Staining and culture techniques (scale bar = 25 μm)

A **Lugol iodine staining** of the rhabditiform larva in stool. This is the most commonly used procedure in clinical microbiology laboratories. A single stool examination detects larvae in only 30% of cases of infection.

B **Human fecal smear stained with auramine O**, showing orange-yellow fluorescence of the rhabditiform larva under ultraviolet light. Routine acid-fast staining of sputum, other respiratory tract secretions (e.g., bronchial washings), and stool may also serve as a useful screening procedure.

C **Agar plate culture method**. Motile rhabditiform or filariform larvae (the latter increase the longer the plate is kept) and characteristic tracks or furrows, which are made by larvae on the agar around the stool sample. This method is laborious and time-consuming (2–3 days), but is more sensitive than other procedures (e.g., wet mount analysis) in detecting larvae in feces. Tracks are marked (arrows and T). S, stool sample on agar plate; L, larva or larvae.

D **Gram staining** demonstrating *S. stercoralis* filariform larvae (FL). Gram staining of a sputum sample is an excellent tool for diagnosing pulmonary strongyloidiasis.

Procedure for agar plate cultures

1. Place stool on agar plate
2. Seal plate to avoid accidental infection
3. Store plate for 2 days at room temperature
4. Larvae crawl over surface and carry bacteria with them, creating visible tracks
5. Examine plates to confirm larvae
6. Wash with 10% formalin and collect larvae by sedimentation

Repeat this procedure for up to 6 or 7 consecutive days, because of low parasite load and irregular output of larvae in many patients. Tests have shown the agar plate method is superior to a) direct smear, b) the formalin–ether sedimentation technique, and c) the filter paper method. However, the agar plate method is not available globally—sometimes only in large towns and teaching hospitals.