

T Level Technical Qualification in Healthcare Science

Occupational specialism assessment (OSA)

Assisting with Healthcare Science

Assignment 3 - Standard Operating Procedures

Assignment brief insert

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Assignment 3

Standard Operating Procedures

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Giemsa stain operating procedure

1. Introduction

The Giemsa stain is a complex staining technique that is routinely used to demonstrate the presence of different cells within a patient's blood sample. T stain enables the direct visualisation of white blood cells (leukocytes) and red blood cells (erythrocytes).

2. Safety considerations

Methanol is flammable and can cause irritation to skin eyes and intoxication when ingested or inhaled for a long period of time.

Follow local control of substances hazardous to health (COSHH) and risk assessments when performing all staining procedures and clinic samples/quality controls (QCs) which may potentially be infectious.

3. Method for blood smear

- prepare a blood smear using a drop or 2 of blood sample
- place the drops of blood on a microscope slide
- using a second microscope slide held at 45 degrees, touch the blood and quickly pull away across the slide
- repeat if required until a thinly spread blood smear slide is achieved
- allow the sample to air dry
- fix in methanol for 60 seconds

4. Method for Giemsa staining of blood sample

- tip off the methanol
- allow the sample to air dry
- flood the slide with the produced Giemsa stain and leave for 20 to 25 minutes
- run tap water on to the slide to flush off the stain and to prevent precipitation on the smear
- allow the sample to air dry
- examine slide using light microscopy to detect the presence of the blood sample using up to 1000x magnification
- record number of red blood cells (RBC) and white blood cells (WBC) and calculate respective percentages

Haemocytometer and cell count standard operating procedure

1. Introduction

Haemocytometers are used to calculate the number of cells within a suspension.

2. Safety considerations

Blood from a patient may contain infectious agents and should be treated as infectious during all procedures. Appropriate personal protective equipment (PPE) measures should therefore be taken and the correct cleaning method applied to spillages of samples and the disposal of samples and equipment contaminated by patient's blood.

Follow local control of substances hazardous to health (COSHH) and risk assessments when performing all staining procedures and clinic samples/quality controls (QCs) which may potentially be infectious.

3. Method for separating whole blood using centrifugation

- place a sample of the patient's whole blood into the centrifuge tube (this will be specific to the centrifuge being used)
- at a speed of 2500 to 3000rpm, centrifuge the blood until it separates into red blood cells, buffy coat (middle fraction) and plasma (note: this may take up to 10 to 15 minutes depending on the centrifuge used)
- using a pipette, remove (aliquot) a volume of up to 100µl of the buffy coat (note: this sample can be inverted to mix the components and then used again for centrifugation if repetition is required)

4. Method for counting white blood cells using a haemocytometer

- clean the haemocytometer chamber and cover slip with alcohol
- place the cover slip over the mounting support of the haemocytometer
- using the buffy coat aliquot from the previous method, add an equal volume of crystal violet stain to that sample
- add the 10µl of the buffy coat and crystal violet mixture to the haemocytometer chamber
- adjust the magnification, light intensity and focus until the 4 corner grids can be seen, and individual cells can be identified within them (note: using the 10x objective lens is often most suitable)
- using one of the corner grids, count the number of white blood cells within the grid
- repeat this process for the remaining 3 corner grids and calculate a mean cell number using the results from all 4 grids
- multiply the mean cell count by 10⁴ to calculate the number of cells per ml of crystal violet

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Change History Record

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