SPECIMEN OPTIMAL SAMPLING

Principle: Blood is a sterile fluid. The presence of micro-organisms in the blood indicates bacteremia (transitory phase) or septicemia (infective phase). In suspected cases of septicemia, blood is inoculated into an enrichment broth to maximise quick recovery of microorganisms.

BLOOD CULTURE SPECIMEN PROCESSING USING AUTOMATED MACHINES

PROCEDURE: STEP BY STEP

Within 4 hours of sample being taken, blood culture bottles should be placed onto the automated machine

e.g. BD Bactec FX machine.

After sample flags positive, it should be processed immediately in a class 1 cabinet (especially if clinical details indicate potential Advisory Committee on Dangerous Pathogens (ACDP) category 3 or 4 microorganism)

Disinfect the cap with 70% alcohol then use a sterile needle and syringe to draw 1ml of the broth culture.

Inoculate onto Blood, Chocolate and MacConkey agar plates and incubate as follows:

- Blood and Chocolate agar : 35-37°C in a CO_2 incubator
- MacConkey agar: 35-37°C in an O_2 incubator

If anaerobic incubation conditions are available, inoculate another blood agar plate or an anaerobic agar plate (e.g. Fastidious Anaerobic Agar (FAA) and incubate at 35-37°C in ANO_2 conditions.

Prepare a smear on a glass slide. Perform a Gram strain and report the preliminary results immediately. *

Examine all culture plates from the previous day. If no growth, re-incubate blood, chocolate and anaerobic agar for up to 5 days.

Pick colonies from pure cultures to set up biochemical tests and antibiotic susceptibility testing (AST) using Kirby-Bauer disc diffusion method then incubate accordingly. Refer to EUCAST/CLSI guidelines for correct procedures.

Where available, use automated methods (i.e. Vitek® 2) for identification and AST.
Ensure appropriate quality controls are in place for all identification and AST methods

Set up purity plates on appropriate media and incubate in suitable conditions.

Identify the microorganism and AST as interpreted by the automated machine or by reading and interpreting biochemical results.

Ensure identifications match preliminary gram stain results and colonial morphology

Interpret AST results using the standard guidelines (CLSI or EUCAST).

Cultures that don't flag positive remain in the system till they flag negative. Report results as "Negative" and send out results immediately.

*When no microrganisms are seen on Gram stain, investigate in accordance with local procedures e.g. check the growth curve on the automated system – exponential growth indicates a true positive, repeat Gram stain, use Sandiford counterstain if available, scan multiple fields at x10 magnification for 'clumps' of organisms e.g. *Cutibacterium* and yeasts. If the Gram stain is truly negative, the bottle can be returned to the automated machine, if the machine supports this function (refer to manufacturers IFU for time restrictions).

References: References: Best Practices of Blood Culture in Low- and Middle-Income Countries: https://doi.org/10.3389/fmed.2019.00131
Collection, transport and storage procedures for blood culture specimens in adult patients: https://doi.org/10.1515/cclm-2018-1146
Implementation of Automated Blood culture with Quality

COMMONWEALTH PHARMACISTS ASSOCIATION THET PARTNERSHIPS FOR GLOBAL HEALTH CwPAMS

CwPAMS is funded by the UK Department of Health and Social Care's Fleming Fund using UK aid. The views expressed in this publication are those of the authors and not necessarily those of the UK Department of Health and Social Care.