



الجامعة التقنية الشمالية
المعهد التقني / الموصل
قسم تقنيات المختبرات الطبية

التشريح

اعداد

م.م. غفران محمد سعدي


المعهد التقني - الموصل
TECHNICAL INSTITUTE - MOSUL

Ministry of high Education and Scientific Research
Foundation of Technical Education
Institute of medical Technology/ AL- Mosul

Training package
In

Micro technique
For

Students of first class
Pathology analysis



Micro technique



First Modular Unit

ξ/ post test:-

- ١- Define the following:-
Micro technique , Biopsy, Autopsy, Autolysis, Bacterial decomposition.
- ٢- Enumerate the methods in common use

Note:-

Check your answers in page ٦, ٧, ٨.

ο/Sources:-

- ١- Micro technique
By: Dr .Majida A .R.(١٩٨٣)
- ٢- BAKER .JR.(١٩٤٥)
Cytological technique ٣nd Ed. London.

Ministry of high Education and Scientific Research
Foundation of Technical Education
Institute of medical Technology/ AL- Mosul

Training package
In
Steps for preparation of tissue section
For
Students of first class
Pathology analysis



Steps for preparation of tissue section



Second Modular Unit

३/ The text:- steps for preparation of tissue section.

- १ - **Anesthetization:** The purpose of this step is to make the animal tissues more relax the solution or the method used:-
 - a- Chromosome – b- Ether – c- cocaine – d- cooling – e – Alcohol.
- २ - **Killing :-** Rapid step of all biological activities of the organism.
- ३ - **Fixation :** treatment the specimen with fixation material to keep Shape and arrangement of the cells as normal.
- ४ - **Washing:** the specimen wash to remove the excess of uncombined fixative because some of fixative influence the precede steps specially staining.
- ५ - **Dehydration :** is removing of complete water from tissue by replaeing it by ascending concentration of dehadrate agents (Alcohol).
- ६ - **Clearing:** is replace the dehydrating agents from the tissue by clearing agents.
- ७ - **Infiltration:** Impregnation the cleared tissue with molten paraffin wax or other embedding media to fill the porous in the tissue to give asupport.
- ८ - **Blocking out:Embedding :-** Enclosed the tissue by certain media to make it to micro section.
- ९ - **Cutting :-** It means the cutting of block to serial micro sections using the micro tomes.
- १० - **mounting :** putting an individed tissue or small ribbon of tissue On the slide and fixed by adhesive- material
- ११ - **Staining:-** Coulored the tissue (section) by certain dyes to identified certain tissue components.
- १२ - **Mounting cover slip:-** Certain media used to fixed a cover slip on slides to maintain the tissue section.

४/ post test:-

- १ - Define only five the steps of preparation section

٢- Write the steps of preparation section,

Note:-

Check your answers in page ٦, ٧, ٨.

o/Sources:-

- ١- Micro technique
By: Dr .Majida A .R.(١٩٨٣)
- ٢- BAKER .JR.(١٩٤٥)
Cytological technique ٢nd Ed. London.

Ministry of high Education and Scientific Research
Foundation of Technical Education
Institute of medical Technology/ AL- Mosul

Training package
In

Fixtation
For

Students of first class
Pathology analysis



Fixation



Thrid Modular Unit

✓/The text:- **FIXATION**

Aim of fixation:-

१. Inhibit or stop (Autolysis) and (bacterial decomposition) of the tissue due to the coagulation of protein.
२. Hardening the tissue.
३. Make the tissue translucent and improve optical differentiation of the tissue.
४. Facilitated the subsequent steps (specially the staining).
०. Fortify the tissue against the harmful effect of the processing subsequent steps length of the exposure time depends on:-
 १. Rapidly of penetration of the fixative.
 २. Tissue thickness.
 ३. Nature of the tissue.

Principle of fixation:-

१. The quantity of the fixative solution must be at least २० times the size of the specimen.
२. The fixative solution must rounded the specimen from all sides.
३. The size of the specimen must be not so large or so small.

Factures of good fixative:-

१. Quickly penetrate the tissue.
२. Not make a shrinkage of the tissue.
३. Keep the cellular components and don't melt the fatty tissue.

ξ/ post test:-

- १- What are the aim of fixation?
- २- Length of the exposure time depends on:-
 - a-.....

b-.....

c-.....

٢- Write on the principle of fixation .

٣- Name the factors of good fixative.

Note:-

Check your answers in page ٦, ٧, ٨.

o/Sources:-

١- Micro technique

By: Dr .Majida A .R.(١٩٨٣)

٢- BAKER .JR.(١٩٤٥)

Cytological technique ٢nd Ed. London.

Ministry of high Education and Scientific Research
Foundation of Technical Education
Institute of medical Technology/ AL- Mosul

Training package
In
Types of Fixtation
For

Students of first class
Pathology analysis



Type of Fixation



Fourth Modular Unit

٣/The text:- Types of fixatives:-

A. According to the coagulation of protein:-

١. Coagulant fixatives:-

These fixatives convert homogenous spongy shape of protein to insure the penetration of the wax into the tissue (like mercuric chloride, ethyl alcohol).

