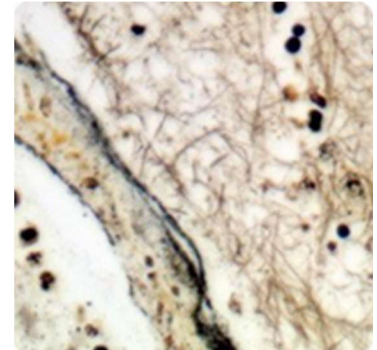


Bielschowsky's Stain Kit (Modified)

Description

The Bielschowsky's Stain Kit (Modified) is designed for histological visualization of nerve fibers, neurofibrillary tangles and senile plaques in Alzheimer's disease.

Axons:	Black
Neurofibrillary Tangles:	Black
Senile Plaques:	Black
Nuclei:	Dark Brown
Background:	Yellow to Light Brown



Uses/Limitations

Not to be taken internally.
 For In-Vitro Diagnostic use only.
 Histological applications.
 Do not use if reagents become cloudy.
 Do not use past expiration date.
 Use caution when handling reagents.
 Non-Sterile

Control Tissue

Cerebral cortex (cut 8-10µm)

Kit Contents

Catalog	Product	Volume	Storage
HSNZ500	Silver Nitrate Solution (20%)	500 ml	2-8°C
HFRL008	Formalin Solution (20%)	8 ml dropper	18-25°C
HCAS008	Citric Acid Solution (Bielschowsky's)	8 ml Dropper	18-25°C
HNAS008	Nitric Acid Solution (Bielschowsky's)	8 ml Dropper	18-25°C
HSTB125	Sodium Thiosulfate Solution (5%)	125 ml	18-25°C

Required but Not Included

Concentrated Ammonium Hydroxide.

Storage

Mixed storage conditions. Store according to individual label instructions.

Precautions

Avoid contact with skin and eyes.
 Harmful if swallowed.
 Follow all Federal, State, and local regulations regarding disposal.

Procedure

1. Prepare Ammoniacal Silver Solution. Prepare working solution using chemically cleaned glassware in a chemical fume hood as follows:
 - a. Pour 25-50ml of Silver Nitrate Solution (20%) into container (volume used is dependent on amount required to adequately fill staining container).
 - b. Add concentrated ammonium hydroxide (not included in kit); drop by drop, while swirling the flask continuously, until precipitate just dissolves and the reagent goes clear.

NOTE: If a small excess of ammonium hydroxide is added and solution will not go completely clear, filter the solution using a paper filter prior to use.

Use extreme care in preparation and use of Ammoniacal Silver Solution. Use mixture once and dispose, observing all local, state and federal laws.

2. Prepare Developer Solution. Use chemically cleaned glassware immediately prior to use as follows:
 - a. 50 ml Distilled Water.
 - b. 8 Drops Formalin Solution (20%). Add and swirl.
 - c. 8 Drops Citric Acid Solution (Bielschowsky's). Add and swirl.
 - d. 4 Drops Nitric Acid Solution (Bielschowsky's). Add and swirl.
3. Prepare Ammonia Water. Mix 320 μ l (8 drops) of concentrated Ammonium Hydroxide (not included) in 50 ml of distilled water.
4. Preheat waterbath to 40°C
5. Deparaffinize sections if necessary and hydrate to distilled water.
6. Place a chemically cleaned staining jar containing 25ml of Silver Nitrate Solution (20%) in waterbath and allow temperature to equilibrate for 10 minutes.
7. Place slide in warmed Silver Nitrate Solution (20%) and incubate for 15 minutes at 40°C.
8. During incubation place Ammoniacal Silver Solution in waterbath in allow temperature to equilibrate.
9. Remove slide from Silver Nitrate Solution (20%) and rinse in 4 changes of distilled water.
10. Place slide in warmed Ammoniacal Silver Solution and incubate for 10 minutes at 40°C.
11. Remove slide from Ammoniacal Silver Solution, shake off excess and

place directly into Developer Solution. Agitate gently until tissue section takes on a yellow/brown hue (5-20 seconds).

12. Remove slide from Developer Solution and immediately place in Ammonia Water for 30 seconds.
13. Rinse in 4 changes of distilled water.
14. Apply adequate Sodium Thiosulfate Solution (5%) to completely cover tissue section and incubate for 2 minutes.
15. Rinse in 4 changes of distilled water.
16. Dehydrate in 3 changes of absolute alcohol for 2 minutes each.
17. Clear, and mount in synthetic resin.