

## Comparing the antibacterial properties of metals

### Practical sheet 2 – Investigating the antibacterial properties of metals

#### Equipment and materials

- 1 cm x 1 cm pieces of metal e.g. copper, zinc, stainless steel, aluminium
- Universal bottles containing 5 cm<sup>3</sup> *Staphylococcus albus* bacteria in nutrient broth (broth seeded with the bacteria)
- sterile Petri dishes containing nutrient agar
- sterile forceps
- inoculation loop
- Bunsen burner and heatproof mat
- marker pen
- tape to secure Petri dishes

#### Method

1. Label the bottles of *Staphylococcus albus* broth cultures with the types of each piece of metal and one bottle 'control'.
2. Working close to the hot flame of a Bunsen burner, use sterile forceps to transfer each piece of metal to the labelled bottles of *Staphylococcus albus* broth cultures and replace the cap. No metal is added to the 'control' bottle. Shake each bottle and leave it for at least 48 hours at 25 °C.
3. Label the bases of the sterile Petri dishes of nutrient agar at the edge so that you know which type of metal will be placed in each one. Label one Petri dish 'control'.
4. Sterilise the inoculation loop in the Bunsen burner flame and allow it to cool in the air for 5 seconds.
5. Open the first of the bottles of metal pieces in bacterial broth. Hold the cap in your hand and flame the neck of the bottle. Insert the inoculation loop into the broth, remove it and replace the cap.
6. Working close to the Bunsen burner, lift the lid of the first Petri dish of nutrient agar just enough to give access (hold it horizontally above the agar). Smear the bacterial broth on the loop onto the agar surface at the left-hand side of the dish. Flame the loop again, let it cool and then repeatedly streak the bacteria from the starting point to dilute them, as shown in the diagram. Replace the lid of the Petri dish and re flame the loop.



- Repeat steps 4 to 6 using the remaining bottles of metal pieces in broth, including the control broth.
- Secure the lids of the plates to their bases by using 4 short strips of adhesive tape.
- Turn the Petri dishes over (to prevent condensation dripping onto the agar surface) and incubate the plates at 25 °C for 48 hours.
- Observe the plates carefully but do NOT open them.



## Questions

- Why were the bottles of bacterial broth and pieces of metal left at 25 °C for 48 hours?
- Why is a 'control' prepared?
- Why are the Petri dishes of agar incubated at 25 °C?
- Why are the Petri dishes taped closed before incubation?
  - Is there any difference between the number of bacterial colonies present on each plate?
  - Suggest a reason for any difference.