narrow range of temperature. Above the optimum temperature, enzymes become denatured and the rate of growth drops, producing what is called the maximum growth. Below the optimum temperature range the rate of growth decrease slowly until a point is reached, called the minimum growth temperature, where growth ceases.

Microorganisms have been divided into three groups based on their optimum temperature:

A- Psychrophiles

Exhibit an optimum range of growth between 0°C and 20°C over this temperature the ribosomes will be unstable. Psychrophiles are found in great numbers in the Antarctic

where temperature fall below 0°C also found in cold soil, streams, rivers, and lake mud. They are also capable of growing in a household refrigerator where they are important agents of food spoilage. These microorganisms are unable to live in high temperature because of the inhibition of enzymes.

B-Mesophiles

They have an optimal temperature range between 20-50°C. most of the bacteria grown in the microbiological laboratories are mesophiles. Many mesophiles have an optimal temperature of about 37°C., which similar to human body temperature. Many of normal resident microorganisms of the human body such as *E.coli* are mesophilic. They are able to grow rapidly and establish an infection within the human body. Some species are pathogenic to humans and other warm-blooded animals.

Mesophilic microorganisms have minimum and maximum temperature of 10°C and 50°C.

C-Thermophiles

Such as *Bacillus staerothermophilus* grown at higher

Thermophiles are found among many groups of microorganisms such as the Cyanobacteria, the photosynthetic bacteria, gram positive, gram negative bacilli, and protozoa.

Most species are found in hot springs in many areas of the world.

Thermophiles have ribosomes, enzymes systems, metabolic pathways, and regulatory devices that are functionally the same as those found in mesophiles.

The ability of thermophiles to survive at elevated temperature is a characteristic that has not been fully explained. Experiments show that the cytoplasmic proteins from thermophiles are resistant to temperatures that denature mesophilic proteins. Resistance to heat is a property associated with the endospores of such bacteria as the *Clostridia* these organisms release potent toxins (*Clostridia botulinum* toxin). Some spore forming *Clostridia* are also thermophilic and the vegetative cells can grow at high temperature.

It has been proposed that most thermophilic groups of microorganisms evolved from mesophily to thermophily.

2-Osmotic pressure

Changing the solute concentration not only alters the availability of water but also alters the osmotic pressure. The cell wall structure of bacteria makes them resistant to changes in osmotic pressure but extremity in osmotic pressure can result in the death of microorganisms.

In hypertonic solutions microorganisms may shrink and in hypotonic solution the cell may burst. An organism that can grow in solution with high solute concentration is called osmotolerant. Some microorganisms are osmophilic requiring a high solute concentration to grow like some fungi.

Some microorganisms known as halophiles specifically require sodium chloride for growth. Moderate halophiles include many marine bacteria grow best at salt concentrations of about 3% NaCl.

Extreme halophiles show maximum growth rate in saturated solutions. These organisms grow well in salt concentration of greater than 15% NaCl and can grow in salt lakes and pickle barrels.

High salt concentration normally disrupts membrane transport systems and denatures proteins.

The extreme halophilic bacterium *Halobacterium* possesses an unusual cytoplasmic membrane and unusual enzymes.

The degree of sensitivity to salt varies for different microbial species like some strains of *Staphylococcus* are salt tolerant and grow at salt concentration greater than 10%, so one of the reasons for mixing high concentration of salt or sugar with foods is to prevent growth of microorganisms through dehydration.

The addition of 12% NaCl is the best technique for reducing microbial growth, but this has no effect on certain halophiles.

3- Hydrostatic pressure

It refers to the pressure exerted by a water column as a result of the weight of the water column. Each 10 meters of water depth is equivalent to approximately one atmosphere pressure.

Most microorganisms are relatively tolerant to the hydrostatic pressure, but cannot tolerate the extremely high hydrostatic pressure of 200 atm since this will inactivate the enzymes and disrupt membrane transport system.

