AS and A-level Biology practicals: Equipment set up

Practical 6: Use of aseptic techniques to investigate the effect of antimicrobial substances on microbial growth

Inoculating broth from slopes

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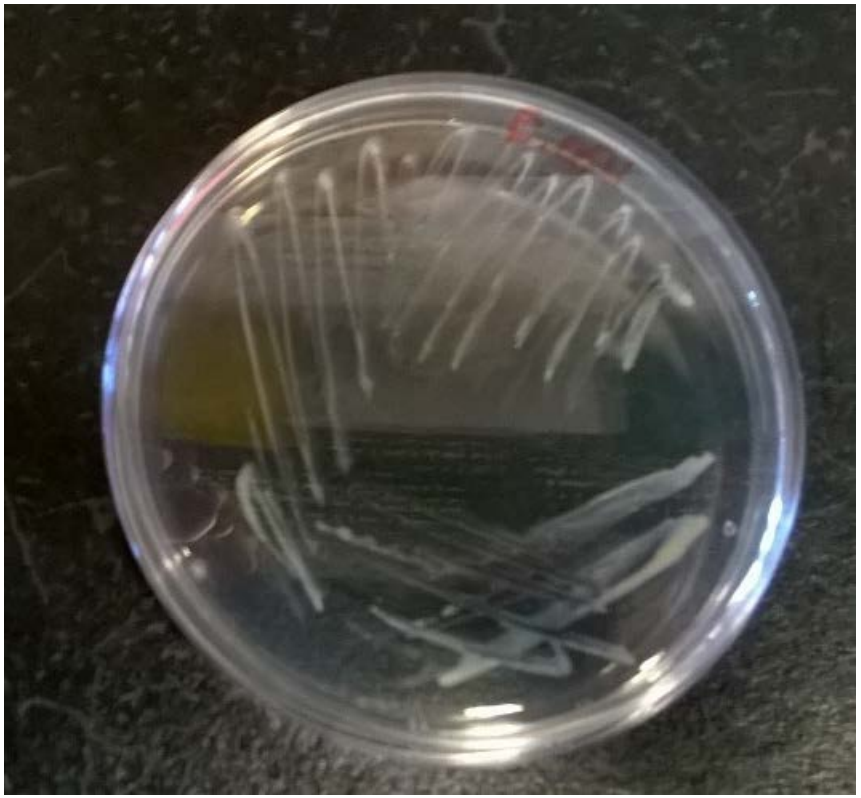
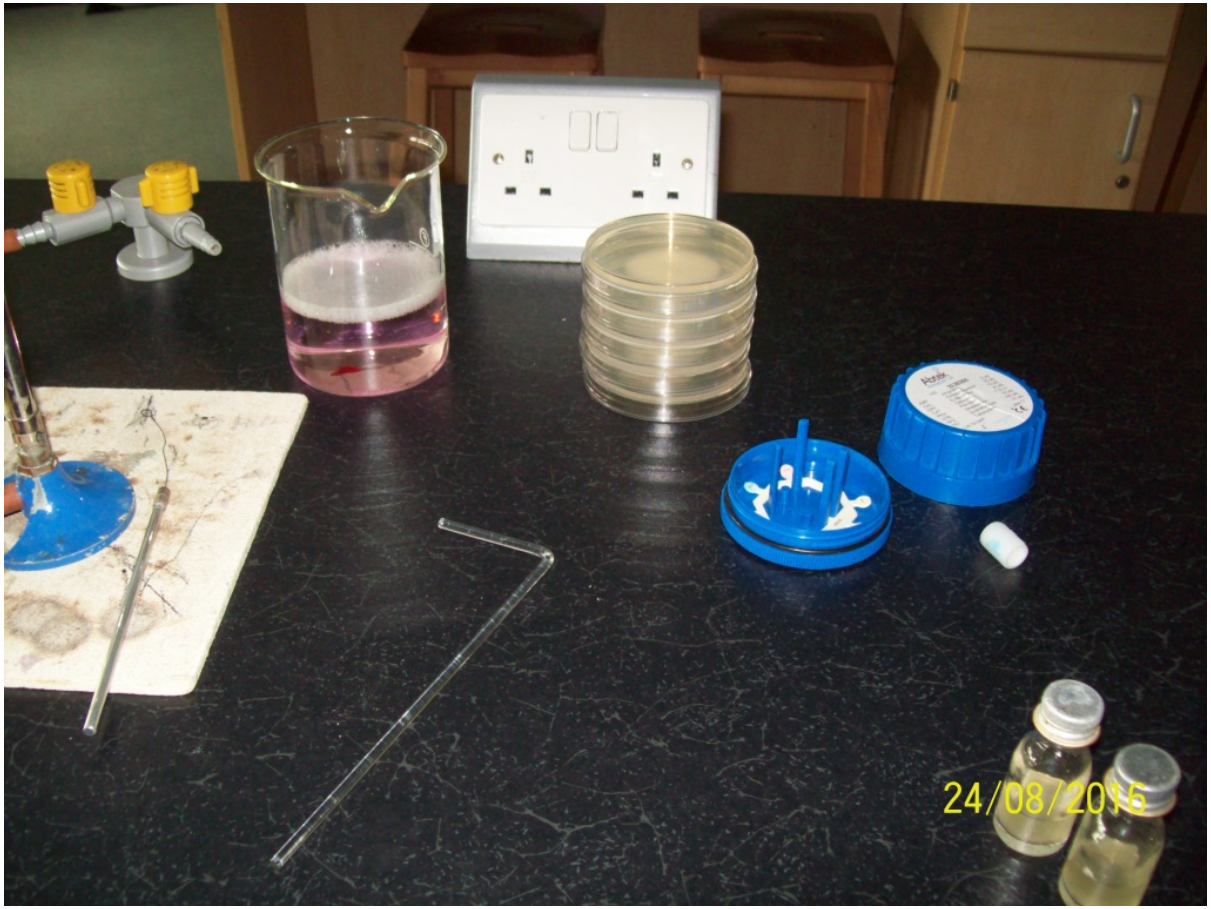
- For student use the main (mother) culture needs to be sub cultured into broth.
- Either it can be prepared directly if only a few bottles of broth are need otherwise it is better to subculture first onto nutrient agar plates so the mother culture is kept for future use.
- Grow the plates for 24-48 hours and use the plates to then subculture into broth for students to make lawns.

