

Bacterial growth and development of microbial community

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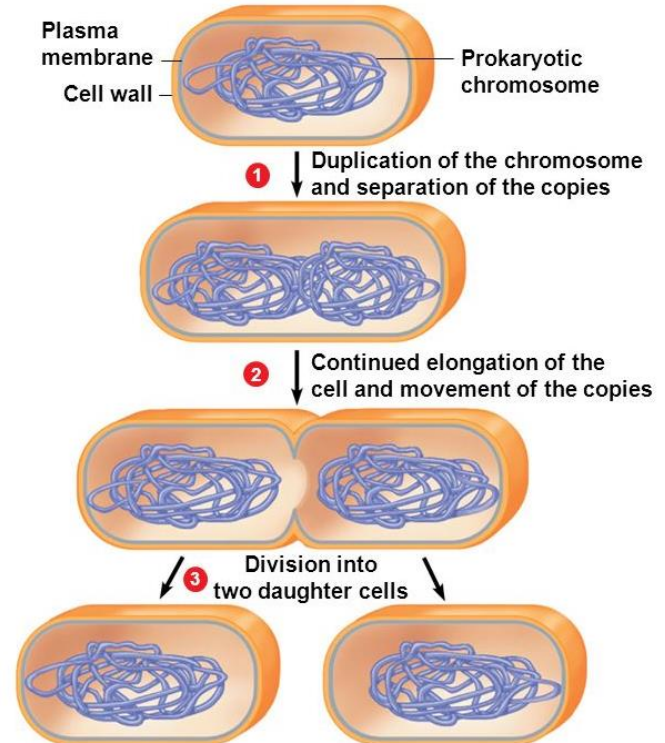
Bacterial growth

Complex process involving numerous anabolic (synthesis of cell components and metabolites) and catabolic (breakdown of cell components and metabolites) reactions.

Generation time

- time required for a cell to divide and form 2 cells
- time required for a population to double
- **from 30 min to >10 days**

Binary fission

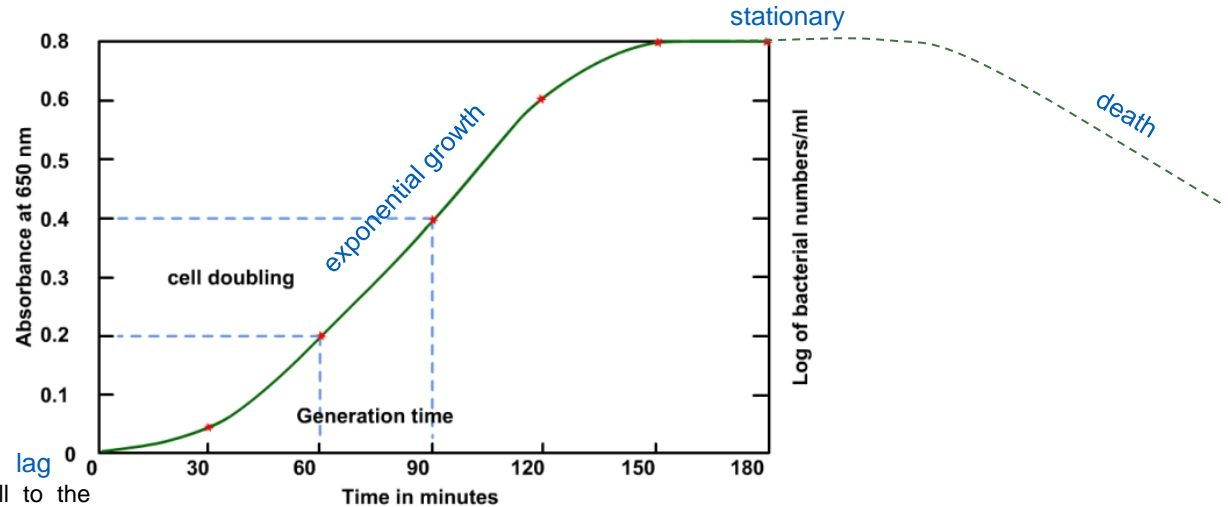


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Bacterial growth

Generation time:

- ✓ Nitrifying bacteria: 8 h (AOBs) /10 hours (NOBs) to several days (opt conditions)
- ✓ Denitrifying bacteria: from several hours to several days
- ✓ Anammox: 7-11 days (T_{opt})



- physiological adaptation of the cell to the new conditions.
- protein synthesis to meet new culture requirements (contaminants)

