

Bielschowsky Control Slides – Technical Memo

CONTROL SLIDES:	Part 4058A	Part 4058B
	10 Slide/Set	98 Slide/Set

PRODUCT SPECIFICATIONS:

Tissue: Positive staining brain.

Fixation: Formalin 10%, Phosphate Buffered (Part 1090).

Section/Glass: Paraffin sections cut at 8 microns on Superfrost™ Plus slides.

Quality Control Stain: Bielschowsky quality control stained slide(s) included.

Reactivity: Guaranteed product specific reactivity for one year from date of receipt. Revalidate after one year to verify continued reactivity.

Storage: 15-30°C in a light deprived and humidity controlled environment.

Intended Use: To verify histological techniques and reagent reactivity.

Before using unstained control slides, review the enclosed stained slide(s) to ensure that this tissue source is acceptable for testing needs.

CONTROL SLIDE VALIDATION:

With Bielschowsky, Lester King Modified Stain Kit:	Part 9154A	Individual Stain Solution
Solution A: Silver Nitrate 20%, Aqueous	250 ml	Part 13807
Solution B: Ammonium Hydroxide 28-30%, ACS	100 ml	Part 1006
Solution C: Developer	25 ml	
Solution D: Sodium Thiosulfate 5%, Aqueous	250 ml	Part 1389

APPLICATION:

Newcomer Supply Bielschowsky Control Slides are for the positive histochemical staining of nerve fibers, neurofibrils/tangles, senile plaques and axons, instrumental in the diagnosis of Alzheimer's disease and other neurological disorders.

PRESTAINING PREPARATION:

- Heat dry sections in oven according to your laboratory protocol.
- All glassware/plasticware must be acid cleaned prior to use.
 - See Procedure Notes #1 and #2.
- Preheat Coplin jar of Solution A: Silver Nitrate 20%, Aqueous in water bath to 37°C.
- Preheat two Coplin jars of distilled water in 37°C water bath.
 - Save for slide rinsing/holding in Steps #7 and #10.

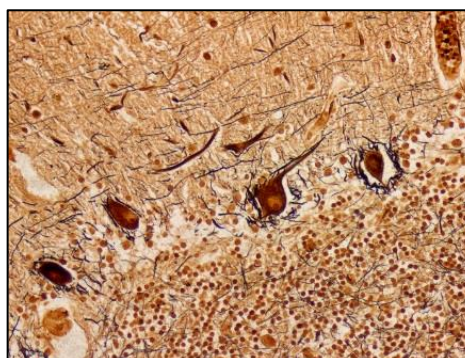
NEWCOMER SUPPLY VALIDATION PROCEDURE:

- Deparaffinize sections thoroughly in three changes of xylene, 3 minutes each. Hydrate through two changes each of 100% and 95% ethyl alcohols, 10 dips each. Wash well with distilled water.
 - See Procedure Notes #3 and #4.
- Place slides in preheated Solution A: Silver Nitrate 20%, Aqueous (Step #3) for 15 minutes.
 - See Procedure Note #5.
- Remove slides from Solution A: Silver Nitrate 20%, Aqueous and hold slides in warmed distilled water.
 - Save Silver Nitrate 20%, Aqueous for Step #8.
- Add Solution B: Ammonium Hydroxide 28-30%, ACS drop by drop in saved Silver Nitrate 20%, Aqueous swirling until precipitate disappears. **Do not go past this point.**
 - Approximately 10 ml of Ammonium Hydroxide 28-30%, ACS will be required.
- Place slides back into the Silver Nitrate Solution with added Ammonium Hydroxide in water bath at 37°C for 10 minutes.
- Remove slides and hold in preheated distilled water.
 - Save Ammoniacal Silver Solution for Step #11.
- Add 1 drop of Solution C: Developer to the saved Ammoniacal Silver Solution with swirling motion.
- Return slides to Ammoniacal Silver Solution with added Developer, in 37°C water bath for 5-15 minutes; average time of 6 minutes.
 - Check slides microscopically at 3 minutes for development of neurons to dark brown.
 - Follow with checks at 1 minute intervals to avoid silver over-development.
- Rinse thoroughly in distilled water for 5 minutes.
- Place in Solution D: Sodium Thiosulfate 5%, Aqueous; 5 minutes.
- Rinse thoroughly in tap water.

- Dehydrate in two changes each of 95% and 100% ethyl alcohol. Clear in three changes of xylene, 10 dips each; coverslip with compatible mounting medium.

RESULTS:

Senile plaques, neurofibrils/tangles	Dark brown to black
Neurons	Dark brown
White and gray matter	Yellowish brown
Nerve fibers, axons	Brown to black



PROCEDURE NOTES:

- Acid clean all glassware/plasticware (12086) and rinse thoroughly in several changes of distilled water.
- Plastic (5500), plastic-tipped or paraffin coated metal forceps must be used with silver solutions to prevent precipitation of silver salts. No metals of any kind should come in contact with silver solutions. Only glass thermometers should be used.
- Drain slides after each step to prevent solution carry over.
- Do not allow sections to dry out at any point during procedure.
- A maximum of 8 slides per 40 ml of Solution A: Silver Nitrate 20%, Aqueous is recommended for proper silver development.
- If using a xylene substitute, closely follow the manufacturer's recommendations for deparaffinization and clearing steps.

REFERENCES:

- Carson, Freida L., and Christa Hladik Cappellano. *Histotechnology: A Self-instructional Text*. 4th ed. Chicago: ASCP Press, 2015. 196-199.
- King, Lester. "The Impregnation of Neurofibrils". *Yale Journal of Biology and Medicine* 14.1 (1941). 59-68.
- Modifications developed by Newcomer Supply Laboratory.