Some microorganisms are referred to as barotolerant (they are classified into obligate and facultative barotolerant) can grow at high hydrostatic pressure.

The inhibition of microbial cell is attributed to the accumulation of acids inside the cell.

4- Acidity and pH

The pH of a solution describes the hydrogen ion concentration (H^+). Microbial growth rates are greatly influenced by pH values. The pH greatly influences the structure and function of proteins and enzymes.

The pH values must be controlled in order to achieve optimal growth rates. This is normally accomplished by buffering the solution. Buffers are used to maintain the pH value within a particular range.

Microorganisms vary in their pH tolerance ranges. Fungi generally show a wider pH range growing well over a pH range of 5-9 compared to most bacteria, which grow well over a pH range of

6-9. Some fungi grow well at lower pH values as low as **zero.** Some protozoa and algae are able to grow at low pH values.

Some bacteria called acidophiles are restricted to grow at low pH value, such as *Thiobacillus*.

Microorganisms live in a wide variety of habitats but most microorganisms grow at pH values near neutrality (pH 6-8).

In general, microorganisms may divide into three categories:

1- *Alkalophiles*

Grow in a pH range of 7-11 with an optimum of 10.

2- *Neutrophiles*

Grow in a pH range of 4-9 with an optimum near neutrality.

3- *Acidsophiles*

Live at low pH values and can be divided into **facultative** and obligate. Such as *Sulfolobus* (bacteria) and *Thermoplasma* (mycoplasma).

Facultative acidophiles grow well at acid pH values but can also grow at pH values of 7 or above, such as fungi.

Obligate acidophiles grow normally at pH values of 3 or less and usually cannot grow above this pH.

5-Radiation

The electromagnetic spectrum divided into certain categories of radiation including gamma rays, X-ray, UV light, high energy, and short-wave length radiation disrupt DNA molecules and exposure to short wavelength radiation may cause mutation many of which are lethal.

The visible light is a source of energy for photosynthetic microorganisms such as Cyanobacteria and purple and green bacteria where the light energy is converted into chemical energy in the cell.

substances. Microorganisms used several different transport mechanisms, the most important of them are:

1- Facilitated diffusion

2- Active transport

3- Group translocation

Facilitated diffusion:

A few substances can cross the cytoplasmic membrane by passive diffusion. In this process molecules move from a region of higher concentration to one of lower concentration. The rate of passive diffusion is depending on the size of molecule and the concentration gradient between the external and internal environment. The rate of diffusion across the selectively permeable membrane is increased by the use of carrier proteins (sometimes called **permeases**), which present in the cytoplasmic membrane and in this case the passive diffusion called facilitated diffusion.

Active transport:

It is the transport of solutes to **higher concentration or against the concentration gradient** with the use of metabolic energy in input. The solute molecules are moved across the membrane **without any modification**.

Group translocation:

A substrate is becoming phosphorylated during the transport process. In this process, a membrane carrier proteins is first phosphorylated in the cytoplasm at the expense of phosphoenolpyruvate; the phosphorylated carrier proteins then binds the free sugar at the exterior me face and transports it into the cytoplasm, releasing it as sugar-phosphate. Such systems of sugar transport are called **phosphotransferase systems**.

Microbial Genetics:

The genetic information of prokaryotic and eukaryotic microorganisms encoded within the **DNA (deoxyribonucleic acid) molecule** and sometimes (as in viruses) in the **RNA (ribonucleic acid) molecule**. These molecules are known as **macromolecules** and they are responsible for the ***transition of hereditary information from one generation to the other.*** Another macromolecule found in the cell is the protein, which is the **result of the genetic code** into its structural or functional form.

A **third type of nucleic acids** was discovered in 1994, called PNA; peptide nucleic acid, PNA is an analogue to DNA in which the backbone chains are modified polypeptides (rather than chains of sugar-phosphate units); two strands of PNA can hybridise to form a DNA-like helical duplex. This molecule may have evolutionary significance; it has been suggested that pre-biotic nucleic acids may not necessarily have had a sugar-phosphate backbone.