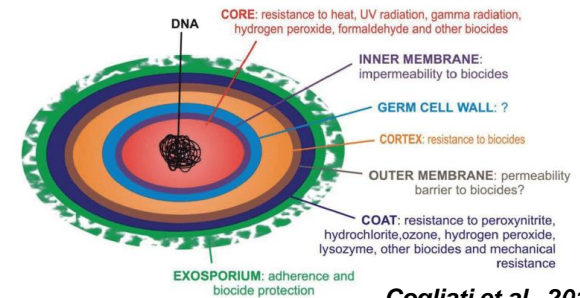
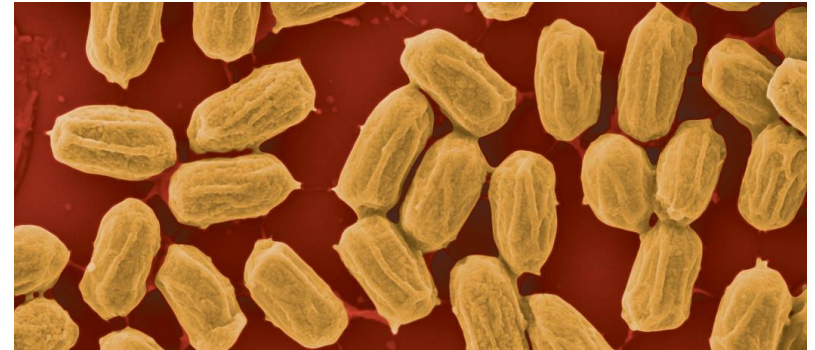
Factors affecting bacterial growth

Physical conditions:

- Temperature
- pH
- Osmotic pressure
- Radiation
- Mechanical & sonic stress

Chemical requirements:

- Nutrients: macronutrients, micronutrients
- Enzymes, organic growth factors
- Oxygen

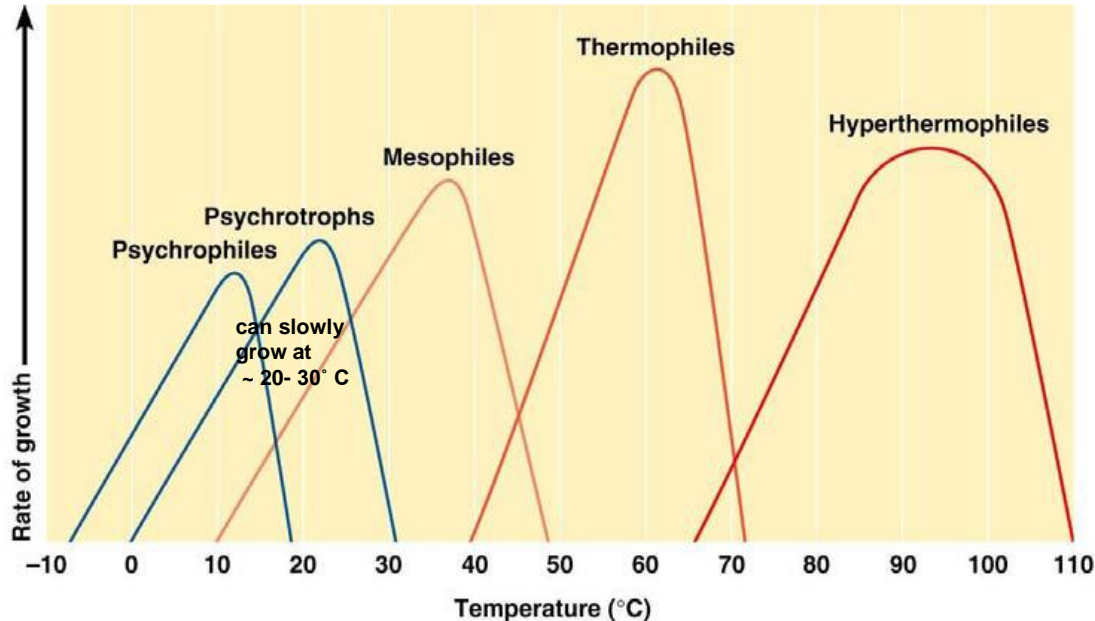


Spores

- ✓ resistant to heat, pressure, radiation and strong acids and bases
- ✓ may survive for many years and decades

Temperature

1. Psychrophiles (“cold- loving”)
2. Mesophiles (“moderateT- loving”)
3. Thermophiles (“heat- loving”)



Minimum: the lowest T that a species will grow
Optimum: best growth T (fastest reproduction)
Maximum: highest T that growth is possible

Nutrients

Macronutrients

- ✓ Carbon (CO₂ or organic compounds)
- ✓ Hydrogen (H₂O or organic compounds)
- ✓ Oxygen (H₂O or organic compounds)
- ✓ Nitrogen (NH₃, NO₃⁻, organic N-compounds)
- ✓ Phosphorus (PO₄³⁻)
- ✓ Sulfur (H₂S, SO₄²⁻, organic compounds)
- ✓ Potassium (K⁺)
- ✓ Magnesium (Mg²⁺, salts)
- ✓ Sodium (Na⁺)
- ✓ Calcium (Ca²⁺, salts)
- ✓ Iron (Fe³⁺, Fe²⁺ or salts)

