Hematoxylin and Eosin Stain  
(H&E)

TECHNIQUE: Formalin fixed, paraffin tissue sections

REAGENTS:

Tacha’s CAT Hematoxylin (Biocare Medical, catalog# CATHE-GL)

1X PBS (pH 7.2-7.3)

Alcoholic-Eosin

1.0% Eosin Y

Eosin Y (CAS# 17372-87-1) --------------------------------------------- 1.0 g  
Distilled water ---------------------------------------------------------- 100 ml

1.0% Phloxine B

Phloxine B (CAS# 18472-87-2) --------------------------------------------- 0.5 g  
Distilled water ---------------------------------------------------------- 50.0 ml

Working Alcoholic-Eosin Solution

1.0% Eosin Y --------------------------------------------- 50.0 ml  
1.0% Phloxine B --------------------------------------------- 5.0 ml  
95% Ethanol --------------------------------------------- 390.0 ml  
Acetic acid, glacial --------------------------------------------- 4.0 ml
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PROCEDURE:

1. Deparaffinize and rehydrate slides to distilled water
2. Stain in Tacha’s CAT Hematoxylin for 1 minute
3. Wash with 4-5 changes of Tap water or until blue stops coming off slides
4. Blue nuclei in 1X PBS for 1 minute
5. Wash with 3 changes of distilled water
6. Counterstain in Alcoholic-Eosin for 1 minute (DO NOT RINSE)
7. Dehydrate through 3 changes of 95% EtOH and 2 changes of 100% EtOH (1 minute each change)
8. Clear in 3 changes of Xylene (1 minute each change) and coverslip

*Do NOT rinse with water after Alcoholic-Eosin! Go directly into dehydration of slides.

RESULTS:

Nuclei ------------------------------- blue
Cytoplasm ---------------------------- pink
Erythrocytes ------------------------- bright red
All other eosinophilic structures ------ red, pink, or orange