BY : BARJES ALZIARA LECTURE 3: BACTERIAL GROWTH AND NUTRITION

MICROBIOTOGY

### Introduction

- Microbial growth refers to both the increase in cell size and number of cells in a population.
- Importance of understanding bacterial growth:
  - Bacterial survival and transmission
  - In vitro diagnostic (laboratory culture)
  - Cessation of bacterial growth for treatment

مشان نعالج صح لازم نعرف نوع البكتيريا اللي بتسبب المرض نعرف عن طرق بقائها ع قيد الحياة ( العمليات الحيوية ) و انتقالها من كائن لاخر

# Microbial growth/Binary Fission

بتعمل دبل لكل العضيات و بتكبر بعدها بتنقسم لثنتين متماثلتين في كل

Bacteria growth by cell division;

binary or transverse division.

During binary fission the parent cell enlarges, duplicates its chromosomes and forms a central transverse septum that divides the cell into two daughter cells.

Chromosome
Cell wall

Replicated chromosome

Septum

Completed sep

طبعا مش كل البكتيريا الها نفس قدرة الانقسام بنفس المدة في بكتيريا اسرع من بكتيريا ثانية

و الانقسام بدل ع اشي جدا مهم هو انه البكتيريا حية و بتعمل عمليات حيوية مشان هيك بتقدر تتكاثر

### Generation time under optimal conditions



· Generation time is the time it takes for a single cell to grow and divide

> هذول ٤ انواع بكتيريا و مدة انقسام كل نوع فيهم من الحكي بنقدر نعرف انه E coli بدها 12.5 دقيقةً مشان تنقسم اماً مثلا ال Treponema pallidum بدها 30 ساعة

**Organism** 

**Generation Time** 

Escherichia coli

12.5 min

Staphulococcus aureus

27-30 min

Mycobacterium tuberculosis (agent of Tuberculosis)

18 - 24 hrs

Treponema pallidum (agent of Syphilis)

30 hrs

هسه clinically الموضوع هذا بفيدنا نكتشف اذا صاحب العينة مصاب ولا لا يعني لازم نزرع عينة و نشوف اذا تواجد عنا بكتيريا على حسب ال generation time

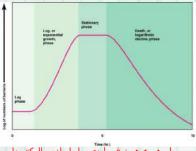
#### The Growth Curve

طبعا هذا الحكي كله ما بيصير بجسم الانسان كله بالمختبر

During lag phase, cells are recovering from a period of no growth and are making macromolecules in preparation for growth

During log phase cultures are growing maximally bacteria multiple at the fastest rate possible under the conditions provided.

حالها للانقسام بانها تصنع انزيمات و جزيئات و بتعمل العمل Bacteria here are(1) susceptible to cell wall active antibiotics.(2) form metabolic end product.



حلو هسه هون في أربع مراحل لنمو البكتيريا الاولى اسمها lag هاى المرحلة بيصير فيها بتجهز البكتيريا replication للعضيات اللي فيها

الثانية log هاي هي مرحلة الانقسام و بهاي المرحلة بكثر عدد البكتيريا بشكل كبير

بتستمر لحد ما يبلش الغذاء ينقص

الثالثة الstationary هون بثبت النمو لانه بسبب قله الغذاء بعض البكتيريا بتصير تموت فتوصل مرحلة ثبات

بين الانقسام و موت البكتيريا اخر مرحلة death مرحلة من اسمها بموت فيها كل البكتيريا سواء من غذاء نقض او من حرارة او ظروف ثانية

- Stationary phase occurs when nutrients are depleted and wastes accumulate (Growth rate = death rate)
- During death phase death rate is greater than growth rate

# Factors Affecting Bacterial Growth

- Temperature
- pH
- Osmotic pressure
- Oxygen
- Nutrition

الحرارة متى ما زادت عن الحد المحتمل راح يحدث denaturation

### Temperature

- Hydrogen bonds will break at high temperatures leads to protein denaturation
- Lipids will be more liquid
- Outside membrane cannot preserve the integrity of the cell and it will disintegrate
- Minimum Temperature: Temperature below which growth ceases, or lowest temperature at which microbes will grow

الحرارة المثالية للبكتيريا مشان تعيش

- Optimum Temperature: Temperature at which its growth rate is the fastest
- Maximum Temperature: Temperature above which growth ceases, or highest temperature at which microbes will grow

### Temperature Classes of Organisms

Psychrophiles ( 0-20°C)

بتحب الحرارة الواطية و بتكون في المحيطات و المناطق الباردة انواع نادرة منها بتصيب الانسان

Cold temperature optima

Most of these microbes are found in the oceans, where the temperature is often 5oC or colder. They can also be found in the Arctic and the Antarctic, living in ice wherever they can find pockets of liquid water.