Sub cultured colonies on nutrient agar plates

- Streak the bacteria for single colonies.
- This method also checks the purity of the original culture.



Antimicrobial discs



Internal structure

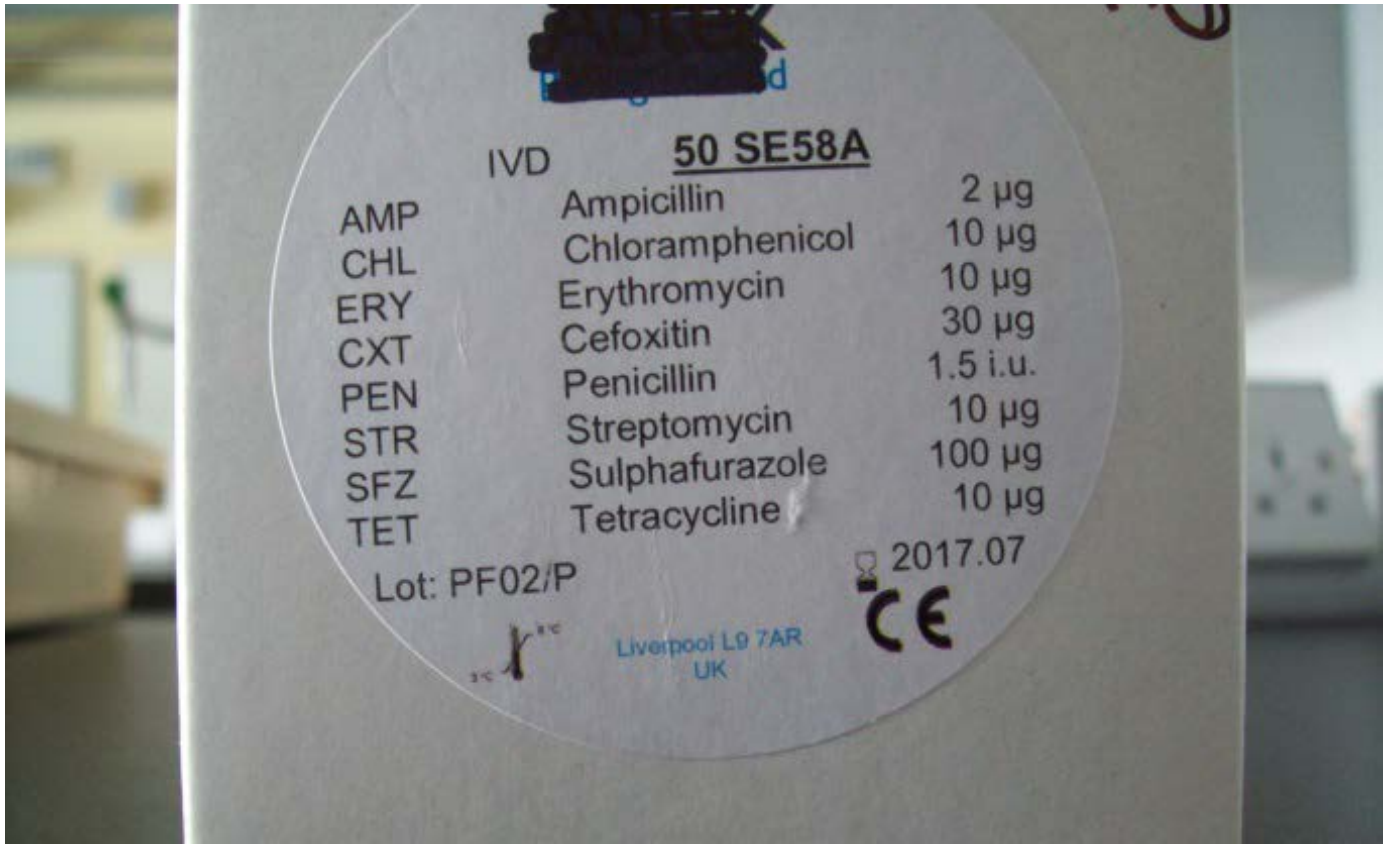
- Equipment for inoculating plates and adding antibiotic rings.
- Glass spreaders can be made from glass tubing with a bend put into them.
- All equipment must be sterilised before use.
- Spreaders and pipettes should be wrapped in foil or non-absorbent paper before sterilising in an autoclave or oven.
- Plastic A4 wallets can be cut into single sheets and soaked in virkon and dried to give a sterile working area.

