

WARTHIN STARRY METHOD FOR SPIROCHETES & OTHER MICROORGANISMS

PURPOSE:	For In Vitro Diagnostic Use: Intended for the qualitative demonstration of Spirochetes and other microorganisms.
PRINCIPLE:	The Spirochetes absorb silver from the Silver Solution. Hydroquinone reduces the silver to the visible metallic form.
CONTROL:	Any tissue known positive for Spirochetes <i>Control Slides can be purchased from Histology Control Systems. See inside back cover, Item# cs012.</i>
SPECIMEN PREPARATION:	Formalin fixed (do not use chromatin fixatives), paraffin embedded sections cut at 6 micrometers
SOLUTIONS:	<ol style="list-style-type: none"> 1. Acidulated Water Item# s108D 2. Silver Nitrate 1% Item# s2165 3. Silver Nitrate 2% Item# s2166 4. Gelatin 5% Solution Item# s190B 5. Hydroquinone 0.15% Solution Acidified Item# s1834 <p><u>Developer Solution:</u> Must be prepared fresh. Use chemically clean glassware. Combine in the order given: Silver Nitrate 2% 1.5 mL Gelatin 5% Solution 3.75 mL Hydroquinone 0.15% Solution Acidified 2.0 mL</p> <p><i>Solutions can be purchased separately from Poly Scientific.</i></p>
NOTES:	Solutions may be mixed at room temperature. For a faster reaction preheat the Silver Nitrate 2%, Gelatin 5% and Hydroquinone 0.15% Aqueous in separate flasks in a flotation bath at 54°C during step 3. Do not use any metal. If overstained or too many Lymphocytes, put in Developer Solution for 2 minutes.
REFERENCE:	Luna, Lee G. <u>Manual of Histologic Staining Methods of the Armed Forces Institute of Pathology</u> . 3rd Ed. McGraw-Hill Book Co. New York. 1968. pp. 238-240.

STAINING PROCEDURE:

1. Deparaffinize and hydrate to Acidulated Water.
2. Impregnate with Silver Nitrate 1% heated in a flotation bath to 43°C for 30 minutes.
Prepare Developer Solution at this point. (See Solutions section)
3. Flood sections that have been laid across glass rods, with the Developer Solution, which must be used as soon as it is mixed. Allow sections to develop until they are light brown or yellow. Check known control under microscope. The Spirochetes should be black and the background light brown or yellow.
4. Wash quickly and thoroughly in hot water, approximately 56°C.
5. Dehydrate in 95% Alcohol, Absolute Alcohol and clear in Xylene, 2 changes each.
6. Mount with Poly Mount (Item# s2153) or any acceptable mounting medium.

RESULTS:

Spirochetes, Other Microorganisms..... Black
Background Pale Yellow to Light Brown

Poly Scientific R&D Corp.

Revision: B-18