Micronutrients

- ✓ manganese
- ✓ zinc
- ✓ cobalt
- ✓ molybdenum
- ✓ nickel
- ✓ copper

Growth factors

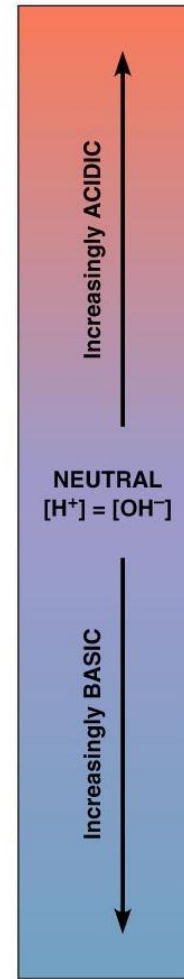
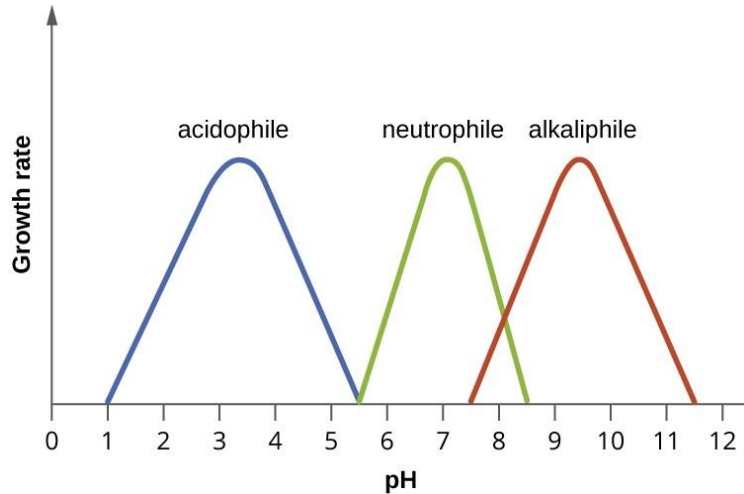
- ✓ Vitamins,
- ✓ amino acids,
- ✓ purines,
- ✓ pyrimidines,
- ✓ folic acid
- ✓ acetate
- ✓ riboflavin

Oligotrophic bacteria

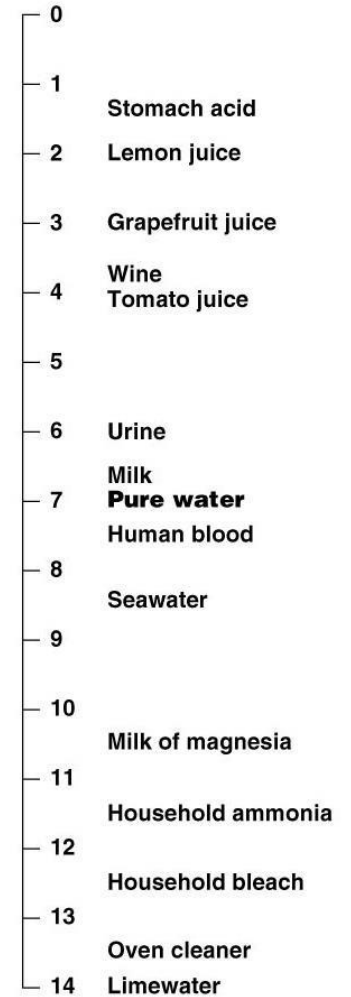
- grow in the environments with extremely low levels of nutrients
- common in drinking water, urine, air etc.
- characterized by slow growth, low rates of metabolism
- alternative energy sources
- different survival strategies

pH

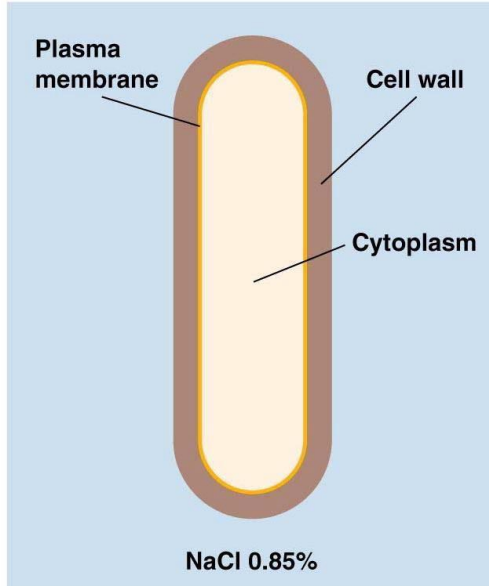
- most bacteria grow in a neutral environment ~ 6.5 - 7.5
- bacteria produce metabolites which may change pH



