#Microbiology

7

Microbial Growth

Reproductive Strategies

- The reproductive strategies of eukaryotic microbes → Sexual → Asexual
  - asexual and sexual, haploid or diploid
- *Bacteria* and *Archaea* → Reproduce Asexually By Binary fission.
  - **haploid only, asexual - binary fission, budding, filamentous**
    (Main Reproductive Process.) (Some Microorganisms Can Reproduce by.)
  - all must replicate and segregate the genome prior to division
    (segregate → How Pairs of Gene Variants Are Separated into Reproductive cells.)

What do we mean by Growth? → Increase in Size and number.

* Bacterial and Archial cells Reproduce Asexually.

* All Bacterial cells must Replicate their DNA before division. (Replicate Chromosome → Segrigation of the Genome)

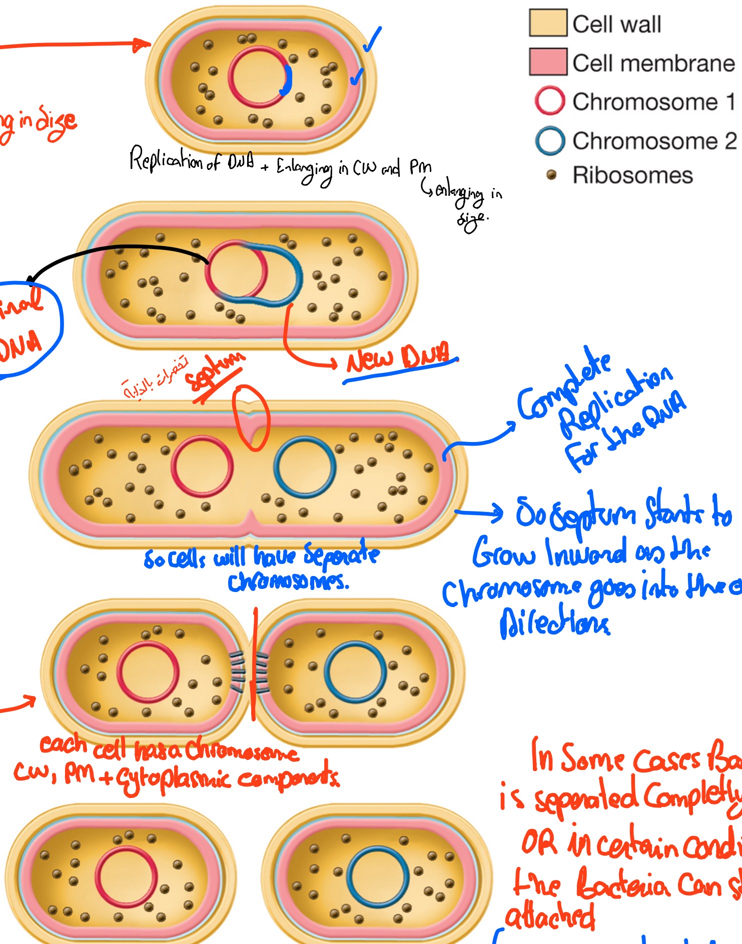
(a) A young cell at early phase of cycle

(b) A parent cell prepares for division by enlarging its cell wall, cell membrane, and overall volume. DNA replication then starts.

(c) The septum begins to grow inward as the chromosomes move toward opposite ends of the cell. Other cytoplasmic components are distributed to the two developing cells.

(d) The septum is synthesized completely through the cell center, creating two separate cell chambers.

(e) At this point, the daughter cells are divided. Some species separate completely as shown here, while others remain attached, forming chains, doublets, or other cellular arrangements.



Bacterial Cell Cycle

- Cell cycle is sequence of events from formation of new cell through the next cell division
  - most bacteria divide by binary fission ✓
- Two pathways function during cycle
  - **DNA replication and partition** ✓
  - **cytokinesis** ✓

*Cells must Go through Phases in order to grow

– Three phases for cell cycle:

1) Period of Growth after cell is Born
2) Chromosome Replication and partition
3) cytokinesis.

first Phase of division

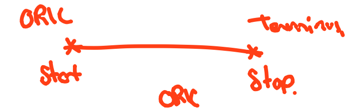
Chromosome Replication and Partitioning - 1

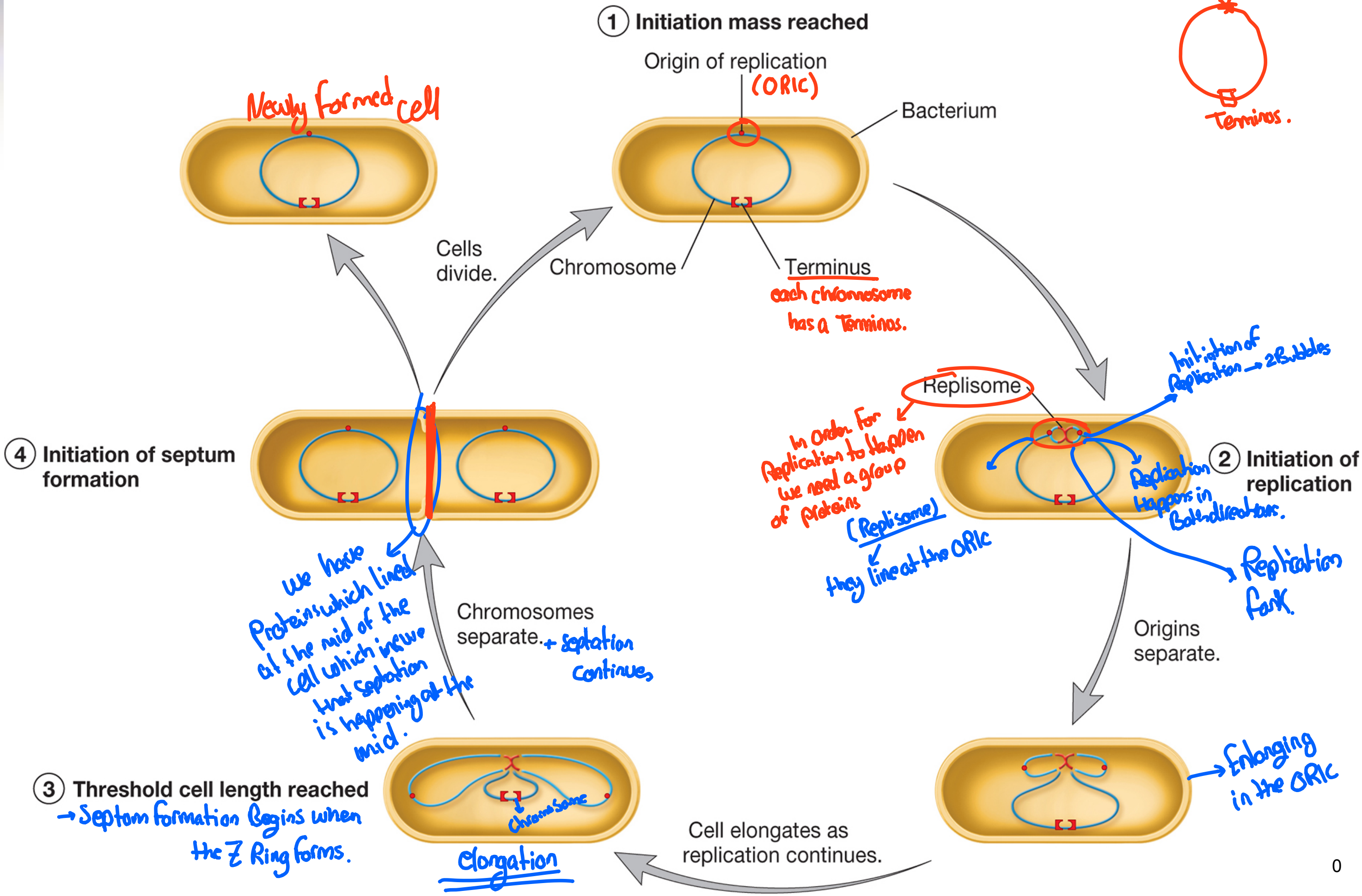
Some microorganisms can have more than one chromosome.

