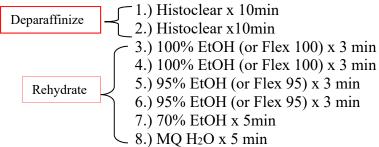
TRAP Staining for Formalin-fixed Paraffin Embedded Sections

Procedure

- 1. Pre-warm MQ H₂O (~50ml per coplin jar) at 37-42°C (for step 3b) ~30min.
- 2. Deparaffinize slides ~60min:

Best if you can heat, but doesn't really matter

Note – never let your slides dry out!!!!!! Throughout this whole procedure



Note all of the above steps do not have to be exact... you can leave for longer

- 3. Prepare the staining solution (according to Beaker B in steps 3 and 4 of the Acid Phosphatase, Leukocyte Kit).
 - a. Make 1 mL Diazotized Fast Garnet GBC Solution (1:1 Fast Garnet GBC Base Solution: Sodium Nitrite Solution). Mix by gentle inversion, let stand for 2 min
 - b. Mix the following solutions while stirring (following volumes = 1 coplin jar):

MQ H₂O pre-warmed to $37^{\circ}\text{C} - 45\text{mL}$

Diazotized Fast Garnet GBC Solution (6a) – 1mL

Naphthol AS-BI Phosphate Solution – **0.5mL**

Acetate Solution – 2mL

Tartrate Solution – 1mL

*scale volumes as necessary for container

- c. Pour solution into coplin jar and ensure that temperature is 37-42°C (Important!)
- 4. Incubate slides at 37-42°C in staining solution for 1 hour.
- 5. Rinse slides thoroughly in MQ H₂O (2x 5-10 min).
- 6. Counterstain with Hematoxylin Solution, Gill No.3 for 2 min.

Put in warm tap water bucket and run warm tap water over it for 8 min

Coverslip with Fluoromount G (Aqueous mounting medium)

Use about 100ul permount per slide

Wipe off excess

Seal with nail polish

Let dry at least 1 hour before looking at on scope (usually O/N)