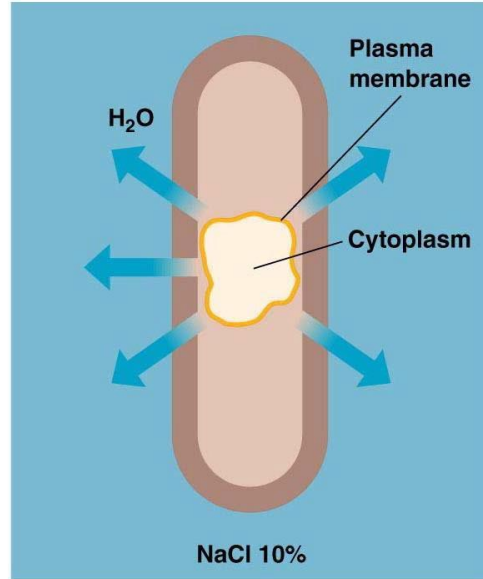
pH scale



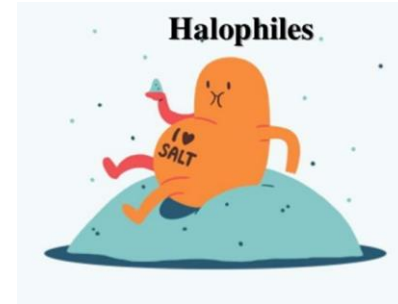
Osmotic pressure



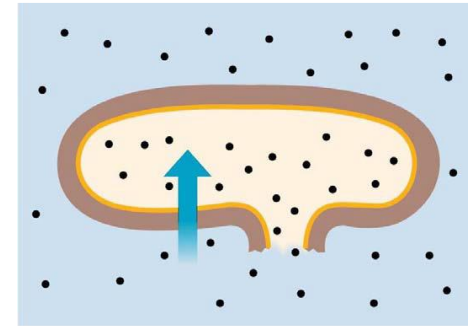
(a) Normal cell in isotonic solution. Under these conditions, the osmotic pressure in the cell is equivalent to a solute concentration of 0.85% sodium chloride (NaCl).



(b) Plasmolyzed cell in hypertonic solution. If the concentration of solutes such as NaCl is higher in the surrounding medium than in the cell (the environment is hypertonic), water tends to leave the cell. Growth of the cell is inhibited.



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(M.phil II)



water moves into the cell and may cause the cell to burst if the wall is weak or damaged (osmotic lysis)

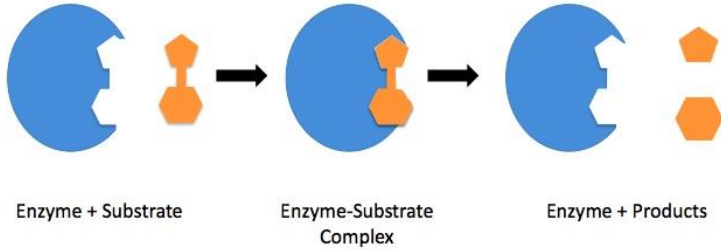
Chemical requirements (media)

- Water (80- 85% weight of cells)
- Carbon (50%)
- Oxygen (20%)
- Nitrogen (14%)
- Hydrogen (8%)
- Phosphorus (3%)
- Sulfur (1%)
- Potassium (1%)
- Sodium (1%)
- Calcium (0.5%)
- Magnesium (0.5%)
- Chlorine (0.5%)
- Iron (0.2%)
- Others (0.3%)

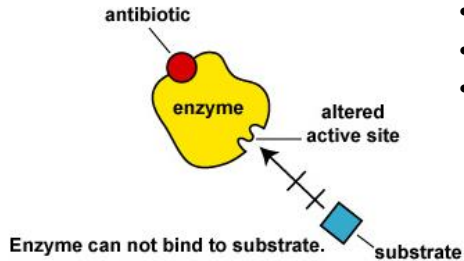


Enzymes

Molecule (proteins, RNA), which speed up chemical reactions

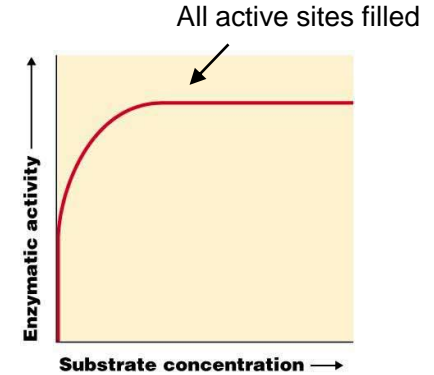
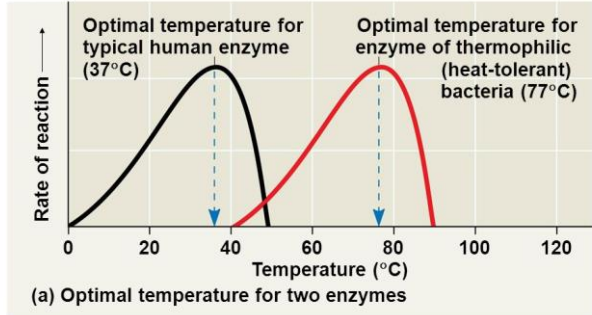