Single cellular Chromosome

Majority

- Most bacterial chromosomes are circular
- **Single origin of replication** – site at which replication begins
- **Terminus** – site at which replication is terminated, located opposite of the origin
- **Replisome** – group of proteins needed for DNA synthesis
- DNA replication **proceeds in both directions from the origin**
- Origins move to opposite ends of the cell


ORIC
Terminus
Start
Stop.
ORIC
Terminus.


1 Initiation mass reached
Origin of replication
(ORIC)
Bacterium
Chromosome
Terminus
each chromosome has a Terminus.
Newly formed cell
Cells divide.
Replisome
In order for Replication to happen we need a group of proteins (Replisome)
they line at the ORIC
Initiation of Replication → 2 Bubbles
Replication Happens in Both directions.
Replication fork
2 Initiation of replication
Origins separate.
Elongating in the ORIC
Cell elongates as replication continues.
Elongation
Chromosome
3 Threshold cell length reached
→ Septum formation Begins when the Z Ring forms.
Chromosomes separate. + Septation continues
we have proteins which lined at the mid of the cell which insure that septation is happening at the mid.
4 Initiation of septum formation

Chromosome Partitioning

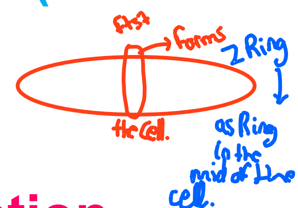
- **Replisome pushes, or condensation of, daughter chromosomes to opposite ends**
- **MreB (*mure*in cluster *B*)** – an actin homolog, plays role in determination of cell shape as spiral inside cell periphery, and chromosome segregation
  - new origins associate with MreB tracks
  - if MreB is mutated, chromosomes do not segregate

if there was any mutation in MreB → No segregation will happen.

Septation will happen in middle only Because we have Proteins that Aids in septation Process → For example the Z Ring.

Cytokinesis - Septation

- **Septation** – formation of cross walls between daughter cells
- **Several steps**
  - selection of site for septum formation
  - assembly of Z ring → Composed of FtsZ → tubilin → Protein Lines at the mid and form the Z Ring.
  - linkage of Z ring to plasma membrane (cell wall)
  - assembly of cell wall (forms) synthesizing machinery
  - constriction of cell and septum formation



Z Ring Insure that division will Happen at a Certaine Place.

Z Ring Formation - Role in Septation

- **Protein FtsZ**
  - tubulin homologue, found in most bacteria and archaea
  - polymerization forms Z ring, filaments of meshwork
- **MinCDE** system in *E. coli* limits the Z ring to the center of the cell
  - MinC, MinD, MinE oscillate from one side of cell to other
  - link Z ring to cell membrane
  - Z ring constricts and cell wall synthesis of septal wall

Prevents FtsZ Polymerization near certain parts of plasma membrane.

MinCDE → Moves from pole to pole

allow the mid of the cell to be empty.

* Cellular Growth and determination of Cell Shape:

Why do Microorganisms have different shapes? Because of the peptidoglycan (Cell wall) → Some Microorganisms lack the cell wall. → Such as micoplasma

Morphic

Doesn't have a specifect shape.

So cell wall is Determinted → Determining the shape of Microorganism (Cocci, Bacilli)

_ But not only the cell wall also the cytoskeletal elements → Gives Different shapes of microorganisms.

→ Cell wall is mainly Composed of NAG + NAM (From Sugars) NAM and NAG Are joined togather by Glycocidic linkage

↓

Can be broken by enzymes (lyzosims)

↓

can be found in Saliva and tears.

↓

Kills Microorganisms that found in eyes and mouth

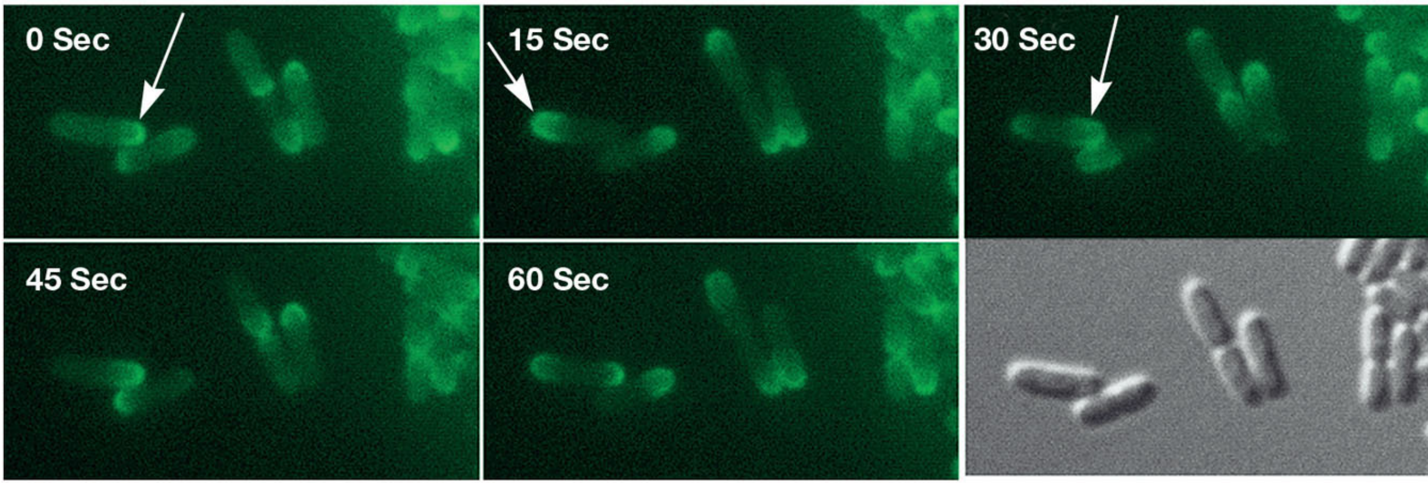
Peptidoglycan Aids in → Support, Protection, Maintain Osmotic pressure in the cell.

Peptidoglycan Biosynthesis → Starts with cytoplasm.

(UDP → Attached to NAG → then it's transfered to NAM) → Cytoplasmic Side of the cell

→ then it goes to the periplasmic membrane

→ Attatched to A Carrier → Flip On the opposit direction → So NAM in direct Contact to the periplasmic space.



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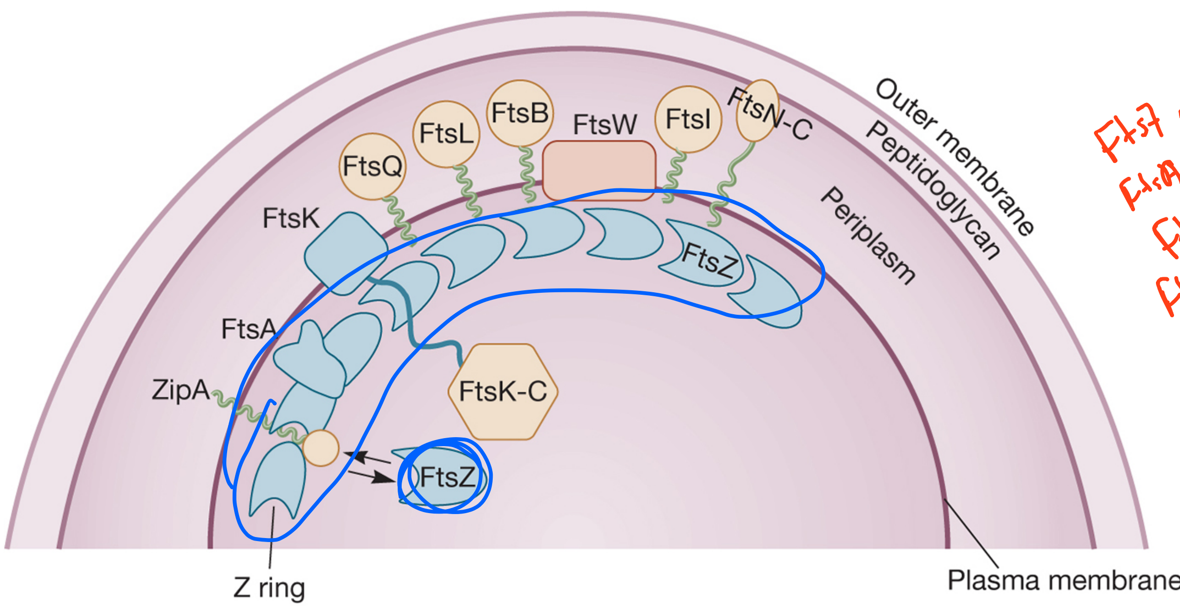


The *E. coli* divisome

What is divisome? Proteins that Are Involved in the division

Cytoskeletal element

MReB → Determine the Diameter and elongates the Z Ring.

moves Around and Protects the shape of cell.