The structure of nucleic acids and their replication

The genetic information of a cell forms a GENOME. The genome of a microorganism is divided into segments consisting of DNA nucleotides sequences known as a GENE. These genes may have structural or functional, metabolic functions.

DNA structure

The DNA is a double helix where each strand is composed of a sequence of nucleotides; phosphodiester bonds link these nucleotides to each other. Each nucleotide is formed of a *deoxyribose sugar, a nitrogen base and a phosphate group.*

Four nitrogen bases are found in DNA: **adenine** (A), **guanine** (G), **cytosine** (C), **and thymine** (T). A and G are purines, while C and T are **pyrimidines**.

The primary structure of DNA

It is resembled by the sequence of nucleotides in a single strand. In this structure when the nitrogen base is bound to the sugar it is known as a **nucleoside**, when a phosphate group is linked to the nucleoside it is known as **nucleotide**.

The secondary structure of DNA

The two strands of the double - helix are **complementary** and **antiparallel**. They are **complementary** because A in one strand always connected by a double hydrogen bonds with T of the complementary strand (forming what is called a base pair); C always connected (base pair) by triple hydrogen bonds with G of the complementary strand.

They are **antiparallel** because the 5' → 3' strand starts with a 5'-$PO_4^=$ group and ends with a 3'-OH free group while the complementary strand has inverse polarity starting with 3'-OH ending with 5'-$PO_4^=$ (3' → 5').

This DNA double-helix model was proposed by **Watson and Crick** 1953 and accordingly the base ratio equation was put fourth:

$$\frac{A + T}{G + C} = 1$$

In a DNA molecule, each turn in a double helix has 10 bases, and the diameter of a single turn is two nm (1 Kilobase = 1000 base pairs and has a molecular weight of 3.3×10^5 per strand).

A DNA molecule always carries a negative charge due to the $PO_4^{=}$ groups. These charges are neutralized by alkaline proteins known as histones in eukaryotes, **histone-like proteins** in prokaryotes.

Structure of RNA molecule

A RNA molecule is usually single stranded; it has a sequence of ribonucleotides each is formed of a ribose sugar, a nitrogen base (A, G, C, and **Uracil (U) instead of thymine**), and a phosphate group. A ribonucleoside is formed of a **ribose sugar** and a nitrogen base.

DNA replication

DNA replicates when the cell **is ready for division. Meselsen and Stahl** first studied this process that proved experimentally using N_{15}-labeled DNA that the DNA replication is semi-conservative.

The process of replication begins when the cell membrane is formed for the new cell; the chromosome is attached to the cell membrane before replication (in prokaryotes the chromosome is mostly **circular** except in some viruses it is linear) in eukaryotes DNA is linear.

The initiation stage in DNA replication is the unwinding of the double helix when each strand serves as a **template**. An enzyme known as RNA **polymerase** is needed to form a primer (a small segment of

RNA) which serves as a leader to which the DNA nucleotides link and bind together in sequence.

The point where DNA starts unwinding is known as the **replicon** (or origin of replication) when the opened double helix with strands apart forms the **replication fork**. The enzyme that participate in unwinding the double helix at the replication fork are; **DNA gyrase** and helix destabilizing protein **helicase**. Another protein takes part in holding the two strands apart it is the **replication protein** (single strand binding protein). The new strand will be complementary to the template strand and antiparallel.

The following step will be the **elongation stage**. In this stage the leading strand will be in the 5' → 3' direction synthesis of DNA in this direction is continuous but for the **lagging** strand, which will be in the 3' → 5' direction DNA synthesis is not possible so along this strand small DNA fragments of separate primers are synthesized in the 5' → 3' direction, these are known as Okazaki fragments.

