

INTENDED USE

Brazilin is used as a red nuclear stain.

PRODUCT SUMMARY

Brazilin reacts like Hematoxylin when used to prepare alum-based stain formulations. Brazilin is oxidized to brazilein with sodium iodate, then complexed with aluminum. The mechanism of staining is the same as with Hematoxylin. In all likelihood, the aluminum atom forms coordinate bonds to nucleic acids. Additionally, four hydroxyls on the alum-brazilein complex offer sites for hydrogen bonding. At suitably low pH, staining is limited to DNA and RNA.

Brazilin may be used to replace Hematoxylin in alum hematoxylin stains (e.g., Harris, Gill, and Delafield formulations) in applications where red nuclear staining is desired. It makes an excellent substitute for nuclear fast red.

INGREDIENTS

Purified Brazilwood

WARNING

Brazilin is a natural dye of low toxicity. Irritant. Avoid contact with skin, eyes, and respiratory system.

For In Vitro Diagnostic Use.

STORAGE

Store at room temperature, away from direct sunlight. Keep containers tightly closed when not in use.

DIRECTIONS FOR USE

1. Solution preparations

When replacing Brazilin for hematoxylin, double the amount of dye specified in the hematoxylin formula. The following is a Harris based formulation. It is available commercially as Brazilliant! (ANATECH Catalog #861).

- a. Alum Solution; mix:
 - Aluminum ammonium sulfate 95.0 g
 - Warm deionized or distilled water 600 ml
- b. Dye Solution; mix:
 - Reagent alcohol 48.0 ml
 - Deionized or distilled water 250 ml
 - Add:
 - Brazilin 9.6 g
 - Mix thoroughly with a magnetic stirrer.
- c. Iodate Solution; mix:
 - Sodium iodate 0.18 g
 - Warm deionized or distilled water 72 ml
- d. Acid Solution; mix:
 - Acetic acid 14.5 ml
 - Deionized or distilled water 30 ml

2. Stain preparation (mix thoroughly in given order)

- a. Combine Alum Solution and Dye Solution.
- b. Slowly add the Iodate Solution.
- c. Slowly add the Acid Solution.
- d. Check pH and adjust to 2.4-2.7 if necessary.
- e. Filter solution before use.

3. Staining

- a. Special stains should be applied before the nuclear stain. Acidic or alkaline solutions applied after the nuclear stain will cause destaining. Exposing sections to bluing agent or tap water before this nuclear stain is recommended when the preceding special stain has a pH below 3.5. The latter will provide darker nuclear staining.
- b. Staining time depends upon section thickness, fixation and desired effects. Suggested times are 5-10 minutes. A slight color shift will occur in the 70% alcohol.
- c. Rinse the nuclear stain using deionized or distilled water. Do not use tap water.

RECOMMENDED STAINING SCHEDULE

- 1. Clearant x 3 3 minutes each
- 2. 100% alcohol x 2 1 minute each
- 3. 95% alcohol 1 minute
- 4. 50% alcohol 1 minute
- 5. Distilled or deionized water x 2 30 seconds each
- 6. Apply special stain
- 7. Tap water 1 minute
- 8. Brazilin nuclear stain 5-10 minutes
- 9. Distilled or deionized water 1 minute
- 10. 70% alcohol 1 minute
- 11. 95% alcohol 1 minute
- 12. 100% alcohol x 3 1 minute each
- 13. Clearant x 3 1 minute each

Results: Nuclei—red

DISPOSAL

- 1. Use a licensed waste hauler.
- 2. Discard into the sanitary sewer system with the approval of local wastewater officials.

MSDS

MSDS are available online at www.anatechltdusa.com.

ORDERING INFORMATION FOR BRAZILIN/BRAZILLIANT!

<u>Cat#</u>	<u>Packaging</u>
860	Brazilin, 50 g jar
861	Brazilliant!, 1 quart

Effective: February 2013 [R625]