Some Proteins of divisomes Aid in Partitioning (Make Sure that each cell has 1 Chromosome).

Table 7.1 Divisome Proteins and Their Functions

Divisome Protein	Function
FtsA, ZipA	Anchor Z ring to plasma membrane
FtsZ	Forms Z ring
FtsK	Chromosome segregation and separation of chromosome dimers
FtsQLB	May provide a scaffold for assembly of proteins involved in peptidoglycan synthesis
FtsI[1], FtsW	Peptidoglycan synthesis
FtsN	Thought to trigger constriction initiation

1 FtsI is also known as penicillin-binding protein 3 (PBP3).

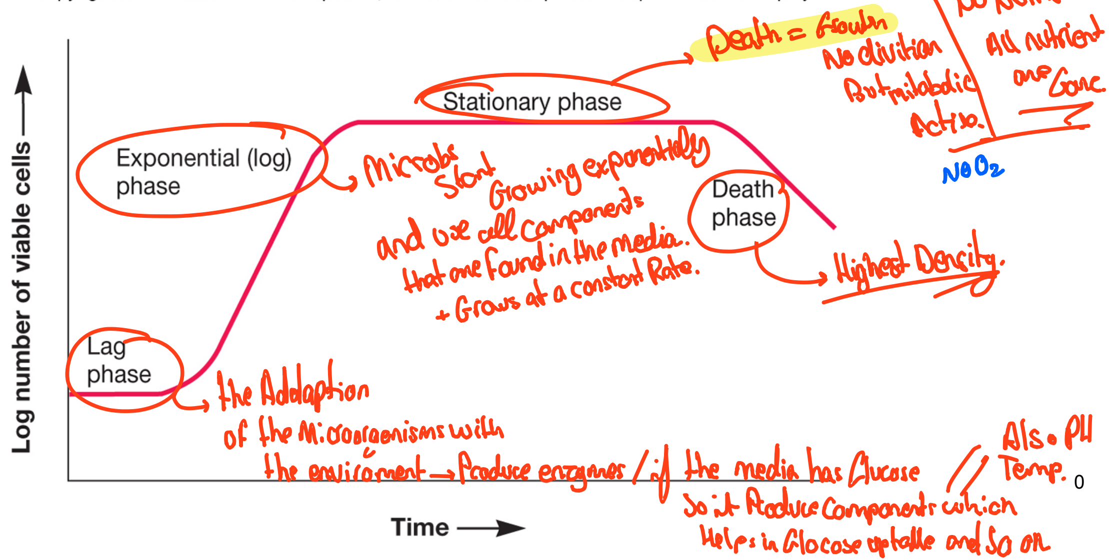
Growth

- Increase in cellular constituents that may result in:
  - **increase in cell number**
  - **increase in cell size**
- Growth refers to population growth rather than growth of individual cells

Microorganisms growth Go through Phases.

The Growth Curve

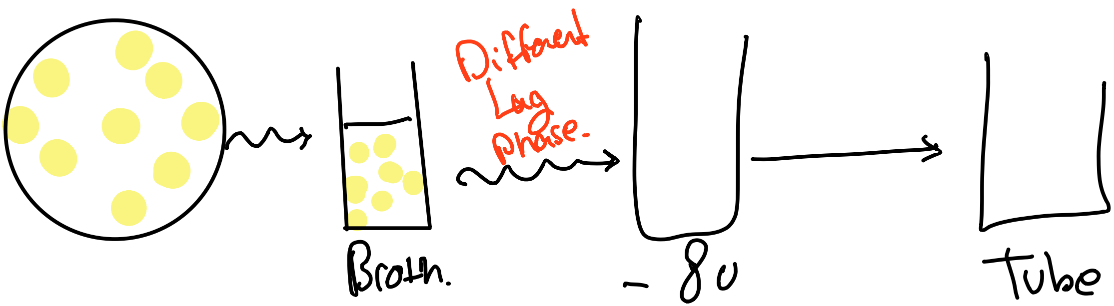
- Observed when microorganisms are cultivated in **batch culture** → Broth // usually liquid Media → Nutrients Are limited once it's Gone. Bacteria will not Grow.
- Usually plotted as logarithm of cell number versus time
- Has four distinct phases



Lag Phase

Some Microorganisms don't have the lag phase depending on where do I take the microorganisms.

- **Cell synthesizing new components**
  - e.g., to replenish spent materials
  - e.g., to adapt to new medium or other conditions
- **Varies in length**
  - in some cases can be very short or even absent



Cells Grow as Quickly as they can in this phase

Exponential Phase

↳ Depending on the environment Log phase varies.

- Also called log phase
- **Rate of growth and division is constant and maximal**
- Population is **most uniform** in terms of chemical and physical properties during this phase

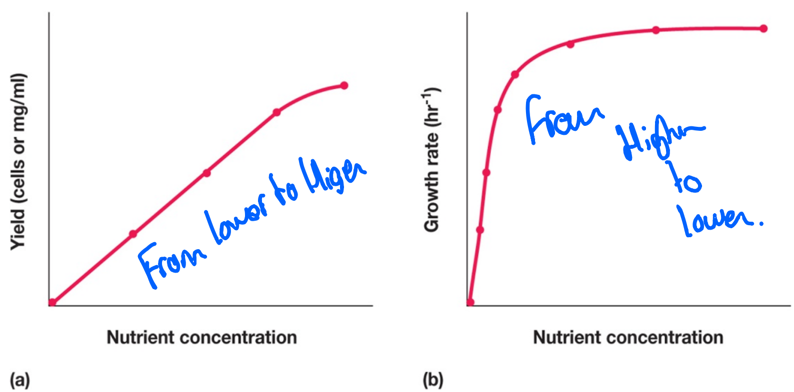
→ Nutrients can be limited ⇒ if they were too little it may effect the Growth

Balanced Growth ~> Constant Rate.

- During log phase, cells exhibit balanced growth
  - cellular constituents manufactured at constant rates relative to each other

Unbalanced Growth

- Rates of synthesis vary relative to each other
- Occurs under a variety of conditions
  - **change in nutrient levels**
    - shift-up (poor medium to rich medium)
    - shift-down (rich medium to poor medium)
  - change in environmental conditions



Stationary Phase

- Closed system population growth eventually ceases, total number of viable cells remains constant
  - active cells stop reproducing or reproductive rate is balanced by death rate ✓

Growth = Death → No increase in the population.

But it stays metabolicly active.

