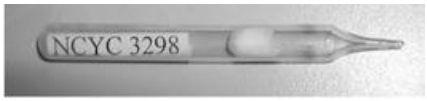
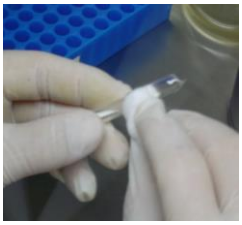
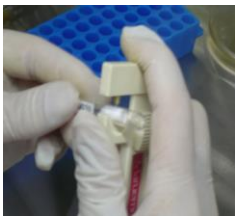
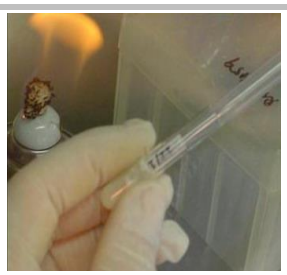
INSTRUCTIONS FOR OPENING AND REVIVING LYOPHILISED CULTURES - Animal Health Laboratories

****Please be noted that aseptic technique must be applied throughout.
****Cultures should be grown on appropriate media and conditions.

What needed are: -
- An ampoule cutter or a file
- Sterile Luria Bertani (LB) broth for *Salmonella* and *Escherichia coli*
- LB agar plates (5 to 6 plates per one strain) for *Salmonella* and *E. coli*
- Columbia broth for *Campylobacter*
- mCCDA agar plates or blood agar (5 to 6 plates per one strain) for *Campylobacter*
- Autopipette with tips or Pasture pipettes
- Inoculating loop

1. Carefully take the ampoule out of the wrap.	
2. Wipe the ampoule neck with 70% alcohol-dampened cotton wool.	
3. Make a deep score on the around the circumference of the ampoule near the middle of the plug using ampoule cutter or a file.	
4. Wrap thick cotton wool around the ampoule and break at the marked area.	

5. Remove the pointed end of the ampoule and cotton into a biohazard container. Pipette 0.5 ml of sterile-appropriate broth into the dried cells, Luria Bertani broth for *Salmonella* and *E. coli* and Columbia broth into for *Campylobacter*. Mix gently and carefully to avoid creating aerosols	
6. For *Salmonella* and *E. coli*, transfer one drop of each strain onto one LB agar plate using or autopipette or pasture pipette. For *Campylobacter*, transfer one drop of each strain onto one mCCDA agar plate or blood agar using or autopipette or pasture pipette. Then, streak the isolate using inoculating loop to get single colonies on plate. The remaining suspension is stored in a screw cap test tube.	
7. For *Salmonella* and *E. coli*, , incubate the inoculated plates and the suspension tubes at 37^0C overnight and observe the bacterial growth. For *Campylobacter*, incubate the plates and the suspension tubes at 42^0C, 48 hours.