

DIFFERENTIAL STAINING OF NEURO TISSUE

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PREPARATION- STEP 1

- ▣ Immediately following post mortem the brain is removed and washed.
- ▣ It is then suspended by the Basilar Artery in 10% Formalin
- ▣ This process of suspension will take approximately 2 weeks.

PREPARATION- STEP 2

- ▣ At the end of 2 weeks the brain is removed from the formalin and all the blood vessels on the surface of the brain is removed using a dissecting forcep.
- ▣ This is very important so that there is no interference of these vessels during slicing which will be done later.
- ▣ The brain is then washed again in cold water and again immersed in 10% formalin for a further 4 weeks.

PREPARATION -STEP 4

- ▣ After the 4 weeks the brain is removed and washed overnight in running water.
- ▣ 25% aqueous solution of gelatin is prepared in warm water (30deg celsius) and the brain is submerged briefly into this solution .
- ▣ The gelatin will prevent fragmentation of brain tissue during slicing.
- ▣ The brain is then left back into the formalin for 48 hrs.

SLICING OF BRAIN

- ▣ Having decided on the plane in which the brain is to be sliced (coronal ,sagittal or horizontal) , a cut is made midway using a sharp knife.
- ▣ Bacon slicer is normally used and ideally the slices are 10-12mm thick.
- ▣ Slices are then washed in running water and placed in 10% Formalin for 48hrs and this step is repeated.
- ▣ After washing the slices are ready for staining.

Staining

- ▣ During this stage the slices are handled very carefully with gloves to prevent any breakages.
- ▣ Staining can be done on a flat tray.
- ▣ Solution A , B and C are used for the staining process.

Solution A

- ▣ Solution A comprises of dissolved 50gm phenol crystals and 5 gm copper sulphate in 1 L of distilled water containing 1.25ml HCl.
- ▣ The brain slices are immersed in this Solution A for 5 min and optimal temperature of this solution is 60 deg celsius.
- ▣ After 5min is Solution A the slices are placed in ice water for 10 seconds.

Solution B

- ▣ After removing from ice water the slices are then placed in Solution B for 30-35 seconds.
- ▣ Solution B comprises of Ferric chloride in distilled water.
- ▣ After removal from Solution B the slices are then washed in running water for 1 minute

Solution C

- ▣ After removing from the running water the slices are then placed in Solution C. for 4 minutes.
- ▣ Solution C comprises of 1% Potassium Ferrocyanide in distilled water.
- ▣ After removal from Solution C the slices are placed overnight in running water.

RESULTS

- ▣ The grey matter will appear blue with gelatin appearing darker blue on the outside.
- ▣ The white matter will appear as very light blue
- ▣ The slices can be then be mounted for display.