Possible Reasons for Stationary Phase

- Nutrient limitation
- Limited oxygen availability
- Toxic waste accumulation → Bacteria Produce wasts.
- Critical population density reached

* At Death Phase → Death is more than Growth

* Long-Term Stationary Phase → Bacteria is getting Adapted to the environment,
takes time to Adapte with Harsh Conditions.. → Natural Selection] Mutation which helps in the Adaptation

Stationary Phase and Starvation Response

- Entry into stationary phase due to starvation and other stressful conditions activates survival strategy
  - 1) morphological changes
    - e.g., endospore formation
  - 2) decrease in size, protoplast shrinkage, and nucleoid condensation
  - 3) RpoS protein assists RNA polymerase in transcribing genes for starvation proteins

* Extremophiles → Grow under Harsh Conditions

Starvation Responses

- **Production of starvation proteins**
  - increase cross-linking in cell wall ✓
  - Dps protein protects DNA ✓
  - chaperone proteins prevent protein damage ✓
- Cells are called **persister cells** → arise due of dormancy // Because of Harsh conditions.
  - **long-term survival** ✓
  - **increased virulence** ✓

Senescence and Death Phase

- Two alternative hypotheses
  - cells are **Viable But Not Culturable (VBNC)**
    - cells alive, but dormant, capable of new growth when conditions are right
- **Programmed cell death**
  - fraction of the population genetically programmed to die (commit suicide)

The Influence of Environmental Factors on Growth

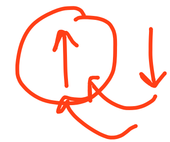
- Most organisms grow in fairly moderate environmental conditions
- **Extremophiles** ✓
  - grow under harsh conditions that would kill most other organisms

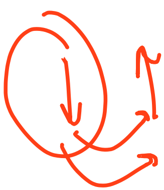
Table 7.2 Microbial Responses to Environmental Factors

Descriptive Term	Definition	Representative Microorganisms
Solute and Water Activity		
Osmotolerant ✓	Able to grow over wide ranges of water activity or osmotic concentration	*Staphylococcus aureus, Saccharomyces rouxii*
Halophile ✓	Requires high levels of sodium chloride, usually above about 0.2 M, to grow	*Halobacterium, Dunaliella, Ectothiorhodospira*
pH		
Acidophile ✓	Growth optimum between pH 0 and 5.5	*Sulfolobus, Picrophilus, Ferroplasma, Acontium*
Neutrophile ✓	Growth optimum between pH 5.5 and 8.0	*Escherichia, Euglena, Paramecium*
Alkaliphile ✓	Growth optimum between pH 8.0 and 11.5	*Bacillus alcalophilus, Natronobacterium*
Temperature		
Psychrophile	Grows at 0°C and has an optimum growth temperature of 15°C or lower	*Bacillus psychrophilus, Chlamydomonas nivalis*
Psychrotroph	Can grow at 0–7°C; has an optimum between 20 and 30°C and a maximum around 35°C	*Listeria monocytogenes, Pseudomonas fluorescens*
Mesophile	Has growth optimum between 20 and 45°C	*Escherichia coli, Trichomonas vaginalis*
Thermophile	Can grow at 55°C or higher; optimum often between 55 and 65°C	*Geobacillus stearothermophilus, Thermus aquaticus, Cyanidium caldarium, Chaetomium thermophile*
Hyperthermophile ✓	Has an optimum between 85 and about 113°C	*Sulfolobus, Pyrococcus, Pyrodictium*
Oxygen Concentration		
Obligate aerobe	Completely dependent on atmospheric O_2 for growth	*Micrococcus luteus*, most protists and fungi
Facultative anaerobe	Does not require O_2 for growth but grows better in its presence	*Escherichia, Enterococcus, Saccharomyces cerevisiae*
Aerotolerant anaerobe	Grows equally well in presence or absence of O_2	*Streptococcus pyogenes*
Obligate anaerobe	Does not tolerate O_2 and dies in its presence	*Clostridium, Bacteroides, Methanobacterium*
Microaerophile	Requires O_2 levels between 2–10% for growth and is damaged by atmospheric O_2 levels (20%)	*Campylobacter, Spirillum volutans, Treponema pallidum*
Pressure		
Piezophile (barophile)	Growth more rapid at high hydrostatic pressures	*Photobacterium profundum, Shewanella benthica*

Solutes and Water Activity

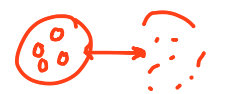
- Changes in osmotic concentrations in the environment may affect microbial cells
  - **hypotonic solution** (lower osmotic concentration)
    - water enters the cell
    - cell swells may burst
  - **hypertonic** (higher osmotic concentration)
    - water leaves the cell
    - membrane shrinks from the cell wall (plasmolysis) may occur





*Compatabile Solutes

Increase in Osmotic Conc. in Hypotonic Solutions



*Solutes Compatible with metabolism and Growth

Water Activity (aw)

- Water activity of a solution is 1/100 the relative humidity of solution
- Also equal to **ratio of solution's vapor pressure (Psoln) to that of pure water (Pwater)**
- Aw = Psoln/ Pwater
  - **low water activity means most water is bound**
- **Osmotolerant microbes can grow over wide ranges of water activity** But optimally at High levels.

amount of water in the cell is very important. (water effects the survival of microorganisms)

→ Drying food
take off water
when the environment is dry it's not good environment for microbial growth OR Adding salts on food such as the cheese → which kills microbes

Solutes and Water Activity

adding salts OR sugars → ↓ aw

- water activity (aw)
  - **amount of water available to organisms**
  - reduced by interaction with solute molecules (osmotic effect)

    higher [solute] lower aw
  - reduced by adsorption to surfaces (matric effect)

* most microorganisms only grow well around .98 OR higher. if aw is much lower microorgan won't survive.

95% OR 80%

the lower the water activity → the solutes are more attached to the cell

Microbes Adapt to Changes in Osmotic Concentrations

- Reduce osmotic concentration of cytoplasm in hypotonic solutions
  - mechanosensitive (MS) channels in plasma membrane allow solutes to leave
- **Increase internal solute concentration with compatible solutes to increase their internal osmotic concentration in hypertonic solutions**
  - solutes compatible with metabolism and growth

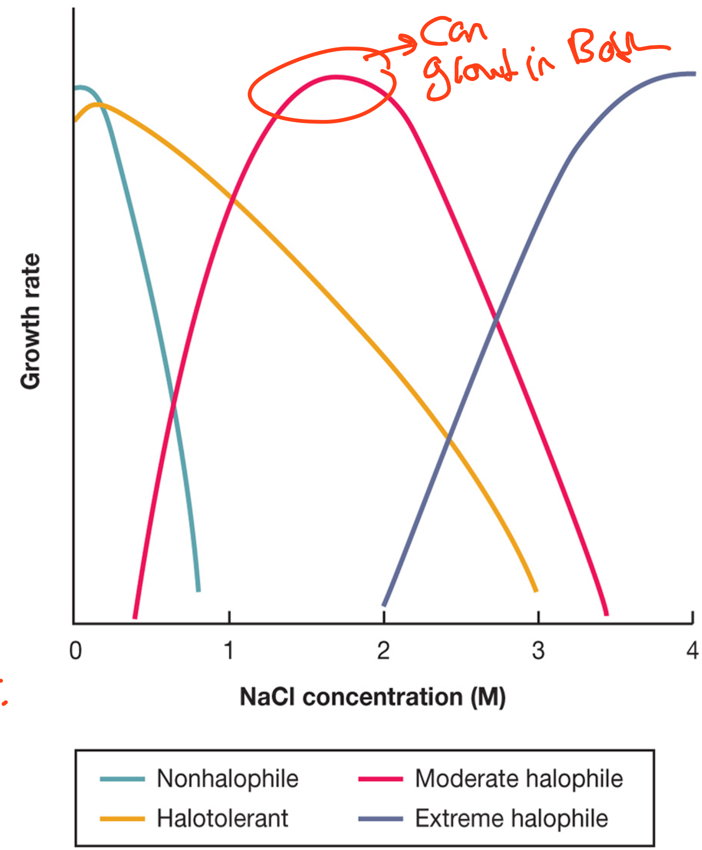
Extremely Adapted Microbes

- **Halophiles** → like environments with High Salt Conc.
  - grow optimally in the presence of NaCl or other salts at a concentration above about 0.2M
- **Extreme halophiles**
  - require salt concentrations of **2M and 6.2M**
  - extremely high concentrations of potassium
  - cell wall, proteins, and plasma membrane require high salt to maintain stability and activity ✓

