

Practical-6(B)

Mayer's Hematoxylin & Eosin



by:
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- **It is a general purpose stain or micro-anatomical stain.**

- **Mayer's hematoxylin** is used because it eliminates the necessity for differentiation and bluing of the section. It can be considered a progressive stain which produces a stained section with clearly defined nuclei while the background is completely colorless.

- **Stain Solutions:**

- Mayer's Hematoxylin
- Eosin Solutions

- **Method of preparation:**

- Dissolve 50g aluminum potassium sulfate (alum) in 1000 ml distilled water.
- When alum is completely dissolved, add 1 gm hematoxylin.
- When hematoxylin is completely dissolved, add 0.2 gm sodium iodated and 20 ml acetic acid.
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- Bring solution to boil and cool, and filter.

Mayer's Hematoxylin

PROGRESSIVE Staining Systems

- Hematoxylin stain is applied
- Tissue is stained with hematoxylin only to a point
- Traditionally no differentiator is used to remove excess hematoxylin
- Stain procedure continues on with counterstain

MAYER`S ALUM HAEMATOXYLIN & EOSIN METHOD

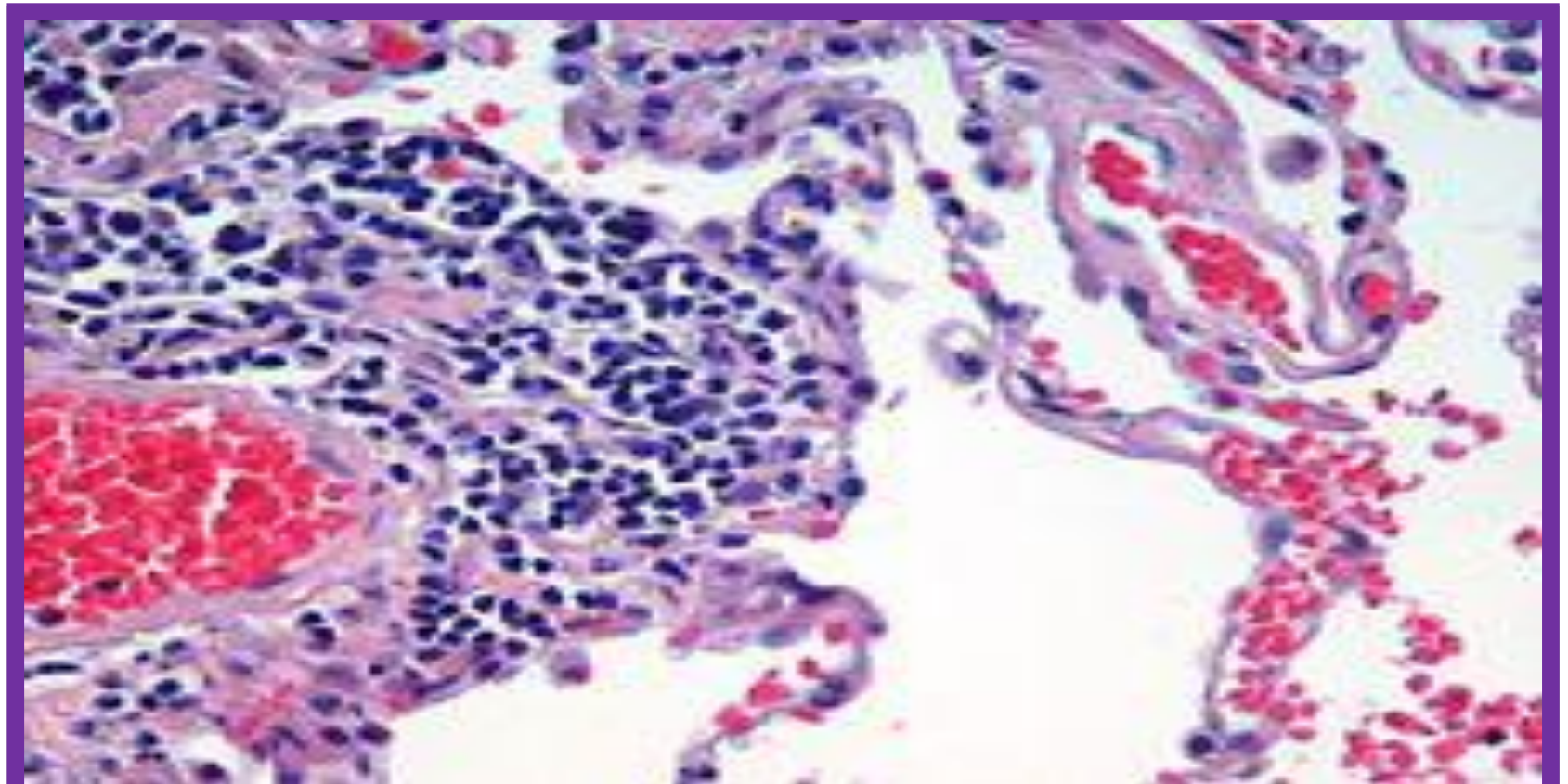
PROCEDURE:		
1. Deep slide	Xylene	3mins.
2. Deep Slide	100% alcohol	2min
3. Deep slide	95% alcohol	1min.
4. Deep slide	70% alcohol	1min.
5. Rinse section to	Tap water	
6. Apply	Mayer`s haematoxylin	10mins.
7. Rinse in	Tap water again	2mins.
8. Blue in	Ammonia water	1min.
9. Apply	Eosin	3mins.
10. Apply	70% alcohol	30seconds
11. Apply	95% alcohol	10seconds
12. Dehydrate	100% alcohol	10seconds
13. Dehydrate	100% alcohol	15seconds
14.Clear in 2 changes	Xylene	2mins each jar
15. Mount in	DPX-cover slipping	

RESULT:

Cell Nuclei _____ blue

RBCs _____ red

Connective tissue & cell cytoplasm _____ shade of pink



MOUNTING:

Mounting usually involves attaching the samples to a glass microscope slide for observation and analysis. In some cases, cells may be grown directly on a slide. For larger pieces of tissue, thin sections (slices) are made using a [microtome](#); these slices can then be mounted and inspected.

Criteria of a good mounting medium:

- It should be miscible with the last fluid from which you are mounting.
- Should have the same refractive index of the glass slide used.
- Should be clear and clean.
- Should flow freely when cover slipping.
- Should harden quickly.
- Should not crack on drying.
- Should not contract too much when setting or drying.
- Should not develop granules on drying.
- Should have the appropriate PH.
- Should be permanent.
- Should not colored with age.
- Should not support life.
- Should not easy to remove its excess from the cover slip.

TWO TYPES OF MOUNTING MEDIUM

AQUEOUS MOUNTING MEDIUM

Generally it is used for temporary mounting media; hours; days; or even weeks.

This includes unstained preparations, fat stains, metachromatic stains, some fluorescent techniques and most enzymes techniques.

THEY CONSIST OF:

Gelatin, gum Arabic or sugar: these will improve the setting quality.

Glycerin: this prevent from cracking and splitting on drying.

Distilled water as a solvent.

A preservative: to prevent the growth of organisms such as phenol.

Potassium acetate: to prevent the stains from dissolving in the mounting medium.

RESINOUS MOUNTING MEDIA

This used for permanent preparations. They are used for routine work except when the substance to stain or the dyes are soluble in the dehydrating, clearing or mounting media.