Listeria monocytogenes

هاي اهم مثال عليها مشكلتها ممكن تعمل تسمم غذائي للشخص يعني اذا حطيت صحن اكل فيه هاى البكتيريا بالثلاجة راح تتكاثر و ممكن تسببلك تسمم

Mesophiles (20 – 45°C)

النوع الثاني امثر واحد بسبب امراض لانه ال optimal temp اله نفس درجة الانسان الباقى ممكن يعمل بس مش قد هاى فهى تعتبر اكثر قروب مهم

Midrange temperature optima

normal human microbiota and pathogens (e.g., E. coli, Salmonella spp., and Lactobacillus spp.) are mesophiles.

Thermophiles(50-80°C)

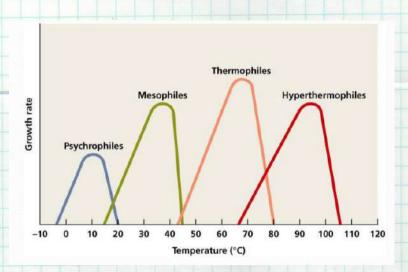
Growth temperature optima between 50°C and 80°C

**Hyperthermophiles** 

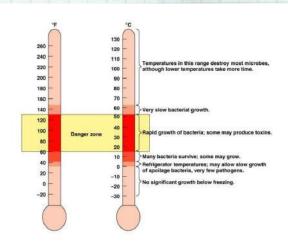
Optima greater than 80°C

These organisms inhabit hot environments including boiling hot springs

## Classification of Microorganisms by Temperature



### Mesophiles



بالنسبة لل mesophiles فاذا رفعنا الحرارة راح تموت بينما اذا خفضناها راح يصيرinhibtion لل growth

### pH and Microbial Growth

#### Each organism has a pH range and a pH optimum

- Acidophiles: Grow optimally between ~pH 0 and
   5.5 (Helicobacter pylori)
- Neutrophiles: Growoptimally between pH 5.5 and 8
- Alkalophiles: Grow optimally between pH 8 11.5
   (Vibrio cholera) نفس الحكي تبع الحرارة ينطبق هون في بكتيريا بتحب الوسط الحمضي في بكتيريا بتفضل المتعادل

Most bacteria grow between pH 6.5 and 7.5

Molds and yeasts grow between pH 5 and 6

Human blood and tissues has pH 7.2+0.2

Lactobacillus bacteria can tolerate acidic environments at pH values 3.5-6.8

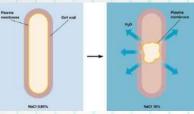
Vibrio cholerae, the pathogenic agent of cholera, grows best at the slightly basic pH of 8.0; it can survive pH values of 11.0 but is inactivated by the acid of the stomach.

### Osmotic Effects on Microbial

Growth

الصورة اليمين البيئة hypertonic اما الشمال hypotonic

- Osmotic pressure depends on the surrounding solute concentration and water availability
- Hypertonic environments, increase salt
   or sugar, cause plasmolysis



حكينا المحاضرة اللي قبل انه البكتيريا hyperosmolar بالنسبة لاجسامنا يعني لو فاتت على جسمنا ممكن يصيرلها swelling طبعا حكينا اللي بحميها من هذا الاشي ال cell wall بس اذا زادت عن الحد ال osmolarity ما راح يقدر يحميها

### Classification:

الدكتور حكا احفظوا هذول التعاريف اعرفوا فرقوا بينهم لانه تعاريفهم متشابهة

هاي احسن بيئة الها اللي فيها كمية مذاب

- Osmophiles: organisms which thrive in high solute
- هاي بتقدر تتحمل البيئة اللي ضغطها الاسموزي عالي بس مش Osmotolerant: organisms which tolerate high solute ضعطها الاسموذي عالي بس مش التقالم ال
- هاي نفس الosmophiles: organisms which thrive in high salt بس osmophiles هاي نفس الosmophiles بسال salt بالتانية
- · Halotolerant: organisms which tolerate high salt
- · Barophiles: organisms which thrive in high pressure
- Barotolerant: organisms which tolerate high pressure