٢. Non coagulant fixatives:-

These fixatives coagulated the protein into normal shape (like – formaldehyde – acetic acid)

B. According to the chemical component:-

١. Simple fixative:- Contain one chemical substance.

٢. Compound fixative:- Contain more than one chemical substance.

١) ١٠% formal saline:- Which contains

- | | |
|-----------------------|---------|
| A. ٤٠ % formaldehyde. | ١٠٠ ml. |
| B. Sodium chloride. | ٨٥ gm. |
| C. Tap water | ٩٠٠ ml. |

Advantages:-

المحاضرة الخامسة

١. Well penetration.
٢. Blood and fatty tissue are preserver.
٣. Causes little shrinkage.
٤. Permit a large variety of staining method.

Disadvantages:-

Store the specimen for many months – formic acid produced which destroys the staining properties.

٢) ١٠ % formalin:- (solution – fluid – fixative)

Preparation:-

- | | |
|-----------------------|---------|
| A. ٤٠ % formaldehyde. | ١٠٠ ml. |
| B. Tap water | ٩٠٠ ml. |

٤٠% = saturated

نقي مشبع

Advantages:-

١. Produce very little shrinkage.
٢. Fixation the sample with it can be followed by most staining technique.

Disadvantages:-

١. Formalin has irritant vapour which may effect the nasal mucus.
٢. Dermatitis may be produced by prolonged exposure to formalin.

٣) Zinger fixative (orange sol.)

Preparation:-

- | | |
|-------------------------|---------|
| ١. Mercuric chloride | ٥ gm. |
| ٢. Potassium dichromate | ٢,٥ gm. |
| ٣. Sodium sulfate | ١ gm. |
| ٤. Distilled water | ١٠٠ ml |

Add ٥ ml

Glacial acetic acid before use.

Disadvantages:-

١. Poor in penetration.
 ٢. Prolonged exposure of tissue. (٢٤h) become brittle.
 ٣. Mercury precipitate. Like course black pigment.
 ٤. Tissue fixed in zinker cutting badly when using frozen sections.
- ٤) Boun's fixative (yellow solution)

Preparation:-

- | | |
|--------------------------|--------|
| ١. Picric acid | ٧٥ ml. |
| ٢. ٤٠% formaldehyde | ٢٥ ml. |
| ٣. Classical acetic acid | ٥ ml. |

Advantages:-

١. Penetrate rapidly.
٢. Permit very good staining of nuclei and connective tissue fiber.
٣. Tissue doesn't need washing with water.

Disadvantages:-

١. Tissue become brittle.
٢. Kidney tissue should never be present in it.

•) Carnoy's fixative

Preparation:-

- | | |
|---------------------|--------|
| ١. Absolute alcohol | ٦٠ ml. |
| ٢. Chloroform | ٣٠ ml. |
| ٣. G. acetic acid | ١٠ ml. |

Advantages:-

١. Quickly penetrate and acting.
٢. Tissue transferred directly to absolute alcohol.
٣. Good fixative for chromosomes.

Post test:-

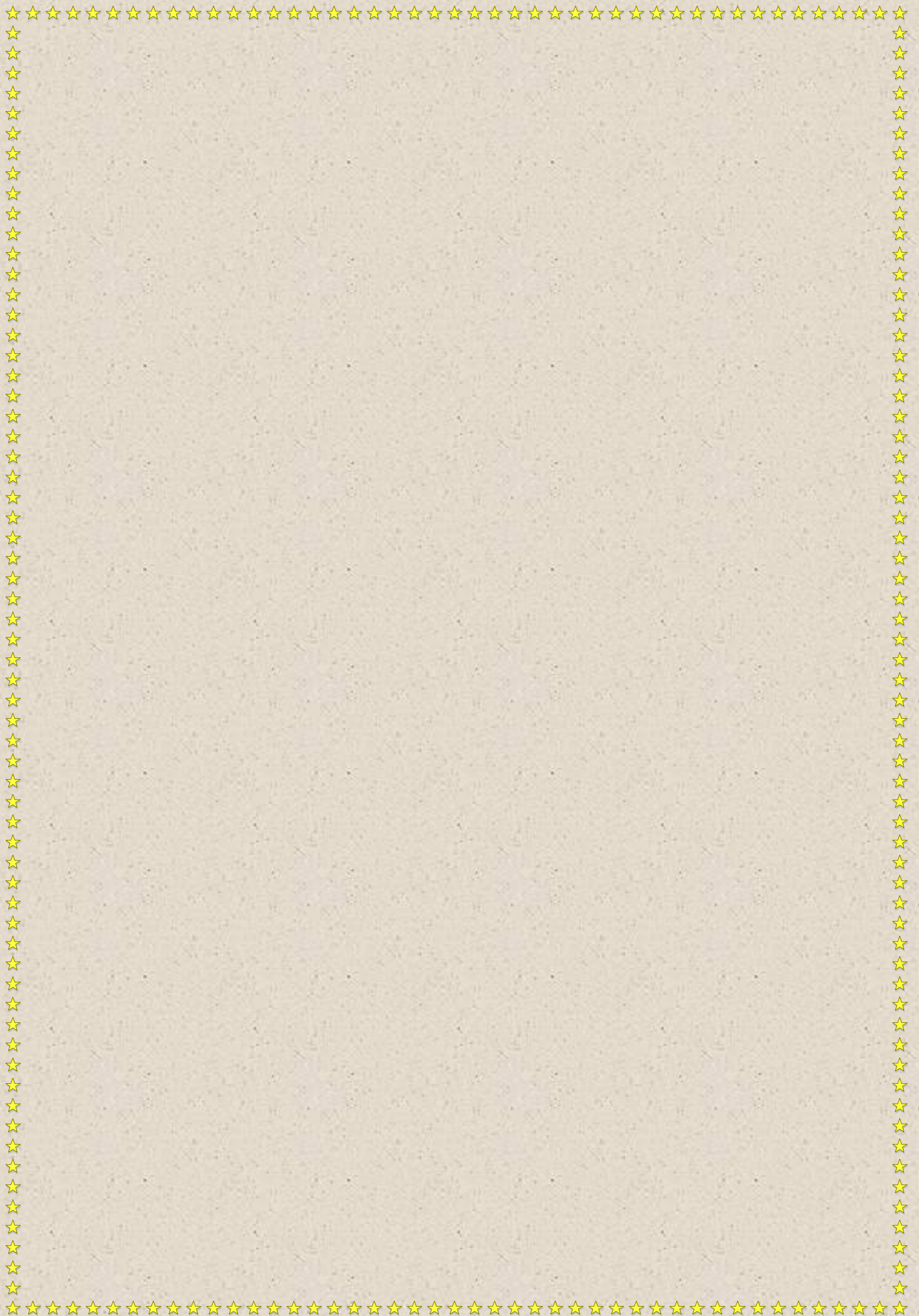
- ١- Classify the types of fixatives.
- ٢- Preparation of ١٠% formal saline and ١٠% formaline solution.
- ٣- Write on advantages and disadvantages of zink fixative.
- ٤- Define : simple fixation and compound fixation.

