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QUESTION:

Write on the purpose of fixation and list five compound fixatives and composition

A fixative is a stabilizing agent and preservation of biological tissues from decay due to autolysis or putrefaction. This process allows for the investigation of tissue structure that is determined by shape and sizes of the macromolecules

1. It acts to disable biomolecules particularly the proteolytic enzymes which either damages or digests the sample.
2. Protects the sample from extrinsic damage as some fixatives are toxic to some microorganism mostly the bacteria that might exist in tissue form. Many fixatives chemically alters the fixed material to make it less palatable.
3. Fixatives alters the cell tissues on a molecular level to increase mechanical strength and stability.
4. Histologically a fixative is used to preserve or harden fresh cell tissue specimens for microscopic examination.
5. Improve optical density.
6. Doesn’t cause shrinking.

COMPOUND FIXATIVES

Compound Fixatives – These are fixatives in which more than one chemical is used .

It is divided into three groups

i) Micro Anatomical .ii) Cytological iii) Histochemical .

MICRO ANATOMICAL

1) Zenker’s Fluid

CYTOLOGICAL - i) Nucleartic

ii) Cytoplasmic

Nucleartic - - The nuclus part of the tissue or cell will fix and leave the cytoplasmic part .

Reagents : -

a) Cornoy’s Fluid

Absolute alcohol - 60 ml

Chloroform - 30 ml

Glecial acetic acid - 10 ml

b) Flening’s Fluid

1% Aquas chromic acid - 15 ml

2% Aquas osmium tetroxide - 4 ml

Glecial acetic acid - 1 ml

c) New Comer’s Fluid

Isopropile alcohol - 60 ml

Petrolium ether - 10 ml

Propionic acid - 30 ml

Ace4tone - 10 ml

Cytoplasmic - - The cytoplasm part of tissue / cell will fix and leave the nucleus .

Reagents : -

a) Champy’s Fluid

3% potassium dichromate – 6 ml

1% chromic acid - 1ml

2% osmium tetroxide - 3 ml

b) Muller’s Fluid

Potassium dichromate - 2.5 gm

Sodium sulfate - 1 gm

Distilled water - 100 ml

Advantages : It preserves nuclear and connective tissue very well . Fixation in Zenker’s fluid facilitates metachromatic staining . Fixation time is 10-20 hours .

Disadvantages : Only thin pieces of tissue can be fixed ( can penetrate up-to 5 mm) . It is expensive and has to be preserved only in special bottles made of nickel alloy . It deteriorates fast . If the tissue is left in the fluid for 3-4 days , it becomes hard and brittle .

2) Formol Neutral Buffer

a) Sodium di-hydrozen phosphate - 3.5 gm

b) Di sodium hydrozen phosphate - 6.5 gm

c) Formalin - 100 ml

d) Distilled water - 900 ml

It is the most commonly used fixative . Tissue is fixed in formalin usually in 12-24 hours . 10% Neutral formalin ( neutralized by calcium carbonate ) in water is also used . Formalin is cheap , preserves most tissue well and its only disadvantage that it is allergenic .

3) Bouin’s Fluid

a) Saturated (Aquas) picric acid - 75 ml

b) Formallin - 25 ml

c) Glecial acetic acid - 5 ml

Advantage : Rapid penetration , preserves glycogen , improves greatly staining of nuclei and connective tissue .

The tissue takes-up yellow colour of picric acid , hence small piece of tissue can easily be spotted .

Disadvantage : It over hardness the tissue if left in the fixative for more than 24-hours . It lyses red cells . Tissue fixed in Bouin’s fluid should be directly transferred to 70% alcohol . It must not be washed in water because it forms water-0soluble picrates .

4) Gender’s Fluid

a) Saturated( Alcohol) picric acid - 80 ml

b) Formalin - 15 ml

c) Glacial acetic acid - 5 ml

5. Heidenhein’s susa

a) Mercuric chloride - 4.5 gm

b) Sodium chloride - 0.5 gm

c) Trichloro acetic acid - 2.0 gm

d) Glecial acetic acid - 4 ml

e) Formalin - 20 ml

f) Distilled water(Up-To) - 100ml