DNA synthesis in both strands requires **DNA polymerase III** to join the nucleotides 3'-OH ⌒ 5'-$PO_4^{=}$ and **DNA polymerase I** is required to remove the primers. The gaps are then filled with the proper nucleotides and joined by ligase enzyme.

Types of RNA and steps in proteins synthesis

There are three types of RNA (**mRNA, tRNA, and rRNA**). Their roles will be described within the process of protein synthesis.

Messenger RNA (mRNA):

It is formed in the nucleus of eukaryotes and nuclear region of prokaryotes. It carries the information **transcribed** from the DNA to the ribosomes (in the cytoplasm) where protein is synthesized. It is **transcribed** from a single strand of DNA and is complementary to that strand.

mRNA is a single strand with a sequence of ribonucleotides to be **translated** by the ribosomes to the required protein.

Transcription is a non-symmetric process since only one strand of DNA is transcribed (except in some viruses where DNA is single stranded by nature).

mRNA is synthesized by a DNA-dependent RNA polymerase known also as **transcriptase**.

Transcription is the **first step** in protein synthesis.

The **second step** in protein synthesis is **translation**; it requires the presence of the other two types of RNA; transfer RNA tRNA and ribosomal RNA (rRNA).

Transfer RNA (tRNA)

It is also known as **soluble RNA** has a distinguished clover leaf structure and two recognition sites; one binds to an activated amino acid, the second is known as the **anticodon** that recognizes the **codon** on the mRNA.

The genetic code

Every **codon** is made of three nucleotides coding for one amino acid, and since there are four nitrogen bases the probabilities of the number of

genetic codes are $4^3 = 64$. There are 20 amino acids therefore there could be more than one code for most amino acids.

For each amino acid there is one or more tRNA that carries the specific activated amino acid to the ribosome.

Ribosomal RNA (rRNA)

It is a major component of the ribosome. There are three rRNA in prokaryotes and four rRNA in eukaryotes each is distinguished by its molecular weight. The rRNA play an important role in protein synthesis because it forms an essential part of the ribosome which is responsible for sequencing the amino acids in the proper order according to the codon sequence of the DNA.

Protein synthesis

There are three stages in proteins synthesis:

1- **Activation:** when each amino acid is activated by a specific amino acyl synthetase. This is also known as the initiation stage. The starting signal is f-met (formyl-methionine). This stage requires If_1, If_2, and If_3 (If = initiation factor, which is a protein).

2- **Elongation stage:** it is achieved by the following steps:

i) Attachment of the activated amino acid to a tRNA molecule The specificity of the tRNA is determined by the anticodon sequence on the anticodon arm.

ii) The mRNA is attached to the ribosome at a specific site

iii) The amino acid is transferred and carried by the tRNA to the ribosome at the site bound to the mRNA codon complementary to the anticodon of the tRNA where each C is bound to G and each A to U. This process needs GTP (Guanosine Triphosphate) as an energy source.

iv) Translocation on the ribosome takes place and other amino acids are placed in position, these amino acids are then connected together by a peptide bond formed by the peptidyl transferase enzyme.

3- **Termination stage:** termination of translation occurs when a stop codon enters the ribosome and three release factors (Rf_1, Rf_2, and Rf_3) help in releasing the tRNA, mRNA, and proteins from the ribosomes.

Plasmids

They are DNA molecules other than the chromosomal DNA and are found in prokaryotic microorganisms and some eukaryotes such as yeasts.

A plasmid is a double stranded DNA segment found in the cytoplasm unrelated to the chromosome and can replicate independently.

Plasmids can be divided into two types according to size:

1- Large size plasmids with high molecular weight 60-120 (kb) known as **R-plasmids (resistance)** carrying genes responsible for antibiotic resistance or **F-plasmids (fertility)** which have the ability to transfer some chromosomal genes within the cells. These plasmids are also known as Conjugative plasmids.

2- Small size plasmids are 2.5-15 kb they carry bacteriocinogenic elements and resistance to some antibiotics or chemicals, they are also known as **non-conjugative** plasmids.