* Non Halophiles → Cannot grow with Salt → Kills the microorganisms

Effects of NaCl on Microbial Growth

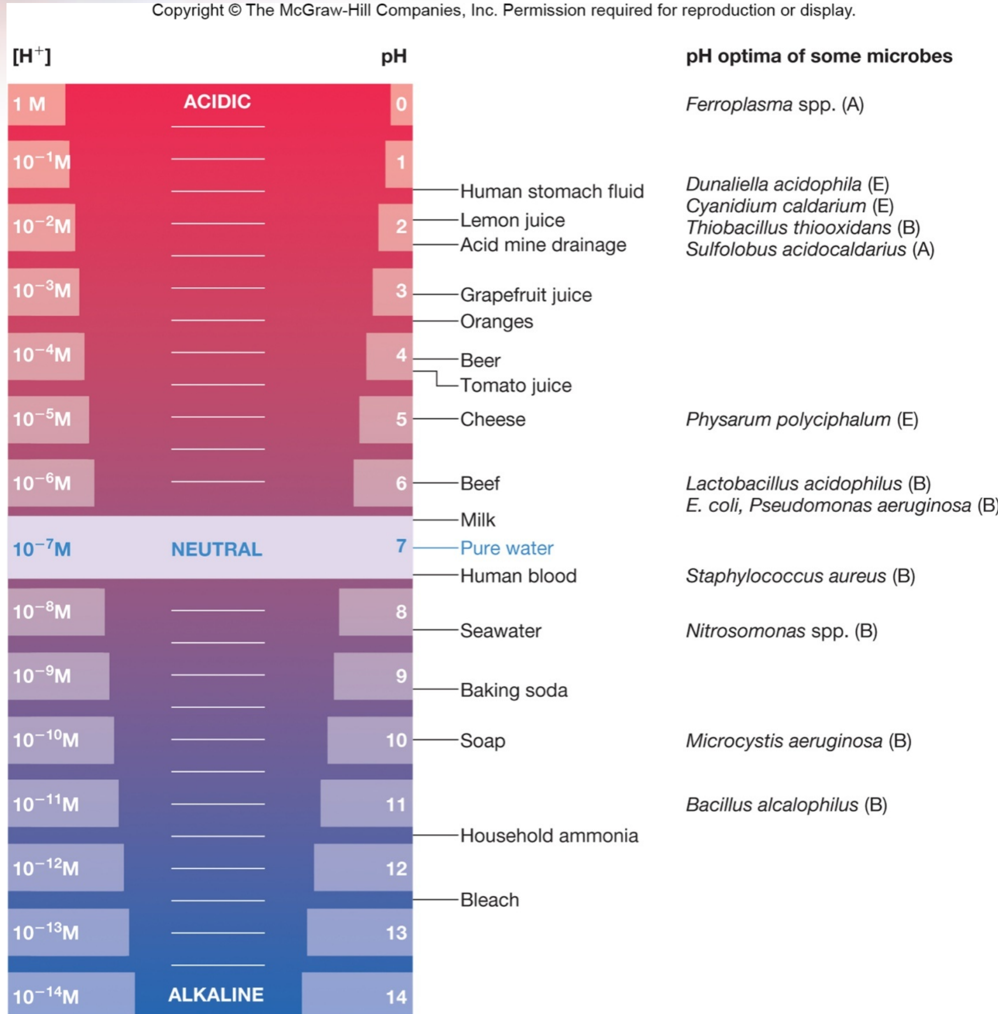
- **Halophiles**
  - grow optimally at >0.2 M
- Extreme halophiles
  - require >2 M



Halotolerant → S. aureus
↳ Can live in Salt enviroments.

pH

- measure of the relative acidity of a solution
- negative logarithm of the hydrogen ion concentration



pH

- **Acidophiles**
  - growth optimum between pH 0 and pH 5.5 Acidic
- **Neutrophiles**
  - growth optimum between pH 5.5 and pH 7 Neutral
- **Alkaliphiles (alkalophiles)**
  - growth optimum between pH 8.5 and pH 11.5 Basic

Pathogenic Bacteria → lives at pH 7

pH

*Most Fungi → More Acidic Surrounding 4PH–6PH.

- Most microbes maintain an internal pH near neutrality
  - the plasma membrane is impermeable to proton
  - exchange potassium for protons
- **Acidic tolerance response**
  - **pump protons out of the cell**
  - **some synthesize acid and heat shock proteins that protect proteins**

Protects from acidic and heat

- **Many microorganisms change the pH of their habitat by producing acidic or basic waste products**

waste Products

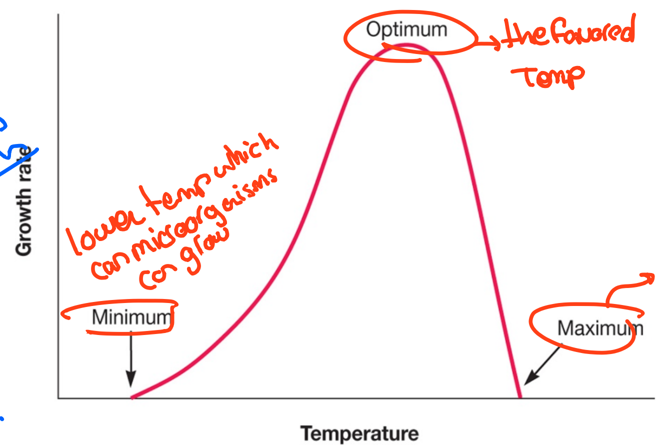
How do microorganisms Resist High or low pH?

Acidic → Pump Protons out of the cell (Transport system).

Basic →

Temperature

- Microbes cannot regulate their internal temperature
- Enzymes have optimal temperature at which they function optimally
- High temperatures may inhibit enzyme functioning and be lethal
- Organisms exhibit distinct **cardinal growth** temperatures
  - **minimal**
  - **maximal**
  - **optimal**



Temperature Ranges for Microbial Growth

- psychrophiles – 0o C to 20o C
- psychrotrophs – 0o C to 35o C
- mesophiles – 20o C to 45o C
- thermophiles – 55o C to 85o C
- hyperthermophiles – 85o C to 113o C

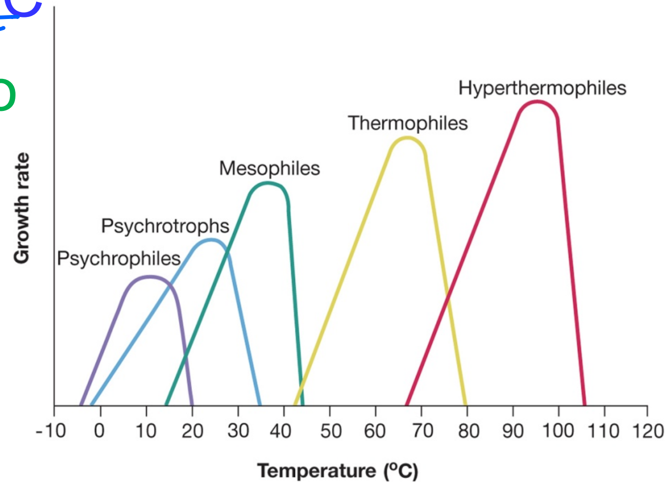


Table 7.3 Temperature Ranges for Microbial Growth

	CARDINAL TEMPERATURES (°C)		
Microorganism	**Minimum**	**Optimum**	**Maximum**
Nonphotosynthetic Bacteria and Archaea			
Bacillus psychrophilus	−10	23–24	28–30
Pseudomonas fluorescens	4	25–30	40
Enterococcus faecalis	0	37	44
Escherichia coli	10	37	45
Neisseria gonorrhoeae	30	35–36	38
Thermoplasma acidophilum	45	59	62
Thermus aquaticus	40	70–72	79
Pyrococcus abyssi	67	96	102
Pyrodictium occultum	82	105	110
Pyrolobus fumarii	90	106	113
Photosynthetic Bacteria			
Anabaena variabilis	ND[1]	35	ND
Synechococcus eximius	70	79	84
Protists			
Chlamydomonas nivalis	−36	0	4
Amoeba proteus	4–6	22	35
Skeletonema costatum	6	16–26	>28
Trichomonas vaginalis	25	32–39	42
Tetrahymena pyriformis	6–7	20–25	33
Cyclidium citrullus	18	43	47
Fungi			
Candida scotti	0	4–15	15
Saccharomyces cerevisiae	1–3	28	40
Mucor pusillus	21–23	45–50	50–58

1 ND, not determined.

Adaptations of Thermophiles

lives on Higher Temperature

- 1) Protein structure stabilized by a variety of means — Proteins (Shock Proteins) → Protect from ↑ Temp
  - e.g., more H bonds ✓ → Solidifies Proteins.
  - e.g., more proline
  - e.g., chaperones → Protection (Folding Proteins)
- 2) Histone-like proteins stabilize DNA
- 3) Membrane stabilized by variety of means → more solid membrane
  - e.g., more saturated, more branched and higher molecular weight lipids
  - e.g., ether linkages (archaeal membranes)

Oxygen Concentration

→ Oxygen is Really essential for microbial Growth, However certain microorganisms do not Require Oxygen → so Microorganisms can be divided into groups Based on the usage of oxygen.

