

How to inoculate culture media

A standard method of inoculation of culture media is used to obtain reproducibility of quantitation results. This procedure for streaking plates is to be used for all cultures unless otherwise specified in the individual procedure.

Supplies/Materials

Culture media appropriate to the source of the specimen. (See individual source procedures).

Inoculating loop or needle

Incinerator

Procedure

1. Inoculate the least-selective plates first.

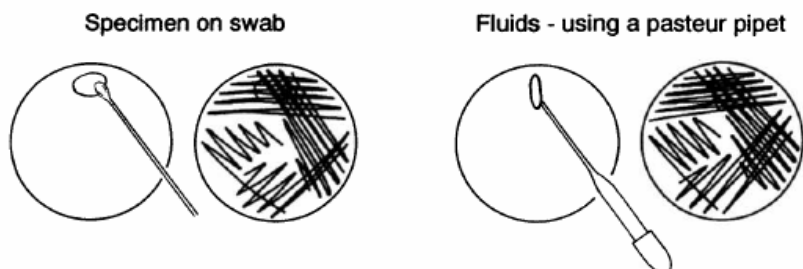
Example: Chocolate, then blood agar, then MacConkey or CNA.

Inoculate any broth tube after the plates. If there are two swabs, leave one swab in the broth. Use the other swab to make the smear. Slides are not sterile and should be prepared after media inoculation.

2. ESwab (contains one swab in liquid transport medium)

- Vigorously shake or vortex the ESwab tube for 5 seconds to release the sample from the swab tip, and evenly disperse and suspend the patient specimen in the liquid transport medium.
- Before inoculating EACH plate/broth/smear, return the swab to the ESwab transport medium tube for 2 seconds to absorb more sample suspension.
 - Unscrew the ESwab cap and use the swab to inoculate the plates and broth. If no slide is inoculated, return the swab to the tube and save the specimen for additional testing if needed.
 - Inoculate the slide last due to the potential for contamination. Remove the contaminated ESwab from the cap with a forceps and discard the swab. Do NOT return the contaminated swab to the transport liquid after slide inoculation. Save the transport liquid specimen for additional testing if needed.
 - Alternatively, a sterile pipet can be used to transfer approximately 100 µL of specimen to each plate/broth and to place 1-2 drops on a slide.

3. Inoculate and streak plates using the four-quadrant method below.



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- a. Quadrant I: Inoculate an area of the plate slightly smaller than a dime. Streak through this area with a sterile loop or needle.
- b. Quadrant II: Streak back into Quadrant I four times, avoiding the initial I inoculum, and streak into Quadrant II.
- c. Quadrant III: Streak back into Quadrant II two to three times and streak into quadrant III.
- d. Quadrant IV: Streak back into Quadrant III **one** time and streak the remainder of the plate for isolation.

Note: Flaming between quadrants is not necessary unless specified in the specific procedure or the specimen appears grossly purulent.