Sudan IV Stain, Herxheimer Alcoholic - Technical Memo

SOLUTION: 125 ml
Sudan IV Stain, Herxheimer Alcoholic  Part 1400A

Additionally Needed:
Alcohol, Ethyl Denatured, 70%  Part 10844
Hematoxylin Stain, Mayer Modified  Part 1202
Lithium Carbonate, Saturated Aqueous  Part 12215 or Scott Tap Water Substitute  Part 1380
Mount-Quick Aqueous Mounting Medium  Part 6271A

For storage requirements and expiration date refer to individual bottle labels.

APPLICATION:
Newcomer Supply Sudan IV Stain, Herxheimer Alcoholic procedure is used for identification of fat/lipid in frozen sections. Herxheimer method refers to an acetone/alcohol solvent mixture; the acetone component of this solution may dissolve out small amounts of lipid.

Sudan dyes are a group of fat/lipid soluble solvent dyes, also known as lysochromes. These solvent dyes readily stain fat/lipid due to the fact that the dyes are more soluble in lipid than in the solvents from which they are applied.

METHOD:
Fixation: Fresh tissue or formalin fixed unprocessed tissue

See Procedure Note #1.

Technique: Frozen sections cut at 8 microns on adhesive slides

Solutions: All solutions are manufactured by Newcomer Supply, Inc.

All Newcomer Supply stain procedures are designed to be used with Coplin jars filled to 40 ml following the provided staining procedure.

STAINING PROCEDURE:

1. Fix frozen section slides in Formalin 10%, Phosphate Buffered for 1 minute.
   a. See Procedure Note #2.

2. Rinse sections carefully in 2 changes of distilled water.

3. Rinse in Alcohol, Ethyl Denatured, 70% (10844).

4. Stain in Sudan IV Stain, Herxheimer Alcoholic for 10 minutes.
   a. Keep tightly capped to avoid evaporation.

5. Differentiate quickly in Alcohol, Ethyl Denatured, 70% to remove excess stain.

6. Wash thoroughly in distilled water.

7. Counterstain with Hematoxylin Stain, Mayer Modified (1202) for 2-3 minutes.

8. Wash gently in several changes of tap water.

9. Blue in Lithium Carbonate, Saturated Aqueous (12215) or Scott Tap Water Substitute (1380) for 10 dips.
   a. The use of a bluing agent is optional.

10. Wash gently in several changes of tap water.

11. Blot excess water from slide; coverslip with Mount-Quick Aqueous Mounting Medium.
   a. See Procedure Note #3.

RESULTS:
Fat Orange/red
Nuclei Blue

REFERENCES:
4. Modifications developed by Newcomer Supply Laboratory.