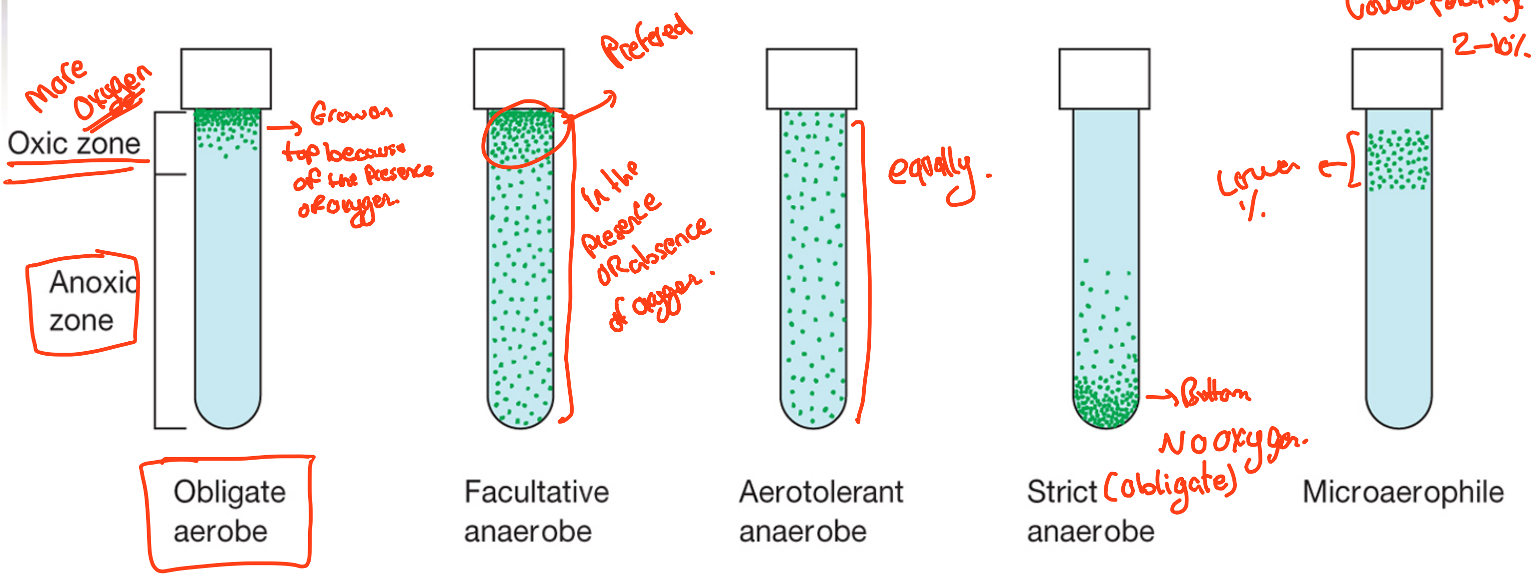
- growth in oxygen correlates with microbes energy conserving metabolic processes and the electron transport chain (ETC) and nature of terminal electron acceptor

Oxygen and Bacterial Growth

- **Aerobe**
  - grows in presence of atmospheric oxygen (O_2) which is 20% O_2
- **Obligate aerobe** – requires O_2 → Die without Oxygen
- Anaerobe
  - grows in the absence of O_2 → Can Grow in Absence of it
- Obligate anaerobe →
  - usually killed in presence of O_2 → if there is oxygen they will die

Oxygen and Bacterial Growth

- **Microaerophiles** → Require lower Oxygen Conc.
  - requires 2–10% O_2
- **Facultative anaerobes** → they don't Require O_2 → Better in oxygen.
  - do not require O_2 but grow better in its presence
- **Aerotolerant anaerobes** → Both Situations.
  - grow with or without O_2



Enzyme content → In order for microorganisms to grow it needs to have enzymes

Obligate aerobe	Facultative anaerobe	Aerotolerant anaerobe	Strict anaerobe	Microaerophile
+ SOD + Catalase	+ SOD + Catalase	+ SOD – Catalase	– SOD – Catalase	+ SOD +/– Catalase (low levels)

Growth in an environment that have oxygen → Effects microbes respiration + metabolism
↓
may produce Radicals
H_2O_2 / → toxic which kills

Basis of Different Oxygen Sensitivities

- Oxygen easily reduced to toxic reactive oxygen species (ROS)
  - superoxide radical O^-
  - hydrogen peroxide H_2O_2
  - hydroxyl radical OH^-

  toxic → so enzymes detoxifies.

- **Aerobes produce protective enzymes**
  - **superoxide dismutase (SOD)**
  - **catalase** ✓
  - **peroxidase** ✓

Strict Anaerobic Microbes

- All **strict anaerobic microorganisms lack or have very low quantities of**
  - **superoxide dismutase**
  - **catalase**
- These microbes cannot tolerate O2
- Anaerobes must be grown without O2
  - **work station with incubator**
  - **gaspak anaerobic system**

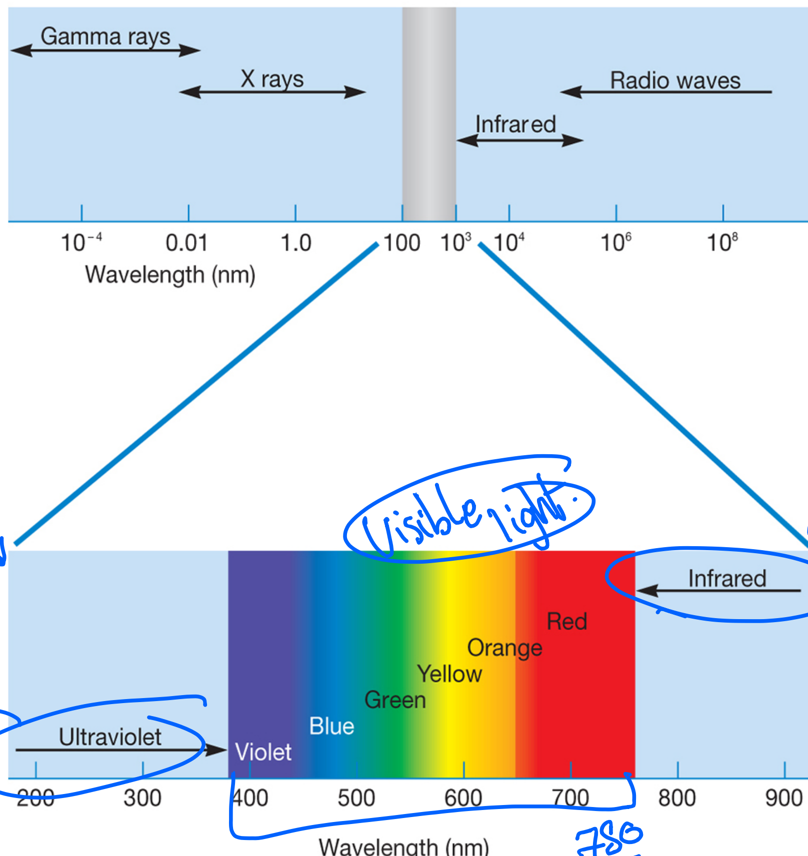
Pressure

Most Microbs lives at 1 atm

- Microbes that live on land and water surface live at 1 atmosphere (atm)
- Some *Bacteria* and *Archaea* live **in deep sea with very high hydrostatic pressures**
- * **Barotolerant** → High Pressure
  - adversely affected by increased pressure, but not as severely as nontolerant organisms
- * **Barophilic** (peizophilic) organisms → extremely High Pressure
  - **require or grow more rapidly in the presence of increased pressure** usually can't live in low Press environ.
  - **change membrane fatty acids to adapt to high pressures**

