

WJEC (Eduqas) Biology A-level

Topic 1.4 - Microbiology

Flashcards

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What are bacterial cell walls made up of?



What are bacterial cell walls made up of?

A three-dimensional mesh of peptidoglycan (murein), a polymer of amino acids and sugars.



What is Gram staining?



What is Gram staining?

A technique used to differentiate between Gram negative and Gram positive bacteria.



Outline the process of Gram staining.



Outline the process of Gram staining.

1. Stain culture with **crystal violet**. Remove and rinse with water
2. Add **iodine solution** and rinse after 1 minute
3. Alternate washes of **alcohol** and water for 30 seconds
4. Counterstain with **red safranin** for 1 minute
5. Dry and examine sample under microscope



Define Gram positive bacteria.



Define Gram positive bacteria.

Bacteria that have a **thick peptidoglycan wall** and a **purple** appearance following gram staining.



Why do Gram positive bacteria appear purple following Gram staining?



Why do Gram positive bacteria appear purple following Gram staining?

The thick peptidoglycan wall retains crystal violet when rinsed with alcohol.



Define Gram negative bacteria.



Define Gram negative bacteria.

Bacteria that have a **thin peptidoglycan wall** with an outer **lipopolysaccharide** membrane and a **red** appearance following gram staining.



Why do Gram negative bacteria appear red following Gram staining?



Why do Gram negative bacteria appear red following Gram staining?

On treatment with alcohol, the lipopolysaccharide layer is lost and the crystal violet washes away. The counterstain safranin stains the thin peptidoglycan layer red.



What is an obligate aerobe?



What is an obligate aerobe?

An organism that requires oxygen for metabolism.



What is an obligate anaerobe?



What is an obligate anaerobe?

An organism that can only survive in environments which lack oxygen.



Define facultative anaerobe



Define facultative anaerobe

- An organism that normally respire aerobically
- It is capable of switching to anaerobic respiration in the absence of oxygen



What are aseptic techniques?



What are aseptic techniques?

A range of techniques used to culture microorganisms under sterile conditions in order to minimise contamination.



List the basic aseptic techniques.



List the basic aseptic techniques.

- Wipe surfaces with antibacterial cleaner
- Set up Bunsen burner nearby - convection currents prevent microbes from entering culture
- Flame inoculating loop and neck of bottles before use
- Minimise time that vessels containing bacteria are open
- Sterilise all equipment e.g. use of an autoclave
- Wear protective clothing
- When inoculating a petri dish ensure that the lid is held at an angle to prevent microbes from the air contaminating the agar
- Hold test tubes at an angle to prevent microbes entering



Outline how to culture microorganisms.



Outline how to culture microorganisms.

- Sterilise inoculating loop in bunsen flame
- Lift dish lid and keep it at an angle
- Transfer bacteria to an agar plate using a sterile inoculating loop or pipette
- Tape on lid at two ends, invert dish and incubate
- In the school laboratory, ensure dish is not airtight and do not incubate above 25°C to avoid growth of pathogens



Explain the difference between a spread plate and a streak plate.



Explain the difference between a spread plate and a streak plate.

- **Spread plate** - microorganisms distributed evenly with a sterile spreader
- **Streak plate** - aims to obtain single colonies by rotating the plate to build layers of the culture on at least three separate streaks



What is nutrient media?



What is nutrient media?

A solid or liquid nutrient-rich medium used in the cultivation of microorganisms.



Describe the composition of nutrient media.



Describe the composition of nutrient media.

Contains a carbon source, nitrogen source, water and growth factors (e.g. salts and vitamins).



Describe the conditions used when culturing microorganisms.



Describe the conditions used when culturing microorganisms.

- Optimum temperature
- Constant pH
- Nutrient supply
- Aerobic conditions



What is the difference between total cell count and viable cell counts?



What is the difference between total cell count and viable cell counts?

In a given area or volume, total cell count is the total number of cells (both living **and** dead) whereas viable cell count is the total number of **living** cells.



Describe how a viable cell count is conducted.



Describe how a viable cell count is conducted.

- Add a known volume of organisms to an agar plate
- Incubate the plate
- Count the number of colonies



What is assumed when conducting a viable cell count?



What is assumed when conducting a viable cell count?

It is assumed that one cell gives rise to a single colony.



What is the problem with the 'one cell
one colony' assumption?



What is the problem with the 'one cell one colony' assumption?

It does not account for clumping of cells in the original inoculum. This may result in a lower estimate of the number of cells



What is a serial dilution?



What is a serial dilution?

A sequence of dilutions, in which the dilution factor is constant, used to dilute a stock solution.