Antimicrobial discs on nutrient agar plates

Antimicrobial discs on nutrient agar plates

- Plates are inoculated with bacteria and the discs added immediately.
- The plates are then incubated.

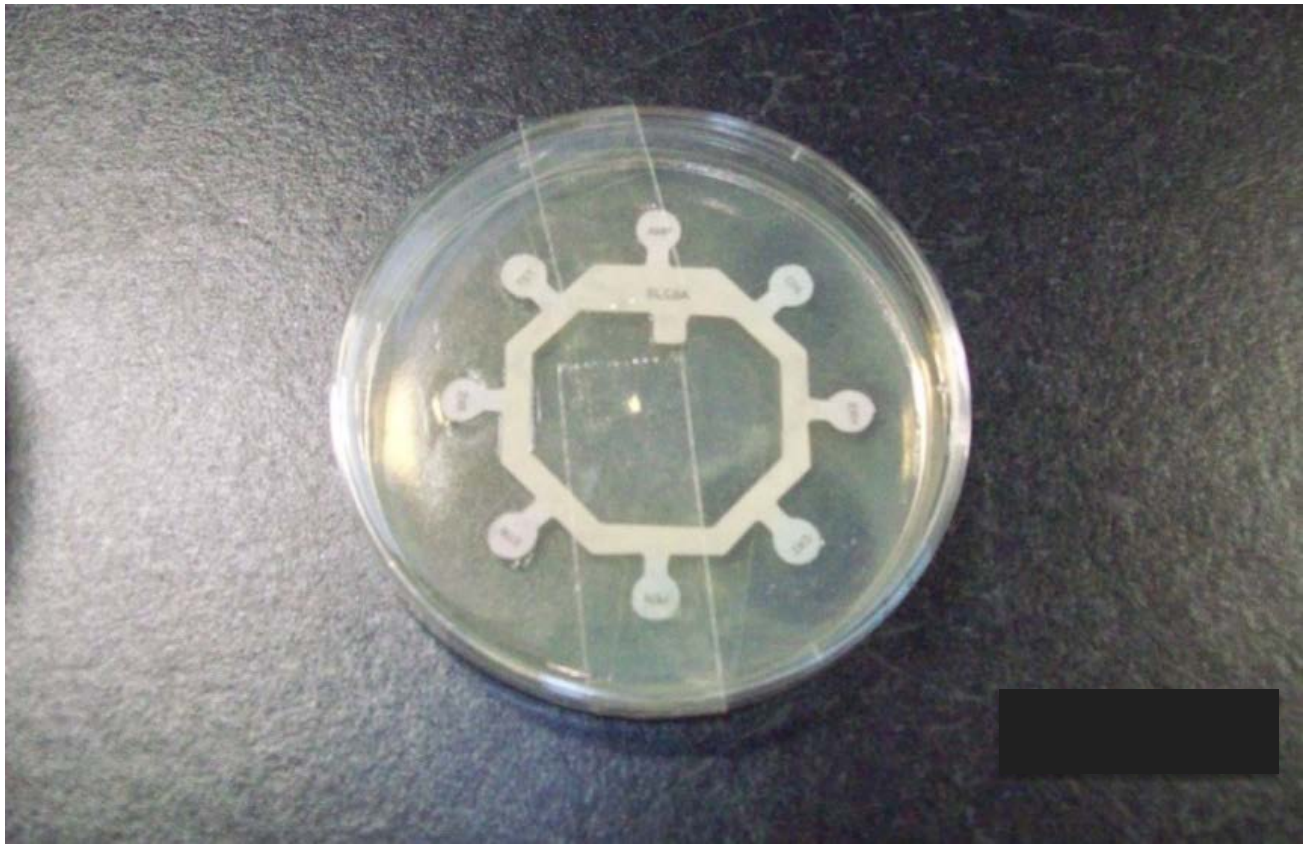
Key for discs



Key for discs

- The keys for the individual antibiotics are found on the box.
- Do not throw the box away if the discs are used during the practical as students need to refer to the key to identify the antibiotics after incubation.

Typical results (E.coli K12)



Typical results (E.coli K12)

- Zones of clearing can be seen around some of the discs.
- All items must be disposed of safely by autoclaving and plates must not be opened after incubation.
- Zones can be measured thorough the base of the plates.