

LAB-3

PERMANENT SLIDE PREPARATION STAGES

1- Fixation

The goal of this process is to maintain the composition of cells and preventing the growth of bacteria in the sample and prevent decomposition also leads to hardening samples and make it possible to cut into thin slices.

Good fixative penetrates tissue quickly, reduces the shrinkage, and does not change the biological components of the tissue. The fixation of two types:

a- Physical fixation : by heating or cooling to the extent of freezing.

b- Chemical fixation : by using chemical fixative like formaldehyde in light microscope. In the electron microscope different fixative is used such as glutaraldehyde and Potassium permanganate ($KMnO_4$).

2- Dehydration

In this step the sample is exposed to an escalating series of alcohols like ethanol or acetone starting from 30% to 100%. After that clearing is made to the sample by Hydrocarbon like xylol in light microscope while in electron microscope propylene is used. (clearing is the process of making the sample or section clear and free of impurities and remove the dehydration solutions.

3- Embedding

Paraffin wax used to embed the samples to be examined by light microscope, after immersion in the wax, sample is poured in templates and more afford to temperatures to resist the flow of electors dite, epon and spur. These materials are liquid in low to be more leave to cool and solidifies. In the electron beddingn miosopres to resisicroscope embedding material liquid in low temperature.

solid and more afford to tempera like araldite, eponelk cal) Is the process of gettng thin sliced and a certain thicken or manual devices called microtome.

4- Sectioning

a tool used cut extremely thin slices of material, known as sections. Im electron radiation.

science, microtomes are used in microscopy, allowing for the preparation

of Microtomes use steel, glass, or diamond blades depending upon the specimen being sliced and the desired thickness of the sections being, cut Steel blades are used to prepare sections of animal or plant tissues for light microscope histology, Glass knives are used to slice sections for light microscopy and to slice very thin sections for electron microscope. Industrial grade diamond knives are used to slice hard materials such as bone, teeth and plant matter for both light microscopy and for electron microscopy diamond knives are used for slicing thin sections for electron sample for observation under transmitted light or microscope. There are many types of mi applications for this design of microtome are as Sledge large samples, such as those embedded in paraffin are of the for ations, Rotary microtome for hard materials, such as as in a synthetic resin, Cryomicrotome for the cutting sample amples, dions, Vibrating microtome The vibrating microtome frozen y thin seicult biological samples Saw microtome The saw UltramicrotomeIt can allow for the preparati seir teeth as well as some ceramics. for hard materials such as teeth or eciaof microtome can also be used for verotome isibrati Preparatio eripples S vibrat materials, such as bones te cerami for very hard

5- Staining

Sections staining in the light microscope are by different dyes such as fastgreen, iosin, hemotoxin,safranin, while in E.M. by anylacetate or leadcitrate.

6- Mounting

By placing a drop of resinous substance such as Canada balsam or picolyte, then covered with a glass cover and the slide is saved until the study.

Practical part:

Picture of plastic Block.

Picture of Ultra microtome.

Picture of microtome.

Picture of Grids that are made from golden and prepared for receive thin slides from tissues which examined by electron microscope.