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ASSIGNMENT!!

1. Write on the purpose of fixation

2. List 5 compound fixatives and compositions

ANSWERS

1. The motivation behind Fixation is to save tissues forever in as life-like a state as could be expected under the circumstances. Fixation ought to be completed as quickly as time permits after expulsion of the tissues (on account of careful pathology) or not long after death (with post-mortem examination) to forestall autolysis. There is no ideal fixative, however formaldehyde comes the nearest. In this way, an assortment of fixatives are accessible for use, contingent upon the kind of tissue present and highlights to be illustrated.

2. List 5 compound fixatives and compositions A. PHOSPHATE BUFFERED FORMALIN

Formulation

• 40% formaldehyde: 100 ml

• Distilled 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 ml

• Mercuric 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.