- The Electromagnetic Spectrum

*Microbs Can die if they Are in direct contact with the Radiation/chemicals
↓
effects Bacterial structures
↓
electromagnetic Radiation



Toxic

Toxic

Penetrate DNA

Radiation Damage

- Ionizing radiation
  - x-rays and gamma rays
  - **mutations death (sterilization)**
  - disrupts chemical structure of many molecules, including DNA
    - **damage may be repaired by DNA repair mechanisms if small dose**
  - *Deinococcus radiodurans*
    - extremely resistant to DNA damage

Radiation Damage…

- Ultraviolet (UV) radiation → Penetrate the cell
  - wavelength most effectively absorbed by DNA is 260 nm
  - **mutations death**
  - causes formation of **thymine dimers** in DNA
  - requires **direct exposure** on microbial surface
  - DNA damage can be repaired by several repair mechanisms

after Absorbtion

5' ATCGATT 3'
3' AATAGCTAA 5'
Complementary strands Bind through H bonds.

UV light if there are 2 T direct the Bond will be formed Between them instead of the Complementary cells

X Rays and Gamma Ray → Kills Microorganisms → mutaion → cell death.

→ Some microbs Are exeption for the Rule → Bacterial endospore

* High dose of Radiation → Sterlization

Radiation Damage…

- Visible light
  - at high intensities generates singlet oxygen (1O2)
    - powerful oxidizing agent
  - carotenoid pigments
    - protect many light-exposed microorganisms from photooxidation

Protection

Microbial Growth in Natural Environments

- Microbial environments are complex, constantly changing, often contain low nutrient concentrations (oligotrophic → environments that have limited nutrients (lower conc. of nutrient) environment) and may expose a microorganism to overlapping gradients of nutrients and environmental factors

* In laboratory all factors can be controlled But in the environment bacteria don't have all the optimal conditions to allow their growth → So microbes can suffer from the lack of nutrients, O_2, Hyperosmotic/Hypo → So they need to survive in these conditions.

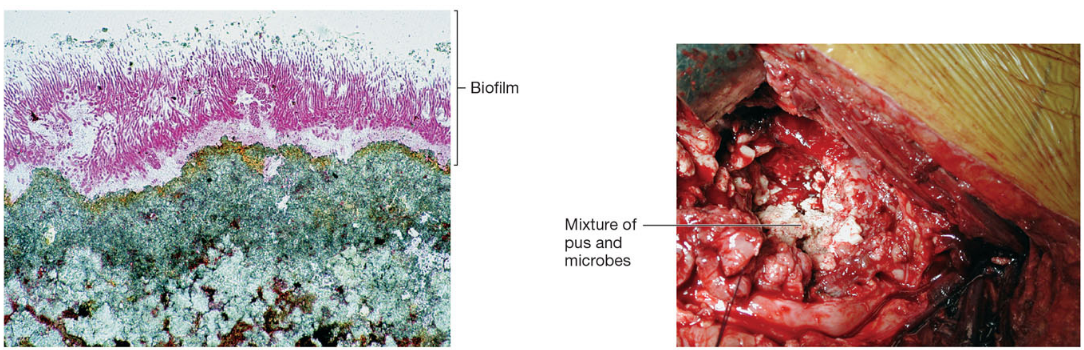
* Microbes in the environment are continously changing → Depending on the factors in the environment.

Biofilms

- Most microbes grow attached to surfaces (**sessile**) rather than free floating (planktonic)
- These attached microbes are members of complex, slime enclosed communities called a **biofilm**
- Biofilms are ubiquitous in nature in water
- Can be formed on any conditioned surface

Many types of microorganisms

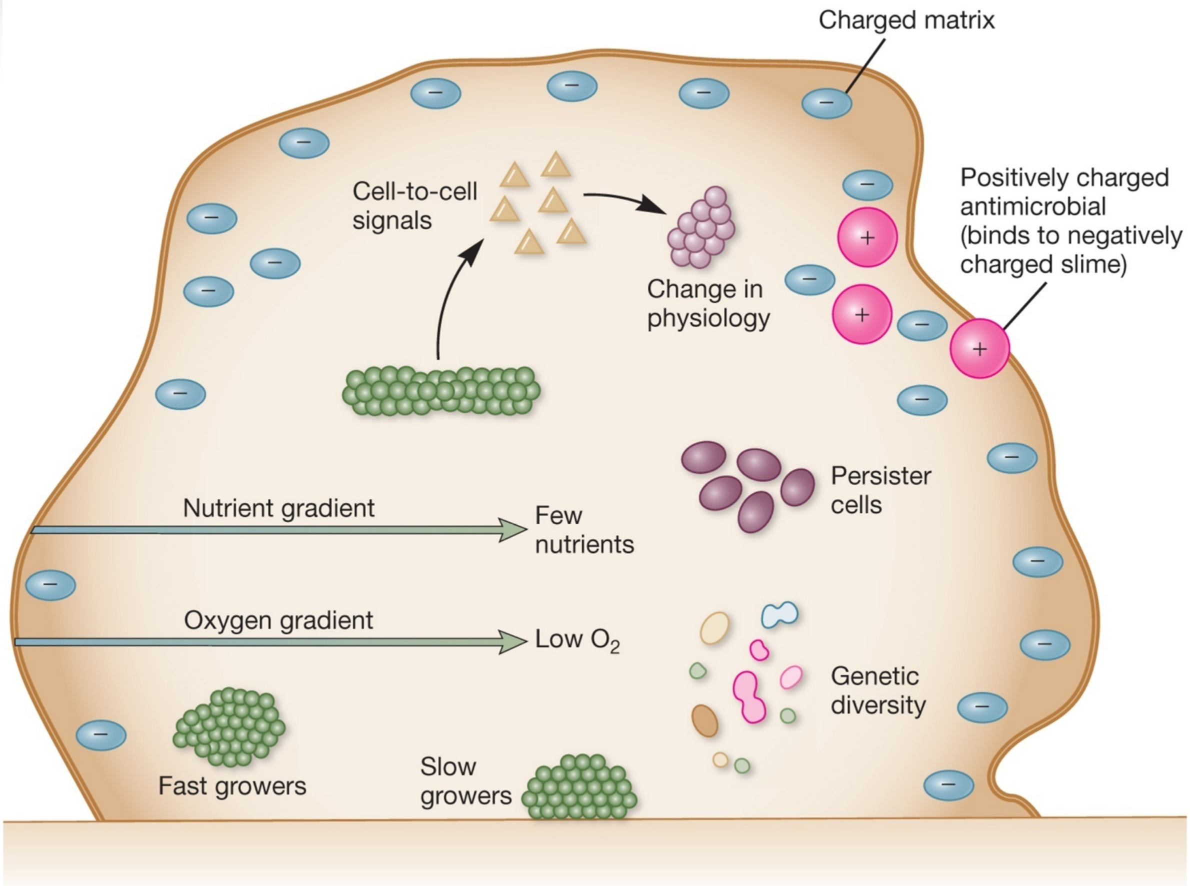
Because of Polysacch which Produce Slime layer makes the microorganism attached.