Factors, affecting enzymes activity



- Temperature
- pH
- Concentration of substrate
- Inhibitors

Each enzyme has an optimal temperature in which it can function

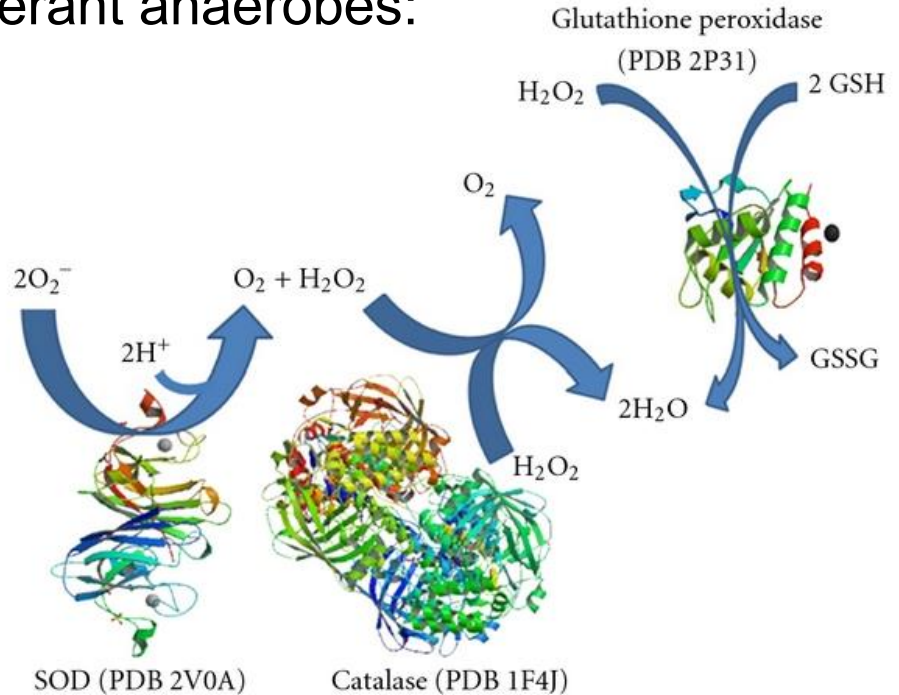


<https://www.youtube.com/watch?v=yk14dOOvwMk&index=17&list=PLTH8ahUlcwvRCscNWDRcD2ZrzBjbrPLt9>

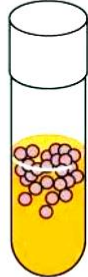
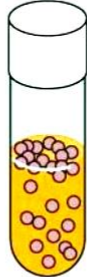



Oxygen tolerance and enzymes

Aerobes and facultative/aerotolerant anaerobes:

- ✓ Catalase
- ✓ Peroxidase
- ✓ Superoxide dismutase (SOD)



