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ASSIGNMENT!!!:

- 1. Write on the purpose of fixation
- 2. List 5 compound fixatives and compositions.

ANSWERS:

1. The purpose of fixation is to preserve tissues permanently in as life-like a state as possible. Fixation should be carried out as soon as possible after removal of the tissues (in the case of surgical pathology) or soon after death (with autopsy) to prevent autolysis. There is no perfect fixative, though formaldehyde comes the closest. Therefore, a variety of fixatives are available for use, depending on the type of tissue present and features to be demonstrated.

2. List 5 compound fixatives and compositions

A. PHOSPHATE BUFFERED FORMALIN

Formulation

40% formaldehyde: 100 mlDistilled water: 900 ml

- Sodium dihydrogen phosphate monohydrate: 4 g Disodium hydrogen phosphate anhydrous 6.5 g
- The solution should have a pH of 6.8
- Fixation time: 12 24 hours

The most widely used formaldehyde-based fixative for routine histopathology. The buffer tends to prevent the formation of formalin pigment. Many epitopes require antigen retrieval for successful IHC following its use. Most pathologists feel comfortable interpreting the morphology produced with this type of fixative.

B). FORMAL CALCIUM

Formulation

40% formaldehyde: 100 ml
Calcium chloride: 10 g
Distilled water: 900 ml
Fixationtime:12–24hours

Recommended for the preservation of lipids especially phospholipids.

C). FORMAL SALINE

Formulation

40% formaldehyde: 100 ml
Sodium chloride: 9 g
Distilled water: 900 ml
Fixation time: 12 – 24 hours

This mixture of formaldehyde in isotonic saline was widely used for routine histopathology prior to the introduction of phosphate buffered formalin. It often produces formalin pigment.

D). ZINC FORMALIN (UNBUFFERED)

Formulation:

• Zinc sulphate: 1 g

• Deionised water: 900 ml

• Stir until dissolved then add – • 40% formaldehyde: 100 ml

• Fixation time: 4 – 8 hours

Zinc formalin solutions were devised as alternatives to mercuric chloride formulations. They are said to give improved results

with IHC. There are a number of alternative formulas available some of which contain zinc chloride which is thought to be slightly more corrosive than zinc sulphate.

E). ZENKER'S FIXATIVE

Formulation:

Distilled water: 950 mlMercuric chloride: 50 g

• Potassium dichromate: 25 g • Glacial acetic acid: 50 ml

• Fixation time: 4 – 24 hours

Gives good nuclear preservation but lyses red blood cells due to the presence of acetic acid. Has been recommended for congested specimens and gives good results with PTAH and trichrome staining. Produces mercury pigment which should be removed from sections prior to staining and can produce chrome pigment if tissue is not washed in water prior to processing. Is an intolerant agent so, after water washing, tissue should be stored in 70% ethanol.