Note:-

Check your answers in page ٦, ٧, ٨.

•/Sources:-

- ١- Micro technique
By: Dr .Majida A .R.(١٩٨٣)
- ٢- BAKER .JR.(١٩٤٥)
Cytological technique ٢nd Ed. London.



Ministry of high Education and Scientific Research
Foundation of Technical Education
Institute of medical Technology/ AL- Mosul

Training package
In

Washing
For

Students of first class
Pathology analysis



Washing



Fifth Modular Unit

٣/The text:- Washing:

It means the removal of the excess of uncombined fixatives.

Fixation in fluids containing potassium dichromate or mercuric chloride like zenker fixative, the tissue must be wash with running tap water for ١٢ – ٢٤ hr.

Fixation in fluids containing picric acid like bouin's fixative, the tissue must be wash ٧٠% alcohol. Because the washing with tap water make the swelling collagen fibers.

Tissue transfers to ٧٠% alcohol for ٣ – ٨ hr.

Dehydration:-

Dehydration is the removing of the water from the tissue by replacing it by dehydrating agents, like alcohol, propanol.

The dehydration was manifested by the immersion of the tissue in ascending strength of alcohol. (usually from ٥%).

The length of time spends in each strength of alcohol and total time of dehydration depends on the tissue thickness and tissue density and fixative used.

Solutions used as dehydrant agents:-

١. Alcohol;
٢. Acetone: (dehydrate quickly).
٣. Diaxone: it can used shead of a high strength at alcohol. Caused little shrinkage it is a poison material.
٤. Tetrandrofurane:- like dioxane.

٤/Post test:-

- ١- Define washing .
- ٢- Solution used in washing.
- ٣- Time of washing .
- ٤- Define dehydration.
- ٥- Solutions used as dehydrant agents.

Note:-

Check your answers in page ٦, ٧, ٨.

o/Sources:-

- ١- Micro technique
By: Dr .Majida A .R.(١٩٨٣)
- ٢- BAKER .JR.(١٩٤٥)
Cytological technique ٢nd Ed. London.

Ministry of high Education and Scientific Research
Foundation of Technical Education
Institute of medical Technology/ AL- Mosul

Training package
In
Clearing
For

Students of first class
Pathology analysis



Clearing

Sixth Modular Unit

٣/The text:- Clearing:

It is the removal of dehydrating agent from the specimen and filled with fluid can mixed with paraffin wax and makes the specimen translucent allow the microscope light to penetrate to insure a good diagnosis or study the specimen.

Clearing agents:

١. Xylol (xylene):- colourless with special order.
٢. Chloroform:- slow in clearing than xylol for this must be put the specimen long time in it.
٣. Benzene:- more speed in clearing, but effect on the bone marrow when treat with it for long period.
٤. Toluene:- very expensive.
٥. Dioxane:- used in hydration and clearing.
٦. Cleve oil.
٧. Cedar wood oil:- these two clearing agent are difficult to remove from the specimen after used for thin must be used three steps in paraffin fluid when enfiltrate.
٨. isopropanol:- used in dehydration and clearing.
٩. Tertiary butyl.