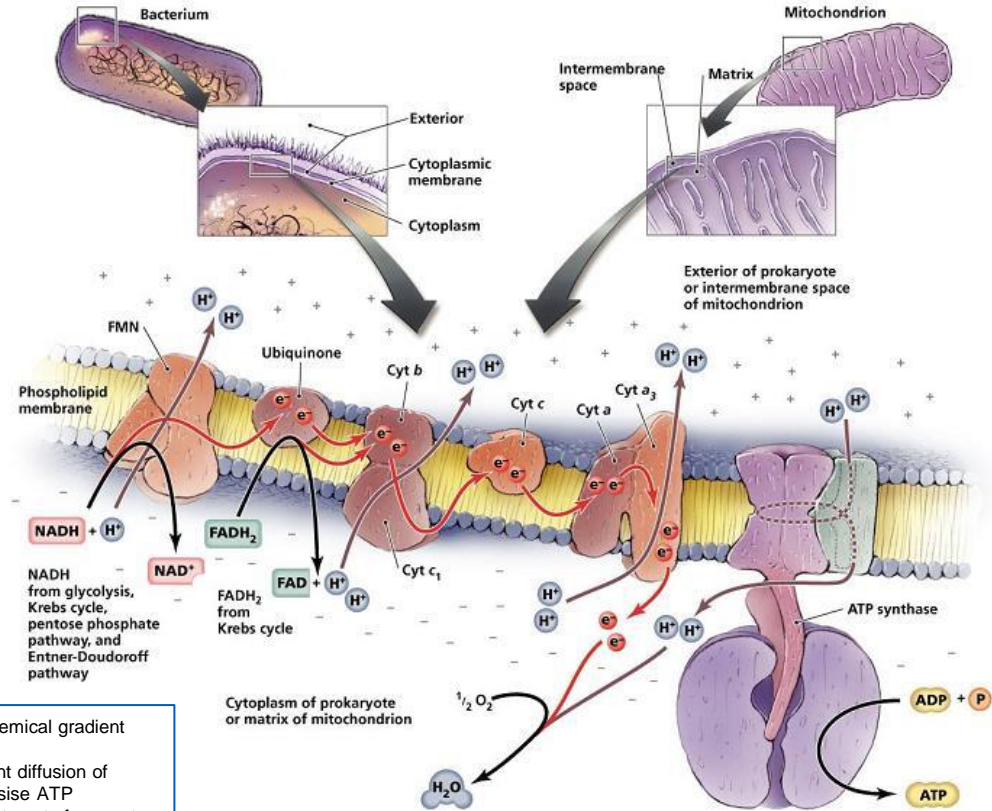
Oxygen

	a. Obligate Aerobes	b. Facultative Anaerobes	c. Obligate Anaerobes	d. Aerotolerant Anaerobes	e. Micro-aerophiles
Effect of Oxygen on Growth	Only aerobic growth; oxygen required.	Both aerobic and anaerobic growth; greater growth in presence of oxygen.	Only anaerobic growth; ceases in presence of oxygen.	Only anaerobic growth; but continues in presence of oxygen.	Only aerobic growth; oxygen required in low concentration.
Bacterial Growth in Tube of Solid Growth Medium					
Explanation of Growth Patterns	Growth occurs only where high concentrations of oxygen have diffused into the medium.	Growth is best where most oxygen is present, but occurs throughout tube.	Growth occurs only where there is no oxygen.	Growth occurs evenly; oxygen has no effect.	Growth occurs only where a low concentration of oxygen has diffused into medium.
Explanation of Oxygen's Effects	Presence of enzymes catalase and superoxide dismutase (SOD) allows toxic forms of oxygen to be neutralized; can use oxygen.	Presence of enzymes catalase and SOD allows toxic forms of oxygen to be neutralized; can use oxygen.	Lacks enzymes to neutralize harmful forms of oxygen; cannot tolerate oxygen.	Presence of one enzyme, SOD, allows harmful forms of oxygen to be partially neutralized; tolerates oxygen.	Produce lethal amounts of toxic forms of oxygen if exposed to normal atmospheric oxygen.

Energy production

Oxidation - Reduction

Energy molecules in cells



1. Proton pumps create an electrochemical gradient (proton motive force)
2. ATP synthase uses the subsequent diffusion of protons (chemiosmosis) to synthesise ATP
3. Oxygen accepts electrons and protons to form water

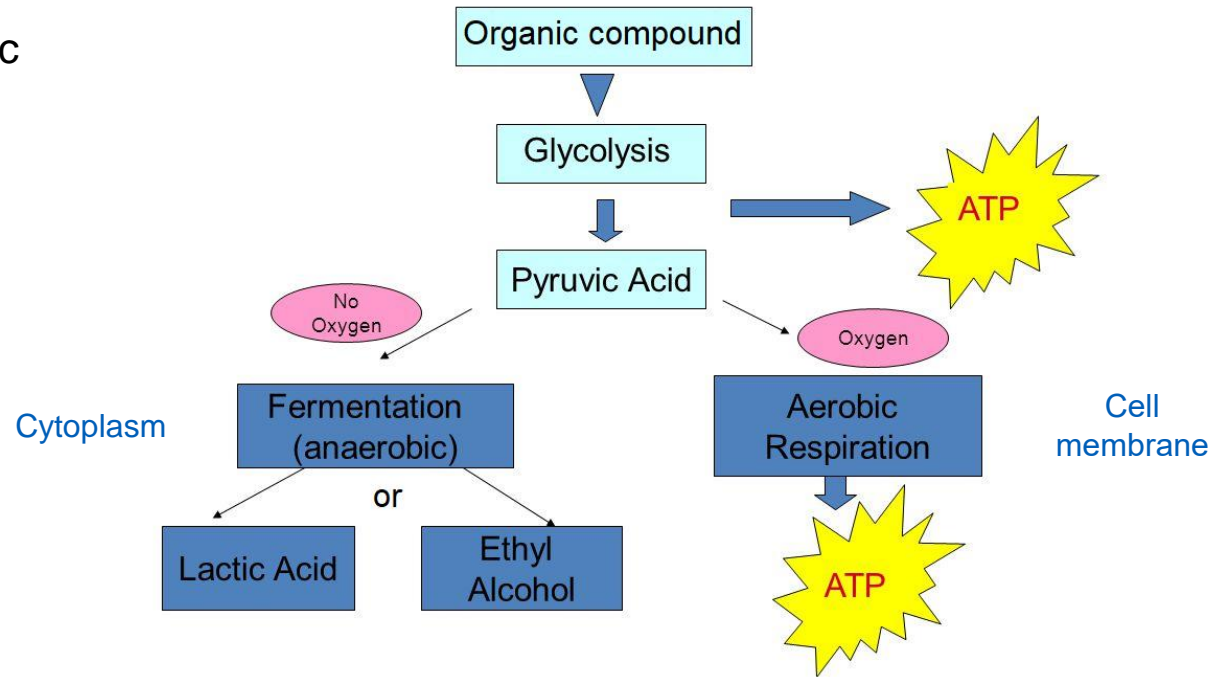
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Carbohydrate Catabolism

Microorganisms oxidize carbohydrates as their primary source of energy

Energy obtained from organic compounds by:

- Respiration
- Fermentation



Aerobic Respiration

- electrons released by oxidation are passed down e- transport chain with oxygen being the final electron acceptor

