## ZIEHL-NEELSEN METHOD FOR ACID FAST BACTERIA (Microwave)

PURPOSE:	For In Vitro Diagnostic Use: Intended for the qualitative demonstration of acid fast organsims.	
PRINCIPLE:	The staining solution, along with the aid of heat, is able to temporarily weaken the lipid shell of certain organisms and dissolve into it. The Acid Alcohol is unable to destain these organisms but it does destain all non acid fast organisms.	
CONTROL:	Any tissue known positive for acid fast organisms Control Slides can be purchased from Histology Control Systems. See inside back cover, Item# cs003.	
SPECIMEN PREPARATION:	Any well-fixed tissue, paraffin embedded sections cut at 5 micrometers	
SOLUTIONS:	<ol> <li>Carbol Fuchsin Ziehl-Neelsen Item# s162</li> <li>Acid Alcohol 1% Item# s104</li> <li>Methylene Blue Working Item# s188B</li> <li>Solutions can be purchased separately from Poly Scientific.</li> </ol>	
NOTES:	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.	
REFERENCE:	Clark, George. <u>Staining Procedures</u> . 4th Ed. Williams & Wilkins. Baltimore, MD. 1981. p. 380.	

## STAINING PROCEDURE:

- 1. Deparaffinize and hydrate to distilled water.
- 2. Place 50 mL of Carbol Fuchsin Ziehl-Neelsen in a plastic coplin jar (loosely apply cap) and microwave for 20 seconds. Stir and re-microwave for 10 seconds. Stir again and add slides to hot solution for 5 minutes.
- 3. Rinse well in running water.
- 4. Decolorize in Acid Alcohol 1% until sections are pale pink.
- 5. Rinse well in running water. Check control.
- 6. Counterstain in Methylene Blue Working Solution until sections are pale blue, one slide at a time.
- 7. Rinse well in distilled water.
- 8. Dehydrate, clear and mount with Poly Mount (Item# s2153) or any other acceptable mounting medium.

## **RESULTS**:

Acid Fast Bacilli	Bright Red
Background	Pale Blue

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