٤/Post test:-

- ١- Define clearing
- ٢- Enumerate the solution used as clearing agents

Note:-

Check your answers in page ٦, ٧, ٨.

٥/Sources:-

- ١- Micro technique
By: Dr .Majida A .R.(١٩٨٣)
- ٢- BAKER .JR.(١٩٤٥)

Cytological technique 2nd Ed. London.

Ministry of high Education and Scientific Research
Foundation of Technical Education
Institute of medical Technology/ AL- Mosul

Training package
In

Infiltration
For

Students of first class
Pathology analysis



Infiltration



Seventh Modular Unit

٣/The text

Infiltration or impregnation:-

It is a method where the molten paraffin wax replaces the clearing agents by three changes of molten paraffin. In other hand, the infiltration means the coating of the specimen tissue by the wax and diffusion of the melting wax in side the spaces of the tissue that took place as a result of the melting of the adipose tissue and the collapse of the other tissue components during the previous process.

The aim of this step is for supporting the tissue against harmful of the embedding media and facilitate the cutting.

Infiltration or embedding media:-

Paraffin wax – Collidin – Gelatin – Plastic.

Paraffin wax:-

There are three types of paraffin wax according to melting point:

١. Soft p. W. The melting point between $48 - 50$ c°.
٢. Medium p.W. The melting point between $50 - 56$ c°.
٣. Hard p. W. The melting point between $56 - 60$ c°.

Type of p. w. use infiltrate – embedding:-

Notice:-

Compact tissue the wax used of melting point $56 - 60$ c° (hard).

soft tissue the wax used of melting point $50 - 56$ c° (medium).

For thin section used hard type of wax (5μ and less).

For thick section used soft type of wax (7μ and more).

٤/Post test:-

- ١- Define infiltration
- ٢- Media used in infiltration:-
a----- b----- c-----
- ٣- Classify the paraffin wax according the melting point.
- ٤- Compact tissue the wax used of melting point(-----)
- ٥- For thin section used hard type of wax(-----)

Note:-

Check your answers in page ٦, ٧, ٨.

٥/Sources:-

- ١- Micro technique
By: Dr .Majida A .R.(١٩٨٣)
- ٢- BAKER .JR.(١٩٤٥)
Cytological technique ٢nd Ed. London.

Ministry of high Education and Scientific Research
Foundation of Technical Education
Institute of medical Technology/ AL- Mosul

Training package
In

Blocking
For

Students of first class
Pathology analysis

By



Blocking



eight Modular Unit

٣/The text

Blocking – embedding – casting out:

It is the surrounding of the infiltration specimen by paraffin wax and orientating the specimen in a compact suitable size and allowed the wax to solidify as compact cubic mass ready to cut into very thin section.

Type of embedding:-

١. Simple embedding and infiltration with same media.
٢. Compound embedding infiltration with certain media. Like celliodin and the embedding by other media. Like wax
٣. Embedding with paraffin wax famous because:-

Advantages:-

١. The process is fast and simple.
٢. The embedding tissue can be store for a long time.
٣. Very thin section (٢ micron) can be get.
٤. A ribbon of sections can be getting.
٥. The sections are easy to be adhering on glass slide.
٦. Paraffin wax with different melting point can be get.

Disadvantages of p. w. method:-

١. Over heated p. W. enters the specimen brittle and making the sectioning was difficult.
٢. Prolonged treatment in paraffin causes shrinkage and hardening of tissue.
٣. Paraffin processing removes fat because the dehydrants and clearing agents are fat solvent.

Trimming:-

It is the removing of the excess wax from the block by shaving the edge of the block from an surface.

Infiltration or impregnation:-

It is a method where the molten paraffin wax replaces the clearing agents by three changes of molten paraffin. In other hand, the infiltration means the coating of the specimen tissue by the wax and diffusion of the melting wax in side the spaces of the tissue that took place as a result of the melting of the adipose tissue and the calapse of the other tissue components during the previous process.

The aim of this step is for supporting the tissue against harmful of the embedding media and facilitate the cutting.

Infiltration or embedding media:-

Paraffin wax – Collidin – Gelatin – Plastic.

Paraffin wax:-

There are three types of paraffin wax according to melting point:

١. Soft p. W. The melting point between $48 - 50$ c°.
٢. Medium p.W. The melting point between $50 - 56$ c°.
٣. Hard p. W. The melting point between $56 - 60$ c°.

Type of p. w. use infiltrate – embedding:-

Notice:-

Compact tissue the wax used of melting point $56 - 60$ c° (hard).

soft tissue the wax used of melting point $50 - 56$ c° (medium).

For thin section used hard type of wax (5μ and less).

For thick section used soft type of wax (7μ and more).

€/Post test:-

- ١- Define blocking
- ٢- What are the types of embedding.
- ٣- Write on advantages and dis advantages of paraffin wax method.
- ٤- Define trimming.

Note:-

Check your answers in page ٦, ٧, ٨.

o/Sources:-


- ١- Micro technique
By: Dr .Majida A .R.(١٩٨٣)
- ٢- BAKER .JR.(١٩٤٥)
Cytological technique ٣nd Ed. London.

