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**FAT - SUDAN BLACK B-PROPYLENE GLYCOL**

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**PURPOSE:** For the demonstration of fat.

**PRINCIPLE:** Sudan Black is slightly basic dye and will combine with acidic groups in compound lipids, thus staining phospholipids also. An alternative stain to the Sudan Black B stain.

**CONTROL:** Use a positive control of a fat smeared slide, and a negative control slide of a paraffin processed tissue, such as lung.

**FIXATIVE:** 10% formalin.

**TECHNIQUE:** Cut frozen tissue sections 10 $\mu$ .

**EQUIPMENT:** Cryostat, coplin jars. (Making stain, stir plate, filter paper, fritted glass filter, and vacuum) and a 60°C oven. Rinse all glassware in DI water.

**REAGENTS:**

**Propylene Glycol:**

Place in two coplin jars, label #1 and #2, can be reused.

**Sudan Black B/Propylene:**

Sudan Black B	0.7 gm
Propylene glycol	100.0 ml

**CAUTION:** Avoid contact and inhalation.

**85% Propylene Glycol:**

Propylene glycol	85.0 ml
Distilled water	15.0 ml

**CAUTION:** Avoid contact and inhalation.

**Hematoxylin:**

Commercial Gill-3

**Glycerin Jelly**

Dissolve sudan black in propylene glycol, slowly, while stirring. Heat to 100°C, but not over 110°C, for a few minutes, stirring constantly. Filter through Whatman #2 filter paper. Cool, and filter again through a frittered glass filter of medium porosity with suction. Store in a 60°C oven. Solution stable for 1 year.

**CAUTION:** Avoid contact and inhalation.

**SAFETY:** Wear gloves, goggles and lab coat. Avoid contact and inhalation. Propylene glycol; mild skin and eye irritant. Cumbustible.

**PROCEDURE:**

1. Pick-up frozen sections on clean glass slides if fresh, albuminized slides if fixed.
2. Fix slides in 10% formalin if fresh.
3. Wash well it tap, rinse in distilled, drain off excess water.
4. Propylene glycol, two changes, 5 minutes each.
5. Sudan Black, 7 minutes, agitate.
6. 85% Propylene glycol, 3 minutes.
7. Rinse in distilled water.
8. Nuclear Fast Red, 3 minutes.
9. Wash in water.
10. Wash in tap water, rinse in distilled.
11. Mount with aqueous mounting media, Glycerin Jelly.

**RESULTS:**

Fat            blue-black  
Nuclei        red

**REFERENCE:**

Carson, F, Histotechnology: A Self-Instructional Text, 1st Ed,1990, pp161-62, ASCP Press  
Crookham,J, Dapson,R, Hazardous Chemicals in the Histopathology Laboratory, 2nd ED, 1991, Anatech

Prepared: \_\_\_\_\_ By: \_\_\_\_\_

Approved: \_\_\_\_\_ By: \_\_\_\_\_

## PROCEDURE CARD

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DATE: \_\_\_\_\_

TECH: \_\_\_\_\_

EXPIRATION: \_\_\_\_\_

**PROPYLENE GLYCOL #1**

DATE: \_\_\_\_\_

TECH: \_\_\_\_\_

**PROPYLENE GLYCOL #2**

DATE: \_\_\_\_\_

TECH: \_\_\_\_\_