

## Instructions for preparing killed cell suspensions and submission of samples to the European Meningococcal MLST Centre (EMMC)

Please follow the protocol below to prepare boiled cell suspensions, suitable for PCR, for submission to the EMMC. Good microbiological practice should be followed throughout.

### Requirements

- freshly grown plates of meningococcal cultures
- boiling water bath
- sterile phosphate buffered saline (PBS)
- 1.5 - 2.0 ml screw-capped Eppendorf tubes (not double-walled tubes)
- sterile swabs/loops
- Nesco/Para film (sealing film) or equivalent

### Protocol

1. Heat the water bath until it boils.
2. Clearly label the screw-capped Eppendorf tubes with the laboratory numbers for each isolate you are submitting. Ensure that these labels will not come off during the heating step.
3. Put 0.5 ml PBS in each Eppendorf tube.
4. Make very **thick** suspensions of organisms by sweeping colonies from each culture plate and emulsifying in the PBS in the tubes.
5. **As soon as possible**, place the Eppendorf tubes in the boiling water bath and leave for 20 minutes.
6. Wrap sealing film around the lids of the tubes.
7. Transport to the EMMC at room temperature using standard mail.