Ministry of high Education and Scientific Research
Foundation of Technical Education
Institute of medical Technology/ AL- Mosul

Training package
In

Cutting
For

Students of first class
Pathology analysis



Cutting



Ninth Modular Unit

٣/The text Cutting the block:-

It is the cut of the block into micro section with thickness between (١ – ٥٠) μ using a tool called microtome.

Types of microtome:-

١. Rotary microtome:- for p. W. Blocks cutting.
٢. Freezing microtome:- for fresh tissue like block by using CO_2 gas.
٣. Sliding microtome:- for cellodine block cutting.
٤. Ultra microtome:- for electron microscope study.

Types of knives:-

١. Wedge shaped knife for wax.
٢. Planoconvex knife for cellodine.

Difficulties in cutting appear because:-

١. Imperfect of dehydration.
٢. Imperfect of clearing.
٣. Imperfect of impregnation with p. W.

Mounting sections on slide:-

It is a fixing of tissue sections on a glass slide using adhesive mixture (material) like Mayer's egg albumin then drying the sections by:-

١. For ٢٤ hr in an incubator at ٣٧ c° .
٢. For ٣٦ – ٤٣ hr at room temperature.

٣. For ٢٠ minutes in an incubator at ٥٧ – ٦٠ c° .

Mounting the sections on slides:-

By using one of three ways:-

١. By hot plate;- using Mayer's egg albumin as adhesive material.
٢. By warm water bath:- using Mayer's egg albumin.
٣. By alcohol burner:- using ٣٠٪ alcohol.

Adhesive mixture Mayer's egg albumine:-

١. egg albumin. ٥٠٪.
٢. glycerin ٥٠٪.
٣. few crystals of thymol – prevent growth of moulds.

٤/Post test:-

- ١- Define cutting
- ٢- What are the microtomes and their knives.
- ٣- Define mounting and name the ways using in this step.
- ٤- Enumerate the ways used in drying the section.
- ٥- prepare the adhesive mixture mayers egg albumine.

Note:-

Check your answers in page ٦, ٧, ٨.

٥/Sources:-

- ١- Micro technique
By: Dr .Majida A .R.(١٩٨٣)
- ٢- BAKER .JR.(١٩٤٥)
Cytological technique ٢nd Ed. London.

Ministry of high Education and Scientific Research
Foundation of Technical Education
Institute of medical Technology/ AL- Mosul

Training package
In

Staining
For

Students of first class
Pathology analysis



Staining



Tenth Modular Unit

٣/The text Staining:-

All types of tissue sections after mounting on slide can not be seen by light microscope because the cell and other tissue components are pale and uniform for differentiation of the tissue components must be stain.

There are three methods for staining the tissue:-

١. –Vital – staining method:- it is an inject the animal by stains then kill the animal and remove the tissue for making sections. This type of staining using for applied research.
٢. Routine staining method:- this method uses in laboratories and apply the haematoxylin and easin stain.
٣. special staining method:- this method uses to study the changing in the nature or amount of the cell and tissue components and using a special stain.

How does the tissue coloured:-

The basic dyes stain acidic components of the tissue and vice versa.

The common acidic components of the tissue are nuclei, mucus, and cartilage which attract with a basic dyes and the basic components, as the cytoplasm which attract with acidic dyes.

Types of stains (dyes):-

١. Natural dyes.
 - A. Plant source --- like Hx.
 - B. Animal source --- like camine.
٢. Artificial dyes.
 - A. Basic dyes:- stain the acidic organelles like nucleus.
 - B. Acidic dyes:- stain the basic organells, like R.B.C., cytoplasm.
 - C. Neutral dyes:- like sudan III, methyl blue.

Mordents:-

It is a substance that has a strong affinity for dye and also for a tissue, e.g. iron alum, potassium alum.

Where does the dye and the mordant unit to form a coloured lake:-

Direct stain:-

It is a staining of the tissue with the dye directly without using of the mordant.

Indirect stain:-

It is a staining of the tissue with the dye by using a mordant.

Stock solution:-

A solution that prepared in a strength more concentrated than what is actually needed.

Regressive stain:-

A dye tends to stain many tissue components but does not become equally firmly attached to all of them then the reaction between the dye and a specific chemical component of the tissue allowed to go too far and the excess of the dye is removed slowly until the desired result has been obtained, e.g HX.

Progressive stain:-

A dye at beginning does not stain the tissue components with perfect intensity then the reaction of staining is allowed to progress until the intensity of staining is satisfactory.

Differentiation:-

It is the remove of the unwanted excess of the dye.

Differentiator:-

The agent that does removing of the excess dye, e.g. acid alcohol, 2% aqueous ferric chloride (2% FeCl₃)

Bleaching:-

It is a removed of the natural pigment or a dye caused by osmium tetroxide, or potassium dichromate using 1% hydrogen peroxide in alcohol or by 1, 2% potassium permanganate.

ξ/Post test:-

- ١- Define stining
- ٢- Discuss the methods used in stining.
- ٣- What are the types of stains (dyes)
- ξ- How does the tissue coloured

Note:-

Check your answers in page ٦, ٧, ٨.

