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Part 4100
Revised March 2025

Calcium Control Slides – Technical Memo

CONTROL SLIDES:

Part 4100A	**Part 4100B**
10 Slide/Set	98 Slide/Set

PRODUCT SPECIFICATIONS:
Tissue: Positive staining placenta.
Fixation: Formalin 10%, Phosphate Buffered (Part 1090).
Section/Glass: Paraffin sections cut at 4 microns on Superfrost™ Plus slides.
Quality Control Stain: Von Kossa Calcium 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 Von Kossa Calcium Stain:	**Individual Stain Solution**
Silver Nitrate 5%, Aqueous	Part 13805
Sodium Thiosulfate 5%, Aqueous	Part 1389
Nuclear Fast Red Stain, Kernechtrot	Part 1255

APPLICATION:

Newcomer Supply Calcium Control Slides are for the positive histochemical staining of calcium or calcium salts in tissue sections.

PRESTAINING PREPARATION:

1. Heat dry sections in oven according to your laboratory protocol.
2. All glassware/plasticware must be acid cleaned prior to use.
    a. *See Procedure Notes #1 and #2*

NEWCOMER SUPPLY VALIDATION PROCEDURE:

3. 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.
    a. *See Procedure Notes #3 and #4.*
4. Place slides in Silver Nitrate 5%, Aqueous according to the following timings and conditions.
    a. *Direct sunlight or ultraviolet light for 10-30 minutes.*
    b. *In front of a 60-100 watt light bulb for 1 hour or longer.*
    c. *See Procedure Note #5.*
5. Check slides periodically and remove from light source when control slide shows black-brown deposits macroscopically.
6. Rinse well in several changes of distilled water.
7. Place slides in Sodium Thiosulfate 5%, Aqueous for 2 minutes.
8. Rinse well in several changes of distilled water.
9. Counterstain in Nuclear Fast Red Stain, Kernechtrot for 5 minutes.
    a. *Shake solution well before use; do not filter.*
10. Rinse well in distilled water.
    a. *See Procedure Note #6.*
11. Dehydrate in two changes each of 95% and 100% ethyl alcohol; 10 dips each. Clear in three changes of xylene, 10 dips each; coverslip with compatible mounting medium.

RESULTS:

Calcium salts	Black to brown/black
Nuclei	Red
Cytoplasm	Light pink

PROCEDURE NOTES:

1. Acid clean all glassware/plasticware (Part 12086) and rinse thoroughly in several changes of distilled water.
2. No metals of any kind should come in contact with silver solutions to prevent precipitation of silver salts. Use plastic forceps (5500) or paraffin coated metal forceps.
3. Drain slides after each step to prevent solution carry over.
4. Do not allow sections to dry out at any point during procedure.
5. Direct sunlight is the preferred method. If procedure is in minimal sunlight increased incubation time will be necessary.
6. Wash well after Nuclear Fast Red Stain, Kernechtrot to avoid cloudiness in dehydration steps.
7. If using a xylene substitute, follow manufacturer's recommendation for deparaffinization and clearing steps.

REFERENCES:

1. Carson, Freida L. and Christa Hladik. *Histotechnology: A Self-Instructional Text.* 3rd ed. Chicago, Ill.: American Society of Clinical Pathologists, 2009. 269-270.
2. Sheehan, Dezna C. and Barbara B. Hrapchak. *Theory and Practice of Histotechnology*. 2nd ed. St. Louis: Mosby, 1980. 226-227.
3. Modifications developed by Newcomer Supply Laboratory.