FINE NEEDLE ASPIRATE BIOPSY (FNAB) PROCEDURE FOR ENLARGED LYMPH NODES

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Consent
Written parental consent should be obtained

Laboratory assistance
Contact Histopathology (extension 5209) to ask for a technologist to assist with the specimen preparation at the procedure

Anaesthesia
Sedation is required in young children (oral sedation [e.g. chloral hydrate] or parenteral sedation [e.g. ketamine]) and paracetamol
Topical anaesthesia (Emla cream) should be used to optimise pain control

Positioning the child
For FNAB of cervical lymph nodes the patient should be positioned supine with a pillow under the shoulders to extend the neck
In an older child, supraclavicular nodes may be aspirated while in the sitting position

Equipment
Sterile pack, sterile gloves, plastic apron and Biotane in alcohol disinfectant,
Needles
- Use 22 gauge (black), needles unless the child is very small, in which case a narrower gauge needle may be used. Be careful about using narrow gauge needles as caseous material is difficult to aspirate
Syringes
- 10cc
Slides
- Glass slides with ground glass edges are obtainable from Histopathology - Patient details (name, surname and folder number) must be written in pencil on the frosted portion of the slides

Cytology fixative spray, TB transport bottles containing Middlebrook media, and green-capped tubes containing 50% alcohol for cell block preparation are obtainable from Histopathology, 1st floor, ICH building

Infection control measures
Because specimens may contain viable mycobacteria and the specimen preparation may induce aerosolisation a N95 mask should ideally be worn during the procedure

Procedure
- Stabilise the mass with one hand and introduce the needle with the other hand
- Always perform a minimum of 3 needle passes
- Always use a sterile needle and syringe for each pass
- Maintain 1-2 cc suction throughout aspirate
- Aspirate using cutting motion until material appears in hub of needle
- Release suction before withdrawing the needle
- Place cotton wool on insertion site and ask an assistant to apply pressure

Distribution of aspirates
Sufficient specimen material should be obtained to:
- Inoculate 1 x TB transport bottle containing Middlebrook media
- Prepare 1-2 x microscope slides for microbiology i.e. 2 slides air-dried
- Prepare 2 x microscope slides for histopathology analysis i.e. 1 slide fixed with cytology fixative spray and 1 slide air-dried
- Flush 1 syringe in a green-capped tube with 50% alcohol for cell block preparation

Specimen preparation
During office hours (07:00 – 17:00, Monday to Friday) a Histopathology staff member should be contacted (extension: 5209), and if available will come to the procedure to assist with the specimen preparation. Outside these hours the clinician who performs the FNAB is responsible for preparing the specimens as follows:

A. **Inoculation of TB transport bottle**
   - Break the aluminium seal of the transport bottle and clean the surface with alcohol swab
   - If pus-filled material has been aspirated, inject not more than two drops (0.3ml of aspirate) into the transport bottle
   - If minimal material is left in the needle, insert the needle into the transport bottle, aspirate a small amount of the liquid into the syringe and express back into the bottle
   - Before injecting the contents into the transport bottle the needle should be checked for firm attachment to the syringe and manually held in place during inoculation

B. **Slide preparation**
   - Remove needle from syringe, introduce 5-10 cc air into the syringe, then re-attach needle and, holding the needle onto syringe, use air to express material in needle onto glass slide
   - Touch needle tip on glass slide during above, 1cm from frosted end
   - Place second slide parallel to first, and, maintaining gentle pressure, pull the 2 slides apart
   - Spray-fix bottom slide with fixative from distance of about 30cm until wet
   - Repeat above procedure to prepare two additional air-dried slides for microbiology

C. **Specimen for cell block preparation**
   - Flush 1 syringe in the green-capped tube containing 50% alcohol

**How to request and submit specimens to laboratory**
- Complete specimen request form, indicating which tests are required: TB microscopy & culture ± histopathology. Do not request general routine microscopy, culture and sensitivity. Specimens should be transported in a sealed plastic bag along with the corresponding request form.
- Specimens do not require specialized storage, however transport to the laboratory and subsequent analysis should be done as speedily as possible to prevent diagnostic delay.
- The attending Histopathology staff member will take the TB microscopy slides, inoculated TB transport bottle and request form to Specimen Reception for transport to Microbiology at Groote Schuur Hospital. The rest of the microscope slides and specimen in 50% alcohol will be stored in Histopathology until the TB microscopy has been finalised.
- If a Histopathology staff member is not present, the responsible clinician must ensure that the specimens are directed to Specimen Reception, ground floor, ICH building

**Specimen processing**
A Histopathology technologist will obtain the TB microscopy result from the NHLS laboratory results web site: WWDISA. Processing of the histopathology specimens depends on the TB microscopy result (refer below):

- If TB microscopy suggests the presence of acid-fast bacilli the specimen in Middlebrook transport media will be cultured for mycobacteria and processed by Hain line probe assay to identify resistant isolates. If BCG infection is a consideration, speciation of a cultured MTB complex isolate should be discussed with microbiology. **The histopathology slides and cell block will not be processed.**
- If TB microscopy fails to identify acid-fast bacilli, the specimen in Middlebrook transport media will be cultured for mycobacteria. **The histopathology slides & cell block will be screened for cell abnormalities by the Histopathology service.**

**Acknowledgement:** This protocol was adapted from FNAB procedure by Prof C Wright, University of Stellenbosch