ο/Sources:-

- ١- Micro technique
By: Dr .Majida A .R.(١٩٨٣)
- ٢- BAKER .JR.(١٩٤٥)
Cytological technique ٢nd Ed. London.

Ministry of high Education and Scientific Research
Foundation of Technical Education
Institute of medical Technology/ AL- Mosul

Training package
In
Special Staining
For
Students of first class
Pathology analysis



Special Staining

Special Staining



eleventh Modular Unit

✓/The text Staining:-

All types of tissue sections after mounting on slide can not be seen by light microscope because the cell and other tissue components are pale and uniform for differentiation of the tissue components must be stain.

There are three methods for staining the tissue:-

1. –Vital – staining method:- it is an inject the animal by stains then kill the animal and remove the tissue for making sections. This type of staining using for applied research.
2. Routine staining method:- this method uses in laboratories and apply the haematoxylin and easin stain.
3. special staining method:- this method uses to study the changing in the nature or amount of the cell and tissue components and using a special stain.

How does the tissue coloured:-

The basic dyes stain acidic components of the tissue and vice versa.

The common acidic components of the tissue are nuclei, mucus, and cartilage which attract with a basic dyes and the basic components, as the cytoplasm which attract with acidic dyes.

Types of stains (dyes):-

1. Natural dyes.
 - A. Plant source --- like Hx.
 - B. Animal source --- like camine.
2. Artificial dyes.
 - A. Basic dyes:- stain the acidic organelles like nucleus.
 - B. Acidic dyes:- stain the basic organells, like R.B.C., cytoplasm.
 - C. Neutral dyes:- like sudan III, methyl blue.

Mordents:-

It is a substance that has a strong affinity for dye and also for a tissue, e.g. iron alum, potassium alum.

Where does the dye and the mordant unit to form a coloured lake:-

Direct stain:-

It is a staining of the tissue with the dye directly without using of the mordant.

Indirect stain:-

It is a staining of the tissue with the dye by using a mordant.

Stock solution:-

A solution that prepared in a strength more concentrated than what is actually needed.

Regressive stain:-

A dye tends to stain many tissue components but does not become equally firmly attached to all of them then the reaction between the dye and a specific chemical component of the tissue allowed to go too far and the excess of the dye is removed slowly until the desired result has been obtained, e.g HX.

Progressive stain:-

A dye at beginning does not stain the tissue components with perfect intensity then the reaction of staining is allowed to progress until the intensity of staining is satisfactory.

Differentiation:-

It is the remove of the unwanted excess of the dye.

Differentiator:-

The agent that does removing of the excess dye, e.g. acid alcohol, 2.5% aqueous ferric chloride (2.5% FeCl₃)

Bleaching:-

It is a removed of the natural pigment or a dye caused by osmium tetroxide, or potassium dichromate using 0% hydrogen peroxide in alcohol or by 1, 2% potassium permanganate.

ξ/Post test:-

- ١- Define stining
- ٢- Discuss the methods used in stining.
- ٣- What are the types of stains (dyes)
- ٤- How does the tissue coloured

Note:-

Check your answers in page ٦, ٧, ٨.

ο/Sources:-


- ١- Micro technique
By: Dr .Majida A .R.(١٩٨٣)
- ٢- BAKER .JR.(١٩٤٥)
Cytological technique ٣nd Ed. London.

Ministry of high Education and Scientific Research
Foundation of Technical Education
Institute of medical Technology/ AL- Mosul

Training package
In

Treatment of bone tissue
For

Students of first class
Pathology analysis



Treatment of bone tissue



Twelevth Modular Unit

3/The text **Treatment of bone**

Decalcification: complete removal of calcium or phosphate salts from bony tissue to softening the tissue using material called decalcifying fluid like nitric acid, formic acid.

Bones contains large quantity of calcium salts; these make it hard so that sections can't be prepared by ordinary techniques. The same is tissue of other calcified structures including teeth and variety of tissue in which calcium salts have been deposit as result of disease.

Decalcification:- is carried by using decalcifying fluids where some of it cause some loss of histological detail and staining quantities.

The decalcifying fluid:-

1. formic acid.
2. hydrochloric acid.
3. 5% nitric acid.
4. formic citrate (formic acid – sodium citrate).
5. nitric acid – formalin.
6. trichloroacetic acid (5%).
7. pereny's solution.
8. haugs solution.

Fixation:-

The best fixture for bone is 10% formalin, but Zenker fixative should not be used because it penetrates poorly.

Notes:-

1. Decalcification by electrolytic method.
 2. Decalcification by ion exchange resins.
 3. Decalcification by chelating agents.
- Thin sizes of bone are necessary for (5 mm thickness).
1. To allow penetration of the fixation.
 2. To reduce the time for decalcification.


o/Sources:-

- ١- Micro technique
By: Dr .Majida A .R.(١٩٨٣)
- ٢- BAKER .JR.(١٩٤٥)
Cytological technique ٢nd Ed. London.