Transfer of genetic materials bacteria

There are three different mechanisms for genetic exchange in bacteria:

1-Transformation

A free DNA molecule is transferred to a recipient cell of bacteria. The double stranded DNA could enter as it is like in gram negative ***Haemophilus*** cells or it could enter single stranded as mostly happens in gram positive cells. After entering the cell the single strand DNA recombines with the DNA of the recipient cell and the new cell is known as the **transformed cell**. A special treatment is needed to facilitate the entrance if the DNA to the recipient cell; this could be accomplished by treatment of recipient cells with $CaCl_2$ then it is known as a competent cell (ready to receive the new DNA).

2-Transduction

It occurs when the DNA of a donor is transferred to the recipient bacteria by a bacteriophage. Transfer of the genetic material by this method is more frequent than by transformation. There are two mechanisms for transduction:

General transduction: when apiece of DNA at random is carried by a phage and is transferred to the recipient cell later forming **transduced** cells.

i- *Specialized transduction:* it occurs when the bacterial DNA has a specific site for the phage DNA to incorporate, so when the phage DNA is released it carries with it the bacterial DNA genes that are closely attached to the phage DNA and transfers it to the recipient cell (an example is the lambda (λ) phage that infects *Escherichia coli*).

3- Conjugation

Another method for genetic transfer, this method requires the presence of the F (fertility or sex) factor present on the F-plasmid, which is responsible for the formation of **sex pili** or **Conjugation Bridge** from **donor (F^+)** cells to the **recipient (F^-)** cells. Some (F^+) cells are known as **high frequency recombinants (HFr)** they are (1000) times more competent than the F cells in transferring the genetic material.

Mutation in bacteria

The term mutation applies to all heritable changes in nucleotide sequence arising within an organism, they may be:

1- **Spontaneous** (naturally occurring)

2- **Induced** by some mutagenic agents, this could be chemical or physical.

Silent mutation: it is when the mutant is not altered phenotypically but only genotypically.

A mutation could occur by **deletion**, **insertion** of a nucleotide, **transition**, (purine into pyrimidine or pyrimidine into purine) or **transversion** (purine into purine or pyrimidine into pyrimidine).

A point mutation occurs when one nucleotide is inserted or deleted.

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Types of mutagens

1- Base analogs (5-bromouracil) instead of thymine.

2- Deaminating agents such as nitrous acid converts adenine to hypoxanthine.

3- Alkylating agents such as ethyl ethane sulphonate (EES) modifies guanine to bind to thymine.

4- Acridine derivatives.

5- Others such as Manganese and formaldehyde.

Bacterial mutants

Several types of mutants are known:

1- Antibiotic or drug resistant mutants.

2- Mutants that differ in their fermentation products.

3- **Auxotrophs**: are mutants that lack the ability to synthesize organic compounds necessary for their growth therefore they are supplemented with vitamins or amino acids and need to grow on rich media.

4- **Phenotypic mutants**: they are altered phenotypically by a change in morphology or color of the colonies.

5- Mutation in cell surface and antigenic structure.

6- Phage resistant mutants.

7- Mutations with altered structures (i.e. loss of flagella, capsule, or spore formation).

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Bacterial nutrition and energy source

Biological energy is derived from the oxidation of inorganic, organic molecules or from the sun.

The cell uses the energy to form specific function, one of which is the biosynthesis of new cellular components. The materials that are used to construct new cellular components are called nutrients.

The nutrients required for growth are those inorganic or organic components that supply the major elements to the cell.

The common nutrients requirements

Analysis of microbial cell composition shows that 95% or more of cell dry weight is made up of few major elements (carbon, oxygen, hydrogen, nitrogen, sulfur, phosphorus, potassium, calcium, magnesium and iron). These are referred to as macro elements or macronutrients because microorganisms in large amounts require them.