Classification of Organisms Based on O2 Utilization

very important الدكتور حكا عنهم

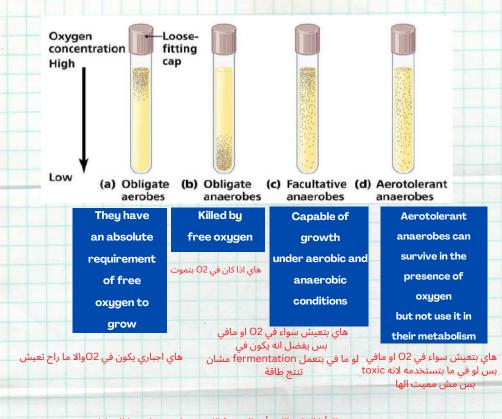
#### Aerobes

- Grow in ambient air, which contains 21% oxygen and a small amount of (0.03%) carbon dioxide.
- Aerobes obtain some of their energy from glycolysis but they get most of the energy through aerobic respiration (via the Krebs cycle and oxidative phosphorylation).
- Aerobes require molecular oxygen as a terminal electron acceptor, so they cannot grow in its absence.

#### Anaerobes

- Anaerobes can not grow in the presence of oxygen
- oxygen is toxic to them.
- Anaerobes do not use free O2
   as their final electron acceptor.
- Instead, they use inorganic oxygen-containing molecules such as nitrate (NO3-), nitrite (NO2-), and sulfate (SO42-), in a process called anaerobic respiration.
- Their metabolism frequently is a fermentative type

بتعيش بالوسط اللي فيه O2 و بتعمل atp عن طريق oxidative phosphorylation بتعيش بالوسط اللي ما فيه O2 و يعتبر toxic الها بتنتج atp عن طريق fermentation



اقرأوا الحكي اللي بأخر الصفحة اللي بعدها بعدين ارجعوا للجداول

### OBLIGATE AEROBES

# THEY HAVE AN ABSOLUTE REQUIREMENT OF FREE OXYGEN TO GROW

#### OBLIGATE ANAEROBES

- KILLED BY FREE OXYGEN
- KILLED NOT BY GASEOUS OXYGEN BUT BY A HIGHLY REACTIVE AND TOXIC FORM OF OXYGEN CALLED SUPEROXIDE AND HYDROGEN PEROXIDE.
- LACK SUPEROXIDE DISMUTASE AND
   CATALASE ENZYMES TO NEUTRALIZE OXYGEN
   FREE RADICALS THUS, THEY SUCCUMB TO THE
   TOXIC EFFECTS OF SUPEROXIDE AND
   HYDROGEN PEROXIDE

#### هذول امثلة

- BACILLUS
- PSEUDOMONAS
- MYCOBACTERIUM

tuberculosis هاي ثاني مرة بحكيها الدكتور اعرفوها احتياط

- ACTINOMYCES
  - BACTEROIDES
  - CLOSTRIDIUM

#### **ABCS**

هذول البكيتريا ممكن يعمللونا infection في المناطق اللي ما بوصلها O2 FACULTATIVE ANAEROBES

- CAPABLE OF GROWTH UNDER AEROBIC AND ANAEROBIC CONDITIONS.
- THEY USE OXYGEN IF IT IS AVAILABLE BUT CAN

FUNCTION WITHOUT IT.

- WHEN OXYGEN IS PRESENT, THEY
  PREFERENTIALLY USE OXYGEN AS A TERMINAL
  ELECTRON ACCEPTOR AND CARRY ON AEROBIC
  METABOLISM
- BUT THEY SHIFT TO ANAEROBIC METABOLISM
  WHEN OXYGEN IS ABSENT

AUREUS

**ENTEROBACTERIACEAE** 

**STAPHYLOCOCCUS** 

AEROTOLERANT ANAEROBES

- THEY ARE ANAEROBIC BACTERIA THAT ARE NOT KILLED BY EXPOSURE TO OXYGEN.
- AEROTOLERANT ANAEROBES CAN SURVIVE IN THE PRESENCE OF OXYGEN BUT NOT USE IT IN THEIR METABOLISM.
- CAPTURES ENERGY BY FERMENTATION, REGARDLESS OF WHETHER THE ENVIRONMENT CONTAINS OXYGEN.

