

BONE MARROW BIOPSIES

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H.I.S.T.O.

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Tec Tips

I acquired this decalcification procedure after the pathologists complained of poor detail with nitric acid and various commercial products. Dr. Mahoney must like it - he kissed me on the cheek! Thanks to Jan Graham in Ft. Dodge for the procedure.

Aspirate:

1. Allow aspirate to clot in petri dish.
2. Transfer clot to specimen container filled with 10% buffered formalin.
3. Process as usual.

Bone Core Fixation:

1. Immediately fix acetic formalin for a minimum of one hour.
2. Fix overnight if brought in later than 2 hours end of the workday.
3. Decalcify first thing in the morning.

Acetic Formalin		Dilute RDO	
80% ethanol	900cc	Distilled Water	1 part
conc. formalin	100cc	RDO	3 parts
acetic acid	50cc		

Bone Core Decalcification:

1. Place fixed core and label in a plastic cassette (RDO discolors metal). Decalcify in dilute RDO for 1 hour. Do NOT leave overnight.
2. Test for decalcification by gently checking for pliability.
3. Rinse in running tap water for approximately 2 minutes.

Processing:

1. If the biopsy came in afternoon, process with rest of specimens.

2. If the biopsy is decalcified in the morning, hand process in 120cc plastic specimen cups, 15 minutes in each container, starting with 70% ethanol or speed process through processor, 15 minutes each station with heat and vacuum, starting with the first dehydrant station.

Microtoming:

1. Cut both aspirate and bone cores at 2 microns.
2. Place 3 levels on one slide. Cut an extra slide of the last level and set aside on the back of the water bath in case a special stain is requested.

Staining:

1. Hematoxylin approximately 5 minutes. Bone core 1 minute (Check under microscope before counterstaining).
2. Eosin for 10 dips, dehydrate, clear, and coverslip.

Discussion:

Over decalcification will result in poor or indifferent histological detail and staining characteristics. Less than one hour is usually not sufficient.