The first six are components of carbohydrates, lipids, proteins, and nucleic acids. The remaining four exist in the cell as cations and play a variety of roles

Potassium (K^+):

It is required for activity by a number of enzymes including some of those involved in proteins synthesis.

Calcium (Ca^{++}):

It has many functions and the most important one is the contribution to heat resistance of bacterial endospore formation.

Magnesium (Mg^+):

It serves as a cofactor for many enzymes also makes a complex with ATP, stabilizes ribosomes and cell membrane.

Iron (Fe^{++} or Fe^{+++}):

It is used in synthesis of cytochromes, as a cofactor for enzymes and electron carrying proteins.

All microorganisms require a number of micromolecules or trace elements in addition to macromolecules.

Most cells need the trace elements Mn, Zn, Co, Ni, and Cu, but in small amounts .As contaminants of water or glassware and regular media components, they are often adequate for growth. These microelements are normally a part of enzymes and cofactors and they aid in the catalysis of reaction and maintenance to proteins structure such as zinc, which is found at the active side of some enzymes, manganese aids many enzymes catalyzing activity, cobalt is a component of vitamin B_{12}.

Major elements that make up the organic matter of typical bacterial cell:

Element	Dry weight / percentage
Carbon	50
Oxygen	20
Nitrogen	15
Hydrogen	8
Phosphorus	3
Sulphur	1

Composition of function of the chemical components of bacterial cell:

Chemical components	Total cell wt.	Function
Water	70	Hydrolysis of organic molecules, solute transport, and source of electrons for some photosynthetic organisms.
Inorganic ions: (Ca, Na, Mg, PO_4, SO_4, etc.)	1	Active components of many enzymes and proteins.
Sugars and precursors	3	Major source of energy, components of cell wall, capsule, slime layer, and components of DNA.
Amino acids and precursors	0.4	Precursors of proteins sources of energy, and components of cell wall.
Nucleotides and precursors	0.4	Precursors of nucleic acids, and source of energy as ATP.
Lipids and precursors	2	Components of gram negative cell wall, cytoplasmic membrane, and source of energy.
Macromolecules; Proteins	15	Components of cytoplasmic membrane, cell wall, pili, flagella, ribosomes, and most of enzymes.
Nucleic acids ANA RNA	 1 6	Primary genetic components of cells, components of ribosomes, mRNA, and tRNA, that function in proteins synthesis.

The nutritional requirements of microorganisms vary among different species also these requirements can change with in a species as a result of gene mutation. A microorganism that requires the same nutrients as the majority of naturally occurring members of its species is a prototroph.

If a prototrophic microorganism mutates in such a way that it can't synthesize a molecule essential for its growth and reproduction it will this molecule, therefore it must obtain it or its precursors from the surrounding environment, such a microorganism is called an auxotroph.

Nutritional types of microorganisms

The size, morphology, and chemical composition of the bacterial cell can vary depending on environment and the genetic make up of the organism. All organisms require source of energy, hydrogen, and electron for growth to take place.

There are only two sources of energy available to organisms:

1- *Light energy*

2- *The energy derived from oxidizing organic or inorganic molecules.*

Phototrophs: microorganisms use light as an energy source, such as *Chlorobium, Chromatium,* and *Rhodospirillum.*

Chemotrophs: microorganisms obtain energy from the oxidation of chemical compounds.

Microorganisms also have only two sources of hydrogen atoms or electrons.

Lithotrophs: microorganisms use reduced inorganic substances as an electron source.

Organotrophs: microorganisms extract electrons or hydrogen from organic compounds.

Photolithotrophic autotrophs (often called phototrophs): microorganisms use light energy and CO_2 as a carbon source.

Chemoorganotrophic heterotrophs (often called chemotrophs or heterotrophs): microorganisms use organic compounds as a source of energy, hydrogen, electron, and carbon for biosynthesis.

Essentially **all pathogenic microorganisms** are chemoheterotrophs while large majority of microorganisms are either photolithorophic autotrophs or chemoorganotrophic heterotrophs.

