MGIT CULTURE PRE/POST TEST-MARKING GUIDE

Name of trainee: ..... **Date:**.....

Answer all questions

1. Describe the principle of MGIT Culture (**7 marks**)

   The MGIT (Mycobacteria Growth Indicator Tube) consists of liquid broth medium that is known to yield better recovery and faster growth of mycobacteria. The MGIT contains 7.0 ml of modified Middlebrook 7H9 broth base. This medium is terminally sterilized by autoclaving. An enrichment, MGIT OADC (Oleic acid, Albumin, Dextrose and Catalase) or MGIT 960 Growth Supplement, is added to make the medium complete. This Growth Supplement is essential for growth of many mycobacteria, especially those belonging to M. tuberculosis complex. Addition of the MGIT PANTA is necessary to suppress contamination. An Oxygen-quenched flouorochrome embedded at the bottom of this 7ml tube, as bacteria grows within this tube free oxygen is utilized and replaced by carbondioxide. With depleted free Oxygen the fluorochrome is no longer inhited thus resulting in flourescence within the tube visualized under UV light by the machine. The intensity of fluorescence is directly prroportional to to the extent of the depletedfree Oxygen.

2. List 3 advantages of Using MGIT CULTURE as a mycobacterial technique (**3marks**)
   Increased recovery of Mycobacteria
   Decreased time to detection compared to solid media
   Long shelf-life of the media; can be store at room temperature

3. State the 4 limitations of using MGIT Culture (**4marks**)
   More prone to contamination
   Not approved for clinical samples such as urine and blood stained sputum
   It's expensive
   More procedures needed to confirm the Mycobacteria strain
   Requires reliable power supply

4. What is the maximum incubation time for a MGIT Sample? (**1mark**)
   42days

5. List atleast five sources of biohazards in the TB culture laboratory. (**5marks**)
   Vortexing/shaking
   Spillage
   Centrifugation
   Improper waste management
   Inoculation/pipetting
   Opening tubes at non-ambient temperature

6. List all the materials and equipment required for preparation of sputum processing reagents for TB culture. (**10marks**)
   Biological Safety Cabinet (BSC)
   1000 µl adjusted pipette
   Sterile pipette tips
   Mycobacteria Growth Indicator Tubes
   Vortex Mixer
   Timer
   MGIT PANTA Antibiotic mixture, MGIT supplement
   Phosphate buffer pH (6.8)
   Sterile NaOH-NALC-Sodium Citrate Solution
   Disposable 50ml falcon tube.

7. Describe the procedure for preparation sputum for inoculation on MGIT tubes.(**10marks**)
   Add an equal volume of NaOH-NALC-Sodium Citrate Solution to the specimen
   Vortex the tube and again hand mix/invert every 5 minutes
   Add PBS up-to the 50 ml mark
   Centrifuge the specimen at 3000g for 15 minutes at 4°C
   At the end of centrifugation allow tubes to sit for 15 minutes to allow aerosols to settle
   Carefully decant off the supernatant leaving behind sediment that is resuspended with 2ml PBS
   The pellet is then use for making smears and inoculation on labelled MGIT or LJ media.

8. Mention at least two Quality indicators for MGIT Culture (**5marks**)
   Contamination Rate.
   Turn Around Time (TAT).

9. What could be the possible causes of false of
   (i) Positive results (**2marks**)
   Mis-labelling at the time of collection/accessioning
   Opening more than one specimen container at a time while processing
   Mix up of test samples/lids, shared reagents/dispensers.
   Failure to take precautions aimed at minimizing aerosol production
   (ii) Negative results (**2marks**)
   Limited bacterial load.
   Harsh decontamination during processing
   Mis-labeling at the time of specimen collection/accessioning

10. Mention five (5) quality assurance activities performed during specimen processing (**5marks**)
    Inclusion of both positive and negative process control samples
    Open specimen tubes gently to avoid aerosol generation
    Add reagents to one open tube at a time
    Leave space between tubes in a rack
    Regularly change gloves
    Disinfect the biological safety cabinet (BSC) work surfaces routinely and have it documented on the cleaning log
    Use daily aliquots of processing reagents
    Pour reagents slowly against the inside wall of the tube to minimize splash

END