<u>Process:</u>	<u>Start molecule</u>	<u>End molecule</u>	<u>Energy molecules produced</u>	<u>Waste</u>
Glycolysis	Glucose	Pyruvate	2 NADH 2 ATP	–
Oxidation of pyruvate	Pyruvate	Acetyl-CoA	2 NADH	2 CO ₂
Krebs cycle (citric acid cycle)	Acetyl-CoA	–	6 NADH 2 ATP 2 FADH ₂	4 CO ₂
Electron transport chain/ chemiosmosis	NADH FADH ₂	NAD+ FAD	ATP	Water

Anaerobic Respiration

- organic compounds oxidized, electrons passed down e- transport chain to some molecule other than oxygen (e.g. NO_3 , SO_4) and oxygen **is not** the final electron acceptor:

Nitrate (NO_3^-)  Nitrite (NO_2^-)

Nitrate respiration

Sulfate (SO_4^{2-})  Hydrogen Sulfide (H_2S)

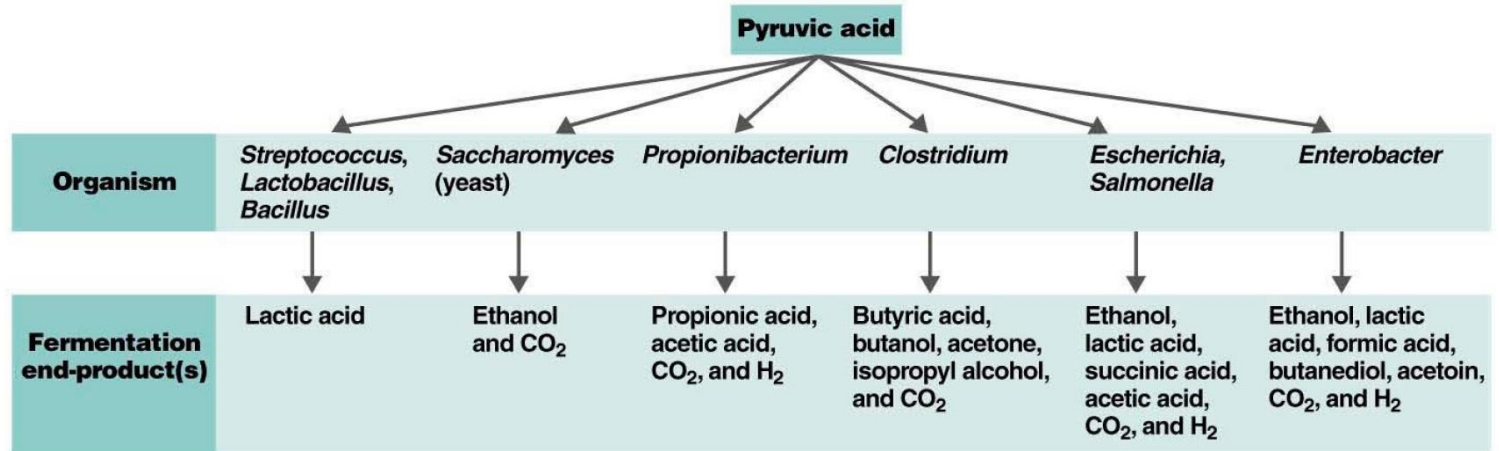
Sulfate respiration

Carbonate (CO_2^{4-})  Methane (CH_4)

Methanogenesis

Fermentation

- anaerobic way to produce ATP
- use organic molecules as their final electron acceptor to produce various end-products