Phototrophs
- → Photolithotrophs(Photoautotrophs) *Chlorobium, Chromatium*
- → Photoorganotrophs *Rhodospirillum*

Chemotrophs
- → Chemoautotrophs (chemolithotrophs)
- → Chemoorganotrophic (heterotrophic)

A particular species usually belong to only one of the four nutritional classes but some show great metabolic flexibility and alter their metabolic pattern in response to environmental change.

Nutritional types of microorganisms:

Major nutritional types	Sources of energy (H, e, and C)	Representative microorganism
Photolithotrophs	Light energy, inorganic Hydrogen / electron donor CO_2 carbon source	Algae, purple and green sulfur bacteria, and cyanobacteria
Photoheterotrophs	Light energy, inorganic Hydrogen / electron donor Organic carbon source	Green sulfur bacteria
Chemolithotrophs	Chemical energy sources (inorganic), inorganic hydrogen / electron donor CO_2 carbon source	Protozoa and fungi
Chemoorganotrophs (heterotrophs)	Chemical energy sources (organic), organic hydrogen / electron donor Organic carbon source.	The majority of non photosynthetic bacteria

Requirements of nitrogen, phosphorus, and sulfur

Carbon: the sole inorganic source of carbon is CO_2 (microorganism called autotrophs) while glucose and amino acids are the sole source of organic carbon, which is needed for anabolism and release energy and microorganism called heterotroph.

Mixotrophs: bacteria that depending on inorganic energy sources and organic carbon sources.

Nitrogen: is needed for the synthesis of amino acids, purines, pyrimidines, nucleic acids, enzymes, and vitamins.

Phosphorus: is present in nucleic acids, phospholipids, nucleotides like ATP, several cofactors, some proteins, and other cell components. It is found in K_2HPO_4 and KH_2PO_4

Sulfur: is needed for the synthesis of substances like amino acids (cystine, and methionine), thiamine, biotin, and some carbohydrate

Growth factors

Microorganisms especially most of photolithotrophic autotrophs often grow and reproduce when minerals and sources of energy, C, N, P, and S are supplied. These microorganisms have the enzymes and pathways necessary for synthesize all cell components required while other microorganisms lack one or more essential enzymes so they can't synthesize all cell components but must obtain them or their precursors from the environment and there are three major classes or growth factors:

1-Amino acids: needed for proteins synthesis.

2-Purines and pyrimidines: needed for nucleic acids synthesis.

3-Vitamins: usually make up all or part of enzymes, and cofactors.

Mineral salts

Microorganisms need mineral salts in a small quantity of inorganic ions (cations and anions) such as:

1- Macronutrients elements:

They are needed for activation of enzymes, enzymes' cofactors, and controlling the osmotic pressure inside the cell. They include Mg^{+2}, Zn^{+2}, Ca^{+2}, Na^{+}, and Cl^{-}. Macronutrients are required in concentrations about 0.1-1 mMol.

2- Micronutrients elements:

They are required in concentrations about 10^{-3}-10^{-5} mM, Such as Co^{+2}.

- There is a group of microorganisms, which needed Na^{+} and Cl^{-} in high concentrations called Halophiles that can be classified into:

I- Slightly holophiles

Those who needed a small quantity of NaCl (2-5 %), include most of marine bacteria.

II-Moderately holophiles

NaCl is needed in a range about (5-10 %) such as *Pseudomonas, Lactobacillus*, and Protozoa.

III- Extremely holophiles

NaCl is needed in a range about (20-30 %) such as *Halobacterium,* and *Micrococcus*. NaCl is necessary to stabilize the binding of cell wall proteins to each other.

Uptake of nutrients by the cell

The first step in nutrients utilization is uptake of the required nutrients by the microbial cell.

No molecules must pass through the selectively permeable cytoplasmic membrane that will not permit the free passage of many