(a) Biofilm on surface of a stromatolite

(b) Infected tissue after hip replacement

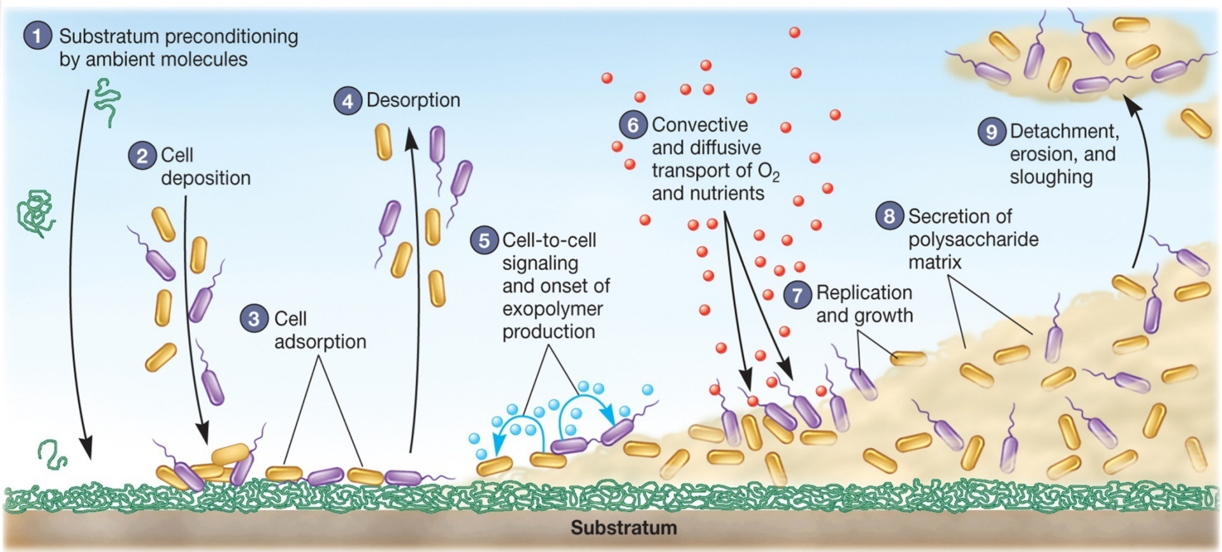
a: Dr. Joachim Reitner; b: Courtesy Nicholas G. Sotereanos, M.D.



Charged matrix
Cell-to-cell signals
Change in physiology
Positively charged antimicrobial (binds to negatively charged slime)
Persister cells
Nutrient gradient
Few nutrients
Oxygen gradient
Low O_2
Genetic diversity
Fast growers
Slow growers

Biofilm Formation

Biofilm may be formed from 1 microorganism.



- ✓ Microbes reversibly attach to conditioned surface and release **polysaccharides, proteins, and DNA** to form the extracellular polymeric substance (EPS)
- ✓ Additional polymers are produced as microbes reproduce and biofilm matures

Biofilms

- a mature biofilm is a complex, **dynamic community of microorganisms**
- **heterogeneity** is differences in metabolic activity and locations of microbes
- interactions occur among the attached organisms
  - exchanges take place metabolically, DNA uptake and communication

Biofilm Microorganisms

- The **EPS** and change in attached organisms' physiology protects microbes from harmful agents
  - UV light, antibiotics, antimicrobials
- **When formed on medical devices, such as implants, often lead to illness**
- **Sloughing off of organisms can result in contamination of water phase above the biofilm such as in a drinking water system**

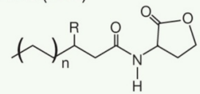
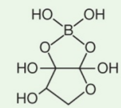
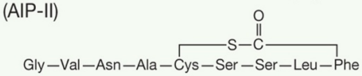
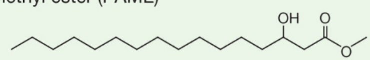
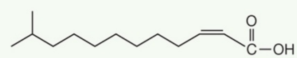
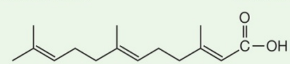
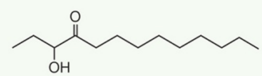
Cell to Cell Communication Within the Microbial Populations

- Bacterial cells in biofilms communicate in a density-dependent manner called **quorum sensing**
- Produce small proteins that increase in concentration as microbes replicate and convert a microbe to a **competent state**
  - DNA uptake occurs, bacteriocins are released

Quorum Sensing

Acylhomoserine lactone (AHL) is an autoinducer molecule produced by many gram-negative organisms

- diffuses across plasma membrane
- once inside the cell, induces expression of target genes regulating a variety of functions

Signal and Structure	Representative Organism	Function Regulated
N-acylhomoserine lactone (AHL) 	*Vibrio fischeri* *Agrobacterium tumefaciens* *Erwinia carotovora* *Pseudomonas aeruginosa* *Burkholderia cepacia*	Bioluminescence Plasmid transfer Virulence and antibiotic production Virulence and biofilm formation Virulence
Furanosylborate (AI-2) 	*Vibrio harveyi*[a]	Bioluminescence
Cyclic thiolactone (AIP-II) 	*Staphylococcus aureus*	Virulence
Hydroxy-palmitic acid methyl ester (PAME) 	*Ralstonia solanacearum*	Virulence
Methyl dodecenoic acid 	*Xanthomonas campestris*	Virulence
Farnesoic acid 	*Candida albicans*	Dimorphic transition and virulence
3-hydroxytridecan-4-one 	*Vibrio cholerae*	Virulence

[a] Other bacteria make a form of AI-2 that lacks boron.

7.8 Continuous culture of microorganisms

1. Distinguish batch culture and continuous culture
2. Differentiate chemostats and turbidostats
3. Discuss the relationship between the dilution rate of a chemostat and population size and growth rate

The Continuous Culture of Microorganisms

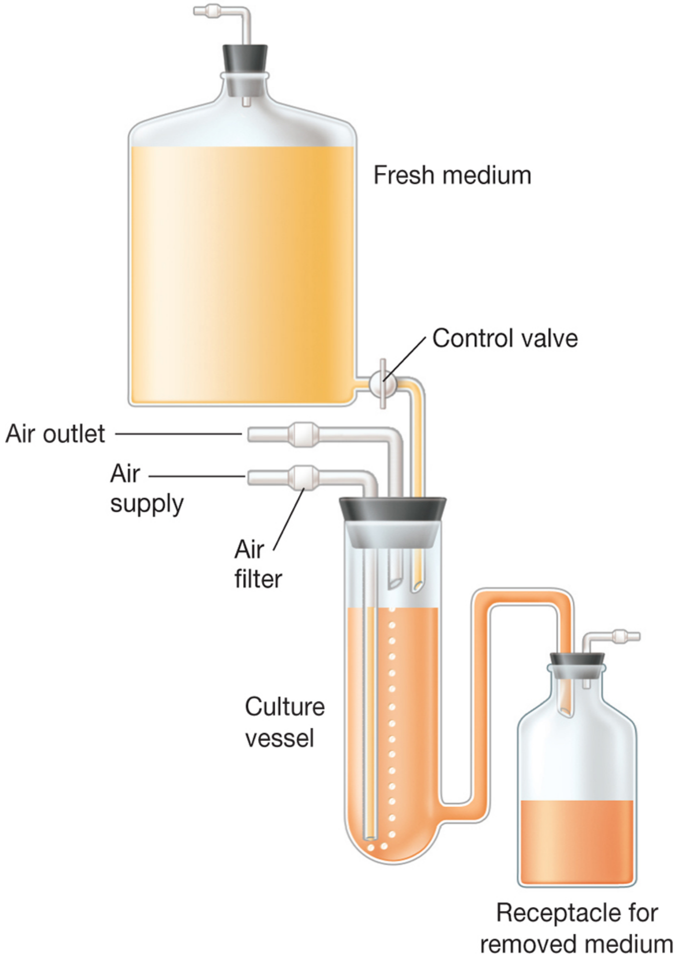
- **Growth in an open system**
  - continual provision of nutrients
  - continual removal of wastes
- **Maintains cells in log phase at a constant biomass concentration for extended periods**
- Achieved using a continuous culture system

Importance of Continuous Culture Methods

- **Constant supply of cells in exponential phase growing at a known rate**
- Study of microbial **growth at very low nutrient concentrations, close to those present in natural environment**
- Study of interactions of microbes under conditions resembling those in aquatic environments
- Food and industrial microbiology

The Chemostat

- Rate of incoming medium = rate of removal of medium from vessel
- An essential nutrient is in limiting quantities



Dilution Rate and Microbial Growth

dilution rate – rate at which medium flows through vessel relative to vessel size

note: cell density maintained at wide range of dilution rates and chemostat operates best at low dilution rate