Ministry of high Education and Scientific Research
Foundation of Technical Education
Institute of medical Technology/ AL- Mosul

Training package
In
Differences between electron and light microscope
For

Students of first class
Pathology analysis



Differences between electron and light microscop



Thirteenth Modular Unit

۳/ The text Differences between electron and light microscope:

	Electron microscope	Light microscope
۱.	High resolution power (۰۰۰,۰۰۰X)	Resolution power not more than (۱۰۰۰X)
۲.	Fixative used Osmic acid, glutvaldehyde.	Fixative used, zenker Boin's, formalin.
۳.	Embedding medium is plastic (Methylacrylate and apoxy resin)	Embedding medium are paraffin, wax, celliodin and ice block.
۴.	Depend on acurren of electrons for pentration the tissue section.	Depend on a beam of light for penetration of the tissue section.
۵.	Sectioning using ultra microtome.	Sectioning using rotary microtome, sliding microtome freeing.

Differences between Paraffin and Celliodin method

	Paraffin method	Celliodin method
۱.	Preparation time is short	Preparation time is long.
۲.	Can get a thin section	Can not get a thin section
۳.	Can get a ribbon of section.	Can not get a ribbon of section.

۴/ Post test:-

- ۱- what are the differences between electron and light microscope.
- ۲- What are differences between paraffin and celliodin method.

Note:-

Check your answers in page ۶, ۷, ۸.

۵/ Sources:-

- ۱- Micro technique
By: Dr .Majida A .R.(۱۹۸۳)

٢- BAKER .JR.(١٩٤٥)
Cytological technique ٧nd Ed. London.

Ministry of high Education and Scientific Research
Foundation of Technical Education
Institute of medical Technology/ AL- Mosul

Training package
In
Frozen section
For

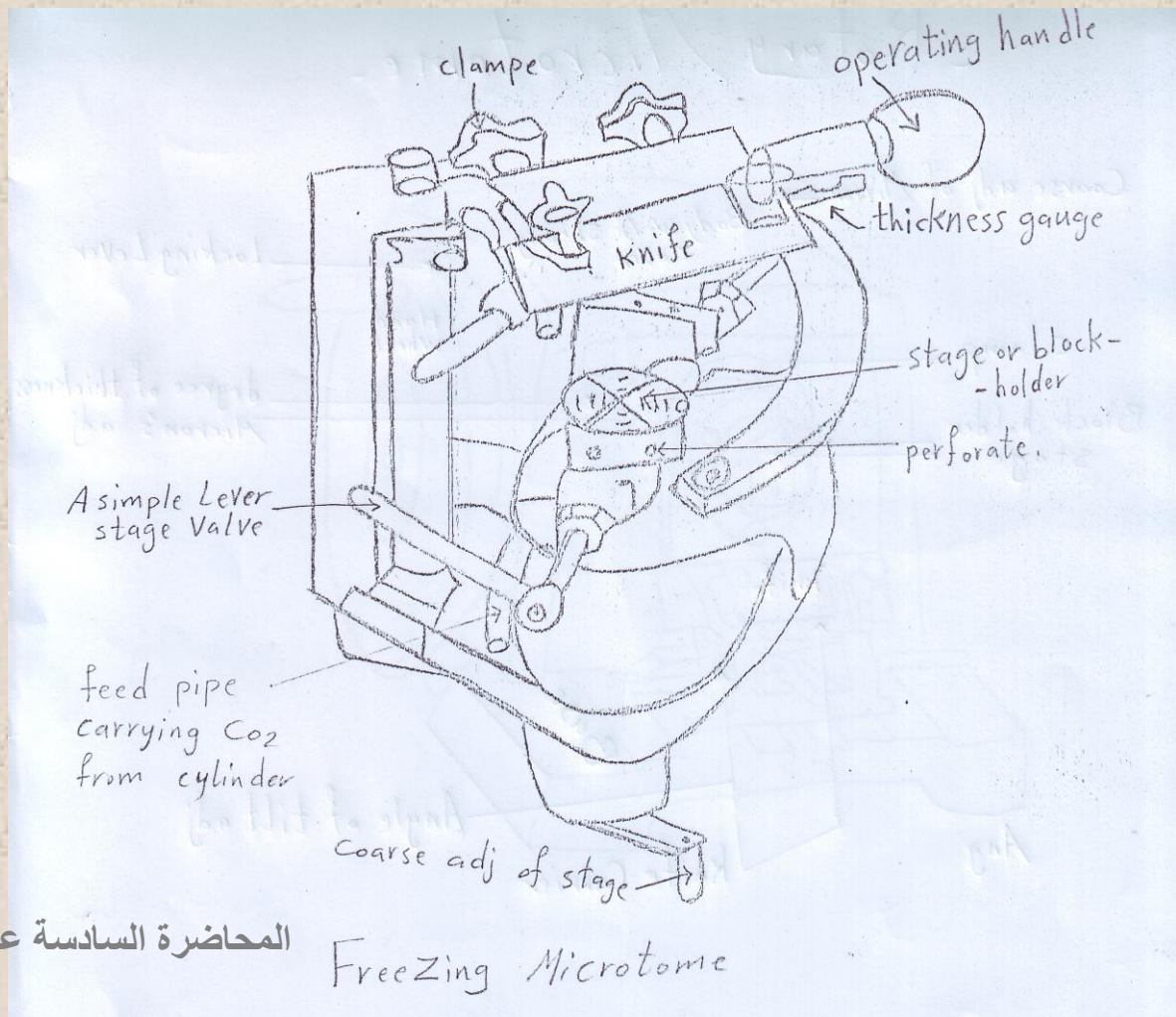
Students of first class
Pathology analysis



Frozen section



Fourteenth Modular Unit



المحاضرة السادسة عشر

٢/ The text:- Frozen section:-

Due to the routine paraffin processing alters the chemical reaction with in the tissue cause a complete loss of enzymes and other soluble substances (ex. Lipids) for these reasons it is necessary to use frozen method to obtain micro sections.

