



APPENDIX K

สำนักหอสมุด

Reagents for tissue fixation and staining

1. Buffered neutral formalin solution (10% neutral formalin)

This buffer contained the following ingredients:

40% formalin	100	ml
KH ₂ PO ₄	4	g and
K ₂ HPO ₄	6.5	g

The buffer was prepared by dissolving all the above ingredients in 900 ml of DW.

2. Mayer's hematoxylin stain

The stain consisted of the following ingredients:

Hematoxylin crystals	1	g
Sodium iodate	0.2	g
Ammonium or potassium alum	50	g
Citric acid	1	g and
Chloral hydrate	50	g

The stain was prepared by first dissolving the alum in DW and hematoxylin crystals were then added while gently mixing. Other reagents were subsequently added and the final volume was adjusted to 1 L with DW. The solution was stirred gently overnight at 25°C and filtered through a Whatman no. 1 filter paper and kept at 25°C. The final color of the stain should be raddish violet.

3. 1% stock alcoholic eosin stain

The stock solution was prepared by dissolving 1 g of Eosin Y (Merck) in 20 ml of DW. After completely dissolved, 80 ml of 95% ethanol was added to the preparation. The solution was filtered through a Whatman no. 1 filter paper and kept at 25°C. To prepare working solution, one part of stock solution was added in three parts of 80% ethanol and 0.5 ml of glacial acetic acid was added to each 100 ml of the stain.

4. 0.01 M PBS, pH 7.2

To prepare in 1 L, the solution was prepared by dissolving 1.22 g of Na_2HPO_4 , 0.17 g of NaH_2PO_4 and 8.77 g of NaCl in 1 L of DW. The pH of the solution was adjusted to 7.2 with 1 N HCl.

5. Sodium citrate buffer, pH 6.0

Solution A (0.1 M citrate acid): $\text{C}_6\text{H}_8\text{O}_7 \cdot \text{H}_2\text{O}$ (21.01 g) was dissolved in 1 L of DW.

Solution B (0.1 M sodium citrate): $\text{Na}_3\text{C}_6\text{H}_5\text{O}_7 \cdot 2\text{H}_2\text{O}$ (29.41 g) was dissolved in 1 L of DW.

The working sodium citrate buffer, pH 6.0 was prepared by mixing 9 ml of solution A with 41 ml of solution B. The pH of this solution was adjusted to 6.0 with 1 N HCl and the volume was made up to 500 ml with DW.