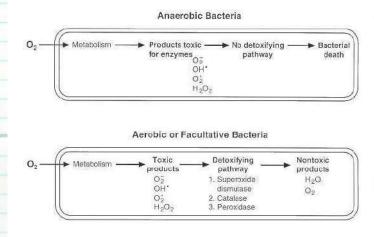
LACTOBACILLUS

تدخل هاي البكتيريا في صناعة الاجبان زي ما قلنا انها بتعمل عملية fermentation

ليش ال O2 يعتبر toxic لبعض البكتيريا و بعضها لا ؟؟ لانه الاوكسجين بس يفوت بيصيرله عمليات ال metabolism هاي العمليات تنتج مادتين (peroxide (H2O2) هذول المواد toxic فاذا ما في نظام يتخلص منهم راح يتراكموا و يسببوا موت الخلية هسه الخلايا من نوع Obligate aerobes او Facultative anaerobes

فيهم نظام اسمه detoxifying system هذا النظام بعمل على تحويل الH2O2 و -O2 ل O2 و H2O عن طريق انزيمين همه ال superoxide dismutase و ال catalase و برضو بيصير هذا الحكي بخلايا الانسان اما ال Obligate anaerobes و Aerotolerant anaerobes ما عندهم هذا النظام فبتراكم عندهم المواد السامة و بعمللها تسمم





- Utilization of O2 during metabolism yields toxic by-products including
   O2-, singlet oxygen (102) and/or H2O2
- Toxic O2 products can be converted to harmless substances if the organism has catalase (or peroxidase) and superoxide dismutase (SOD)
- SOD converts O2- into H2O2 and O2
- Catalase breaks down H2O2 into H2O and O2
- Any organism that can live in or requires O2 has SOD and catalase (peroxidase)

### Oxygen and Microbial Growth

Using oxygen (O2) in metabolism creates toxic waste

Microbes that are able to use aerobic respiration produce enzymes

to

detoxify oxygen:

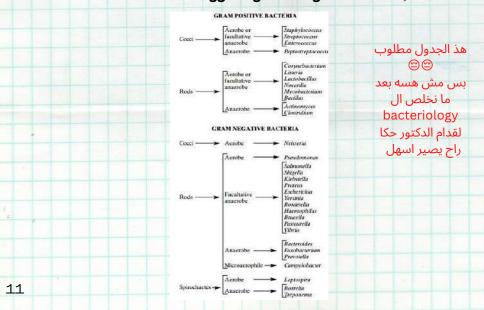
Catalase: H2O2 ---> H2O and O2

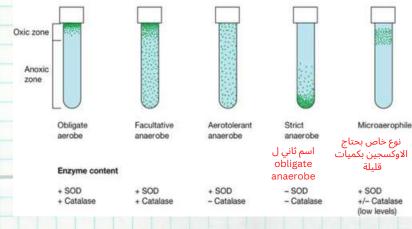
Superoxide dismutase (SOD): oxygen radical ---> H2O2 and O2

Microbes that don't make these enzymes cannot exist in the presence of oxygen.

The enzyme superoxide dismutase reduces the superoxide to oxygen gas and hydrogen peroxide (H2O2).

Subsequently, the toxic hydrogen peroxide generated in this reaction is converted to water and oxygen by the enzyme catalase,





- Obligate aerobes and most facultative anaerobes have both superoxide dismutase and catalase.
- Some facultative and aerotolerant anaerobes have superoxide dismutase but lack catalase.
- · Most obligate anaerobes lack both enzymes.

#### CAPNOPHILES

CAPNOPHILIC (OR CARBON DIOXIDE-LOVING)
BACTERIA REQUIRE INCREASED CONCENTRATION
OF CARBON DIOXIDE (5% – 10%) AND
APPROXIMATELY 15% OXYGEN.