- Frozen sections may cut from fresh unfixed tissue and from fixed tissue.
- The fixative used with frozen technique is ١٠% formal saline and formaldehyde.
- The thickness of frozen sections ranging between ٧ – ١٥ micron.
- Type of block used with frozen microtome is fresh tissue turn to ice block using a blasts CO₂ gas.

The fixed tissue is easier to cut on a freezing microtome than unfixed tissue.

Don't use (avoid) fixative containing alcohol because of the inhibitory effect of the alcohol in freezing.

Fixative causing excessive hardening of the tissue are also must be avoided (e.g. – zenker fluid).

Mounting media used with frozen sections is Glycerin Jelly.

Preparation:-

Gelatin 10 gm

D: W 100 ml

Glycerol 100 ml

Difference between paraffin and freezing method

	Paraffin method	Freezing method
1.	Supporting medium is p.w.	Supporting medium is ice..
2.	Continuous ribbon can be obtained.	Not continuous ribbon but single section can be obtained.
3.	Fixed tissue used	Fixed and unfixed tissue can be used
4.	Section obtained from (4 – 8) thickness.	Section obtained from (10 – 20) in thickness.
5.	Using rotary μ	Using freezing μ

The stain was sudan III

There is no need for dehydration because it dissolves the fat.

4/Post test:-

1- why used frozen section.

2- Complete the following:-

a- The fixative used with frozen technique is-----

and ----- .

b- The thickness of frozen section ranging between-----
micron.

c- Mountng media used with frozen section is -----

ƣ- Preparation of glycerine jelly .

ξ- What are the difference between the paraffin and freezing method?

ο-which stain used in frozen section?

Note:-

Check your answers in page ٦, ٧, ٨.

ο/Sources:-

١- Micro technique

By: Dr .Majida A .R.(١٩٨٣)


٢- BAKER .JR.(١٩٤٥)

Cytological technique ٣nd Ed. London.

Ministry of high Education and Scientific Research
Foundation of Technical Education
Institute of medical Technology/ AL- Mosul

Training package
In
Exfoliative cytology
For

Students of first class
Pathology analysis



Exfoliative cytology



Fifteenth Modular Unit

3/The text:- Exfoliative cytology:-

Is the study of the superficial cells which have been exfoliated from mucus membrane, renal tubules and so on, and also includes the study of those cells, which have been scraped or pulled of such membrane. Such cells may be also found in body fluids for example sputum peritoneal fluid most of techniques employed are for the rapid and early diagnoses of malignancy.

It is important that smears are sufficiently thin and that they are put into fixative while still moist slides.

High concentration of RNA in cells has been used as an indication of maligna.

The acridine – orange technique was popular and may be subsequently stained by the (papanicolaou's).

The sections are smeared directly onto viscid in nature leig cervical, vaginal, nipple and prostatic.

Then placed in the fixative immediately fixation freshly prepared smears should be immersed immediately in the ethal – alcohol solution method used to obtain cytological material are:-

1. Nature techniques.
2. Mechanical method – by scraping the cells by a special tool (uterus, vagina).
3. Aspiration:- method by pull the internal fluid (pretoneal fluid) chest fluid.

Types of carcinoma:-

There are three types of carcinoma, these are:-

1. Squamous type.
2. Adeno car (glandular type).
3. Oats type

character of malignant cells:-

1. The cell size and shape differ per day.

- ٢. The nucleus of malignant cell larger than that of normal cells.
- ٣. The nuclei of malignant cells differ in size.
- ٤. The nuclei of malignant cells was larger according to the cytoplasm.
- ٥. Hyperchromatosis:- the chromatin of malignant cells darker than that of the normal cell.
- ٦. Cells division:- (Hyperchromatosis nucleus larger nucleus take different shap nucleus showing many nuclei cell division).

٤/Post test:-

- ١- why used frozen section.
- ٢- Complete the following:-
 - a- The fixative used with frozen technique is----- and ----- .
 - b- The thickness of frozen section ranging between----- micron.
 - c- Mountng media used with frozen section is -----
- ٣- Preparation of glycerine jelly .
- ٤- What are the difference between the paraffin and freezing method?
 - ٥-which stain used in frozen section?

Note:-

Check your answers in page ٦, ٧, ٨.

٥/Sources:-

- ١- Micro technique
By: Dr .Majida A .R.(١٩٨٣)
- ٢- BAKER .JR.(١٩٤٥)
Cytological technique ٢nd Ed. London.