

Steiner Staining Kit

(Cat# K1435-125; for Spirochetes; Store at Multiple Temperatures)

I. Introduction:

The **Steiner Staining Kit** (for Spirochetes) is designed for demonstrating Fungi, Helicobacter Pylori, Legionella pneumophila, and Spirochete infected tissue. Kit may be used on formalin fixed, paraffin-embedded tissue as well as frozen sections.

Spirochetes: Black to Brown

Helicobacter Pylori: Black to Brown

Fungi: Black to Brown

Legionella pneumophila Black to Brown

Background: Yellow to Tan

II. Application:

- Histological applications
- For *in vitro* diagnostic use

III. Sample Type:

- Formalin fixed, paraffin-embedded as well as frozen tissue sections.
- Control Tissue: Helicobacter Pylori infected stomach.

IV. Kit Contents:

Components	K1435-125	Part Number	Storage Temperature
Oxidizer Solution	125 ml	K1435-125-1	RT
Zinc Formalin Solution	125 ml	K1435-125-2	RT
Gum Mastic Solution	125 ml	K1435-125-3	2-8°C
Hydroquinone	1.5 gm	K1435-125-4	RT
Silver Nitrate Solution (0.2%)	125 ml	K1435-125-5	2-8°C
Silver Nitrate Solution (1%)	9 ml	K1435-125-6	2-8°C

V. User Supplied Reagents and Equipment:

- Distilled water
- Coplin jars
- Forceps
- Filter paper
- Water bath
- Absolute alcohol
- Synthetic resin

VI. Shipment and Storage:

All the reagents are shipped at room temperature and stored at multiple temperatures.

VII. Reagent Preparation:

- Do not use if reagents become cloudy.
- Use caution when handling reagents.
- Non-Sterile

VIII. Procedure (Standard):

Prepare Reducing Solution at Step #10 of this Procedure:

Combine:

25 ml 1% Hydroquinone (0.25 gm Hydroquinone in 25 ml Distilled Water)

15 ml Gum Mastic Solution (2.5%)

Mix thoroughly and filter through medium filter paper.

Then Add:

6 Drops (240 µl) Silver Nitrate Solution (1%)

Mix thoroughly.

1. Preheat Water Bath to 70°C.
2. Deparaffinize sections if necessary and hydrate to distilled water.
3. Incubate slide in Oxidizer Solution for 20 min.
4. Rinse thoroughly in distilled water. *Note: Place 20 ml of Silver Nitrate Solution (0.2%) in water bath to preheat.*
5. Incubate slide in Zinc Formalin Solution for 5 minutes.
6. Rinse thoroughly in distilled water.
7. Incubate slide in preheated Silver Nitrate Solution (0.2%) for 5 minutes at 70° Centigrade. *Note: Discard solution after this step*
8. Rinse slide thoroughly in distilled water.
9. Dehydrate slide in 2 changes of Absolute Alcohol. ***Note: Prepare Reducing Solution (above) and place in water bath to preheat.***
10. Incubate slide in Gum Mastic Solution for 3 min.
11. Air dry slide for 1 minute or until gum mastic is completely dry.
12. Incubate slide in preheated Reducing Solution for 10-15 minutes or until section is tan to brown at 70° Centigrade. *Note: Discard solution after this step*
13. Rinse slide quickly in distilled water.
14. Dehydrate quickly in 3 changes of absolute alcohol.
15. Clear, and mount in synthetic resin.

Procedure (Microwave):

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Prepare Reducing Solution at Step #10 of this Procedure:

Combine:

25 ml 1% Hydroquinone (0.25 gm Hydroquinone in 25 ml Distilled Water)

15 ml Gum Mastic Solution (2.5%)

Mix thoroughly and filter through medium filter paper.

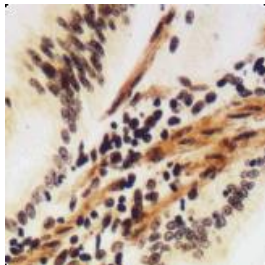
Then Add:

6 Drops (240 µl) Silver Nitrate Solution (1%)

Mix thoroughly.

1. Deparaffinize sections if necessary and hydrate to distilled water.
2. Incubate slide in Oxidizer Solution for 20 minutes.
3. Rinse thoroughly in distilled water.
4. Incubate slide in Zinc Formalin Solution for 5 minutes.
5. Rinse thoroughly in distilled water. *Note: In a loosely capped Slide Jar heat 20ml of Silver Nitrate Solution (0.2%) in a microwave oven for 10 seconds at full power. Repeat as needed until solution is hot, but do not allow solution to boil. Remove Slide Jar from microwave, tighten cap and agitate to equalize temperature.*
6. Incubate slide in hot Silver Nitrate Solution (0.2%) for 2 min with occasional agitation. *Note: Discard solution after this step.*
7. Rinse slide thoroughly in distilled water.
8. Dehydrate slide in 2 changes of Absolute Alcohol.
9. Incubate slide in Gum Mastic Solution for 3 min. *Note: Prepare Reducing Solution (above).*
10. Air dry slide for 1 min or until gum mastic is completely dry.
11. In a loosely capped Slide Jar heat 20ml of Reducing Solution in a microwave oven for 10 seconds at full power. Repeat as needed until solution is hot, but do not allow solution to boil. Remove Slide Jar from microwave, tighten cap and agitate to equalize temperature.
12. Place slide in loosely capped Slide Jar and return to microwave. As before heat Slide Jar containing slide for 10 seconds at full power. Repeat as needed until solution is hot, but do not allow solution to boil. Incubate slide in hot Reducing Solution for 3 min and then reheat again at full power until solution is hot. Incubate slide for an additional 2-3 min or until section is tan to brown. *Note: Discard solution after this step.*
13. Rinse slide quickly in distilled water.
14. Dehydrate quickly in 3 changes of absolute alcohol.
15. Clear, and mount in synthetic resin.

IX. Data:



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FOR RESEARCH USE ONLY! Not to be used on humans.