المحاضرات العملي

المادة: التحضيرات المجهرية microscopic technique

المحاضرة : الرابعة

المرحلة : الأولى. **الدراسة : الصباحية**

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9_Sectioning:

The sample is fixed on the sample holder of the microtome, and the cutting device must be equipped with a very sharp knife The thickness of the desired sector is determined (3-7) microns for paraffin and a thickness of (10-15) microns for celluloid. Good sectors are usually in the form of tapes or a series of sectors. It is preferable that these tapes be placed on a black plate so that it is easy to distinguish the sectors and take the appropriate them to place it on the glass slide.

10_Mounting :

It means placing the histological sector on a glass slide. The sector is transferred to a microscope slide with a drop of water on it. Then it is placed on a slide dryer and left until the water droplet evaporates and the sector sticks well. On the previously painted slide.

11_Staining:

The paraffin wax must be completely removed from the sectors using xylol, and then the xylol must also be disposed of with absolute alcohol, after which the sectors must be transferred to an environment similar to the one in which the dye is dissolved in water or alcohol.

The duration of staining depends on the type of dye, its concentration, and the nature of the tissue. The tissue may be dyed with more than one dye, or the researcher may resort to what is called counterstaining, such as using hematoxylin to stain the nuclei and eosin to stain the cytoplasm.

After the completion of the staining process, the process of preparing the microscopic slide for permanent preservation begins by using a waxy material or a preservative plastic such as Canada balsam or (D.P.X) material, then the cover of the slide is placed at an acute angle of 45 degrees and very carefully so that no air bubbles form, and this is how what is known as the slide is made. After the permanent slides are left to dry on a desiccator the sections can be examined under a microscope.

Advantages of paraffin wax technology:

1- This technique takes a short time to prepare, not more than two days .

2- It gives a successive series of sectors called tapes that are connected to each other, and these tapes are important for research work.

3- It gives very thin slices.

4- sections This method is easy to dye.

Its disadvantages:

1_ This method uses fixatives and an oven, which may damage the tissue and the details of the cell structure.

2_ The use of stabilizers may dissolve the fat content of the tissue during preparation, so it is not possible to study the fats in this way.

3_ This method is not ideal in tissue chemistry because heat destroys enzymes, for example.

Second:Celloidin Technique :

The previous method is similar to the paraffin wax method, except that in this method paraffin is replaced by celluloid.

Its advantages:

1_ It gives full clarity clips where tissue details can be seen.

2_ Not using heat in it preserves the tissue structures as they are.

3_Large pieces of tissue can be used, such as the whole eye.

Its disadvantages:

1_ It takes a long time (about a month) to prepare the sector.

2_ It is not possible to obtain tapes from the sectors because they are thick and separated from each other.

3_ The celluloid strip is very difficult to cut and dye.

Third: the freezing technique

In this method, the soft or fixed tissue is frozen and hardened, and sections are made from it with a microscope and an ice tom, or what is called (cryostat).

Its advantages:

1_ A quick and simple method that is usually used during surgeries that require a quick diagnosis of cancer.

2_ The chemicals in the fabric do not change because no heat is used.

3_ It is used in tissue chemistry to study the activity of cellular enzymes, lipid detection and interaction Antibodies with antigens.

Its disadvantages:

1_ Do not give a series of syllables.

2_ It gives thick sectors due to the difficulty of cutting and dyeing.

Decalcification process

It is known that the presence of calcium salts in the tissues causes hardening of the tissue so that it is not possible to cut these tissues by microtome. Therefore, these salts must be removed so that we can obtain a soft tissue sample that can be cut without causing indentations or damage to the cutting knife.

*** Definition of decalcification: is the method by which we remove deposits of calcium salts from rich tissues such as bones, teeth...**

The process is done by placing the sample in a calcium-reducing solution such as Brienne's solution, buffered formic acid solution, Gooding's solution and Senyurt's solution, Schmidt's solution, and Lily's solution.