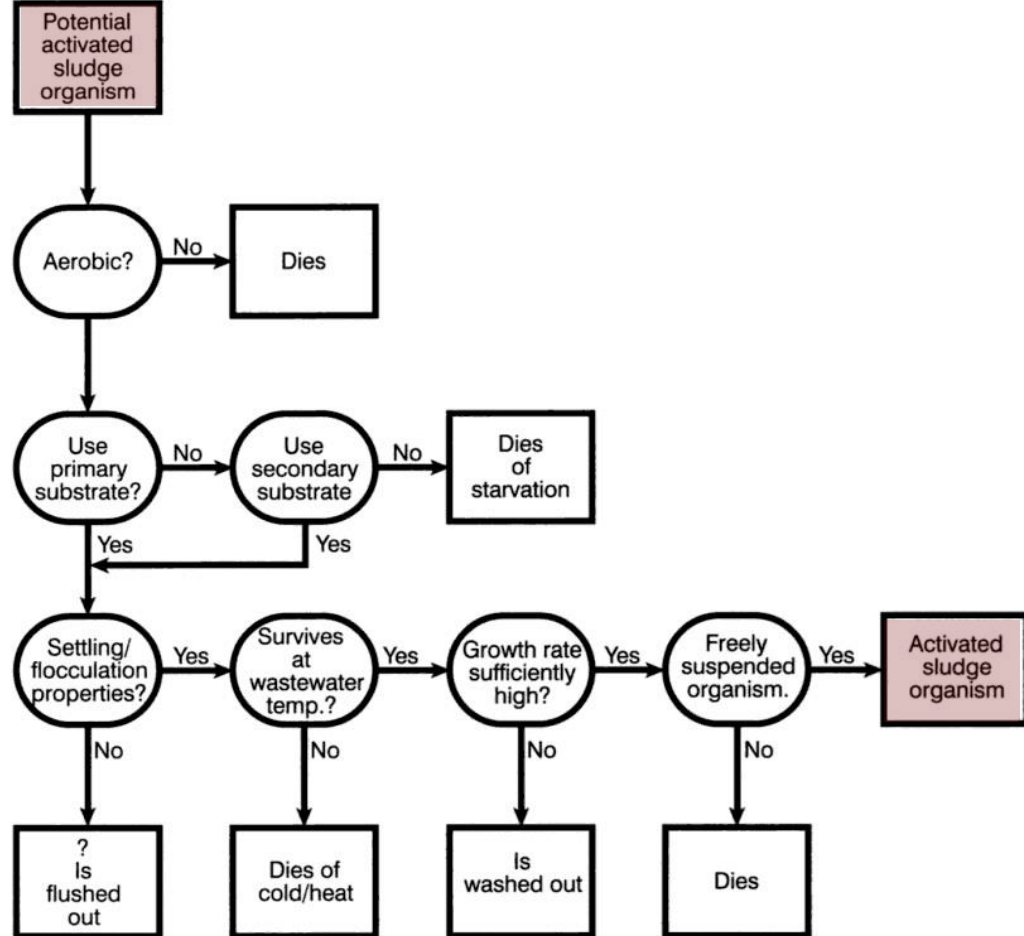
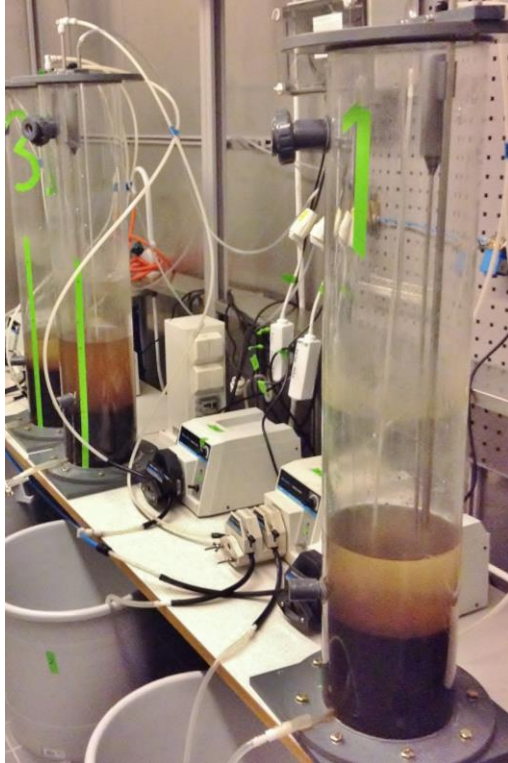


Used for bacterial identification

(b)

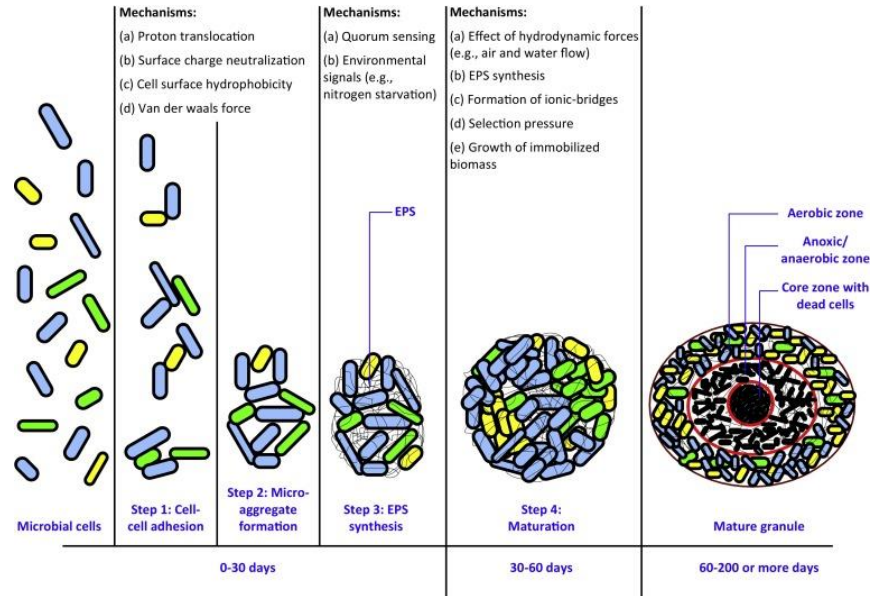
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Selection process for microbial community



Sludge retention time (SRT)

- the time spent by microorganisms in the system / the time available for microorganisms to reproduce – **sludge age**



Group discussion

1. Which of the discussed today parameters can be affected by sludge age?
2. What might happen to bacterial community if sludge age of the process is too short / too long?
3. How may too short/ long sludge age influence removal efficiency of the process? (for example, nitrogen removal or removal of organic pollutants, toxins etc.)