THIS CONDITION CAN BE ACHIEVED BY A CANDLE JAR (3% CARBON DIOXIDE) OR CARBON DIOXIDE INCUBATOR, JAR, OR BAG.

HAEMOPHILUS INFLUENZAE, NEISSERIA GONORRHEA





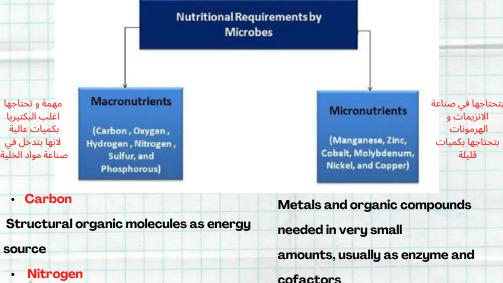
Neisseria gonorrhea

تسبب احد الامراض المنقولة جنسيا لدى الرجال Haemophilus influenzae بتسبب عدوى في الاذن الوسطى و التهاب في الجيوب

#### Microbial Nutrition

Microbial Nutrit

- Organisms use a variety of nutrients for:
- 1- their energy needs
  - 2- to build organic molecules & cellular structures
- Nutrients are those substances that are energy sources to drive the metabolic or biosynthetic processes by the microbial cells



13

In amino acids, proteins

In amino acids, thiamine, biotin

In DNA, RNA, ATP, and membranes

Sulfur

**Phosphorus** 

### Special requirements

هذا الجدول مراجعة لحالات البكتيريا مع الاكسجين و كل نوع راح نوخذ عنه لقدام

- Amino acids
- Nucleotide bases
- Enzymatic cofactors or "vitamins"

Classification	Characteristics	Examples
Aerobes	grow in ambient air, which contains 21% oxygen and a small amount of (0.03%) carbon dioxide.	Bacillus
	Aerobes obtain some of their energy from glycolysis but they get most of the energy	
	<ul> <li>through aerobic respiration (via the Krebs cycle and oxidative phosphorylation).</li> <li>Aerobes require molecular oxygen as a terminal electron acceptor, so they cannot grow in its absence.</li> </ul>	
Obligate aerobes	They have an absolute requirement of free oxygen to grow	Bacillus, Pseudomonas, Mycobacterium
Anaerobes	<ul> <li>Anaerobes can not grow in the presence of oxygen oxygen is toxic to them.</li> <li>Anaerobes do not use free O2 as their final electron acceptor.</li> <li>Instead, they use inorganic oxygen-containing molecules such as nitrate (NO3-), nitrite (NO2-), and sulfate (SO<sub>4</sub><sup>2</sup>-), in a process called anaerobic respiration.</li> <li>their metabolism frequently is a fermentative type</li> </ul>	
Obligate anaerobes	killed by free oxygen     Obligate anaerobes are killed not by gaseous oxygen but by a highly reactive and toxic form of oxygen called superoxide and hydrogen peroxide.     obligate anaerobes lack superoxide dismutase and catalase enzymes to neutralize oxygen free radicals thus, they succumb to the toxic effects of superoxide and hydrogen peroxide.	Actinomyces* Bacteroides Clostridium ABCs
Facultative anaerobes	Facultative anaerobes are versatile organisms capable of growth under aerobic and anaerobic conditions.     They use oxygen if it is available but can function without it.     When oxygen is present, they preferentially use oxygen as a terminal electron acceptor and carry on aerobic metabolism, but they shift to anaerobic metabolism when oxygen is absent	e.g., Enterobacteriaceae family, Staphylococcus aureus, etc
Aerotolerant anaerobes	They are anaerobic bacteria that are not killed by exposure to oxygen. Aerotolerant anaerobes can survive in the presence of oxygen but not use it in their metabolism.	Lactobacillus, for example, always captures energy by fermentation, regardless of whether the environment contains oxygen.
Capnophiles	Capnophilic (or carbon dioxide—loving) bacteria require increased concentration of carbon dioxide (5% – 10%) and approximately 15% oxygen.     This condition can be achieved by a candle jar (3% carbon dioxide) or carbon dioxide incubator, jar, or bag.	Haemophilus influenzae, Neisseria gonorrhea
Microaerophiles	Requires low but not full oxygen tension     Higher oxygen tensions may be inhibitory to them.	Campylobacter jejuni, Helicobacter pylori