

BODIAN'S METHOD FOR NERVE FIBERS & NERVE ENDINGS (Microwave)

PURPOSE:	For In Vitro Diagnostic Use: Intended for the qualitative demonstration of nerve fibers.
PRINCIPLE:	Silver proteinate is taken up by nerve fibers while the copper metal in the solution inhibits the connective tissue from binding the silver. The silver is then reduced by the Hydroquinone Solution to the visible form. Gold Chloride displaces nonreacted silver ions and then is reduced by Oxalic Acid to increase staining intensity. Sodium Thiosulfate removes any loose metallic ions and Aniline Blue is used as a counterstain.
CONTROL:	Normal cerebral cortex <i>Control Slides can be purchased from Histology Control Systems. See inside back cover, Item# cs031.</i>
SPECIMEN PREPARATION:	Formalin fixed, paraffin embedded sections cut at 6-8 micrometers
SOLUTIONS:	<ol style="list-style-type: none"> Protargol Dry Pack Item# s2014-3x1GM See directions below.* Bodian's Reducing Solution, See directions below.* Gold Chloride 1% Aqueous Item# s203 Oxalic Acid 2% Aqueous Item# s2012 Sodium Thiosulfate 5% Aqueous Item# s1895 Aniline Blue Bodian's Method Item# s117 Copper Wire Item# P206-5GM <p><i>Solutions can be purchased separately from Poly Scientific.</i></p>
NOTES:	<p>*These solutions must be made fresh: Protargol Solution: Protargol Powder 3x1 g, Distilled Water 3x100 mL. Sprinkle 1 gram Protargol onto surface of 100 mL of distilled water. Allow it to dissolve. Discard after use. Bodian's Reducing Solution: Hydroquinone 3x1 g, Formaldehyde 3x5 mL, Distilled Water 3x100 mL. Mix 1 gram of Hydroquinone & 5 mL of Formaldehyde with 100 mL of distilled water. Expiration date: 2-3 weeks after reconstitution.</p> <p>Copper wire must first be cleaned with a solution composed of 15 mL of Hydrochloric Acid and 5 mL of Nitric Acid then rinsed in distilled water several times. The copper wire may be cleaned and reused.</p>
REFERENCE:	Clark, George. <u>Staining Procedures</u> . 4th Ed. Williams & Wilkins. Baltimore, MD. 1981. p. 179.

STAINING PROCEDURE:

Note: Variations in timing may occur due to the power wattage of the microwave oven. Provided times and power levels are based on 1000 watt microwave oven.

- Deparaffinize and hydrate sections to distilled water.
- Preheat Protargol Solution in plastic coplin jar for 30 seconds. Then add copper wire and slides to solution.
- Allow to remain on counter in warm solution for 5-10 minutes.
- Rinse slides in 3 changes of distilled water.
- Place slides in Reducing Solution for 10 minutes.
- Rinse in 3 changes of distilled water.
- Tone sections in Gold Chloride 1% Aqueous for 5 minutes.
- Rinse sections in 3 changes of distilled water.
- Develop in Oxalic Acid 2% Aqueous, check with microscope until the background is gray and nerve fibers appear clearly stained (3-5 minutes).
- Rinse in 3 changes of distilled water.
- Treat sections with Sodium Thiosulfate 5% Aqueous for 5 minutes.
- Rinse in distilled water.
- Counterstain if desired in Aniline Blue Bodian Method.
- Dehydrate rapidly in 95% Alcohol, 100% Alcohol and clear in 2 changes of Xylene.
- Mount with Poly Mount (Item# s2153) or any other acceptable mounting medium.

RESULTS:

Nerve Fibers..... Black
 Background..... Blue
 Nuclei..... Black

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