ELASTIC TISSUE FIBERS - WEIGERT'S RESORCIN-FUCHSIN

PURPOSE: A "progressive" staining technique to demonstrate elastic fibers.

PRINCIPLE: The complex formed from the basic fuchsin, an iron resorcin lake, binds to the elastic fibers, resulting in the blue-black staining. The nuclear detail is stained with an iron hematoxylin which will not over differentiate in the acidic elastic stain solution.

CONTROL: Artery or skin

FIXATIVE: 10% formalin or any well fixed tissue.

TECHNIQUE: Cut paraffin sections at 4m or 5m.

EQUIPMENT: Rinse glassware in distilled water: coplin jars.

REAGENTS:

Resorcin-fuchsin Stain

Basic fuchsin	2.0 gm
Resorcinol	4.0 gm
Distilled water	200.0 ml

In a 500 ml beaker, bring solution to a strong boil, add:

29% Ferric chloride	25.0 ml

Stir and continue to boil 2-5 min. Cool and filter, discard filtrate. Dry precipitate and filter paper. Returnto beaker, add:

95% alcohol	200.0 ml

Heat carefully. Stir constantly, dissolve precipitate, discard filter paper. Cool, filter, restore volume to 200 ml with 95% alcohol, add:

Hydrochloric acid	4.0 ml

Label and date, stable for 1 year.

Weigert's Hematoxylin:
See Masson's Trichrome

Van Gieson's Solution:
See EVG

SAFETY: Wear gloves, goggles and lab coat. Work in well ventilated area, work under fume hood. Avoid contact and inhalation.

Hydrochloric acid: strong irritant to skin, eyes and respiratory system. Target organ effects via inhalation on skin, respiratory, reproductive and fetal systems. Corrosive.

Basic fuchsin (pararosaniline) is a known carcinogen. Wear gloves, goggles, particle mask and lab coat, while preparing solution. Work under the hood, keep hot, uncapped, solutions under the hood.

Picric acid: toxic, can become explosive if allowed to become dry.

Iodine: dermal sensitizer, irritant to eyes, skin and respiratory system.

PROCEDURE:

1. Deparaffinize and hydrate to water.
2. Weigert's working hematoxylin (fresh), 10 minutes.
3. Wash in tap water.
4. Scott's Bluing, 30 seconds.
5. Wash in running tap water, 5 minutes, rinse in distilled water.
6. Resorcin-fuchsin solution:
   a. microwave, 30 seconds
   b. allow to stand at room temperature, 1.5 hours.
   c. repeat for a total staining time of 3 hours. Save solution.
7. Rinse off excess stain in 95% alcohol.
8. Wash in tap water, rinse in distilled.
9. Van Gieson solution, 1 minute.
10. Dehydrate quickly through alcohols and xylene, coverslip.

RESULTS:

Elastic fibers	blue-black
Collagen	pink-red
Nuclei	pale blue-black
Other	yellow

NOTES:

1. When microwaving recorcin-fuchsin solution, place coplin jar in 500 ml beaker containing approximately 200 ml distilled water. This will help to keep the soluton from boiling over due to the high alcohol content.
2. Pour the recorcin-fuchsin stain back into the solution bottle.

REFERENCES:

Sheehan D, Hrapchak B, Theory and practice of Histotechnology, 2nd Ed, 1980, pp 194-195, Battelle Press, Ohio

Crookham, J, Dapson, R, Hazardous Chemicals in the Histopathology Laboratory, 2nd ED,1991, Anatech

Safety and Compliance In the Histology Laboratory: Biohazards to Toxic Chemicals, Fredenburgh J, Grizzle W., NSH Symposium, Oct. 1993, Philadelphia

Prepared:_____By: _____

Approved:_____By:_____

PROCEDURE CARD

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Other	yellow

NOTES:

1. When microwaving recorcin-fuchsin solution, place coplin jar in 500 ml beaker containing approximately 200 ml distilled water, to keep the solution from boiling over.
2. Pour the recorcin-fuchsin stain back into the solution bottle when staining is complete.

Weigert's Hematoxylin
See Masson's Trichrome

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Label and date, stable for 1 year.

CAUTION: Corrosive, flammable, carcinogen.

TECH:_____

DATE:_____

EXPIRATION:_____