Copper, Animal Control Slides – Technical Memo

CONTROL SLIDES:

<table>
<thead>
<tr>
<th>Product Code</th>
<th>Description</th>
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<tbody>
<tr>
<td>Part 4130A</td>
<td>10 Slide/Set</td>
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<tr>
<td>Part 4130B</td>
<td>98 Slide/Set</td>
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PRODUCT SPECIFICATIONS:

Tissue: Positive staining animal liver.
Fixation: Formalin 10%, Phosphate Buffered (Part 1090).
Section/Glass: Paraffin sections cut at 4 microns on Superfrost™ Plus slides.
Quality Control Stain: Rhodanine 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 histotechnical 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.

APPLICATION:

Newcomer Supply Copper, Animal Control Slides are for the positive histochemical detection of copper in tissue sections.

PRESTAINING PREPARATION:

1. Heat dry sections in oven according to your laboratory protocol.
2. Prepare Working Rhodanine Solution; combine and mix well.
   a. Shake Solution A: Rhodanine Stock Stain 0.2%, Alcoholic well before each use.
   b. Solution A: Rhodanine Stock Stain 0.2%, Alcoholic 3 ml
   c. Distilled Water 47 ml

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 alcohol, 10 dips each. Wash well with distilled water.
   a. See Procedure Notes #1 and #2.
4. Stain in Working Rhodanine Solution (Step #2) at 60°C for 1-2 hours or at 37°C for 18 hours.
   a. Microwave Modification: See Procedure Note #3.
   b. Place slides in a plastic Coplin jar containing Working Rhodanine Solution and microwave for 6 minutes at 70°C.
5. At the end of incubation (for both oven and microwave), to avoid unwanted slide precipitate, pour off warm Working Rhodanine Solution into a second Coplin jar; reserve and set aside.
6. Rinse slides well in several changes of distilled water.
7. Check positive control slide microscopically to determine adequate copper/reddish brown development.
   a. Return slides to reserved Working Rhodanine Solution if additional incubation is required.
8. Prepare dilute Mayer Hematoxylin Stain Solution directly before use; combine and mix well:
   a. Solution B: Hematoxylin Stain, Mayer Modified 20 ml
   b. Distilled Water 20 ml
9. Stain in dilute Mayer Hematoxylin Stain Solution for 10 minutes.
10. Rinse in distilled water.
11. Rinse in Solution C: Sodium Borate 0.5%, Aqueous; 2-3 quick dips.
12. Rinse well in distilled water.
13. 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:

<table>
<thead>
<tr>
<th>Component</th>
<th>Color</th>
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<tbody>
<tr>
<td>Copper</td>
<td>Copper/reddish brown</td>
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<tr>
<td>Nuclei</td>
<td>Light blue</td>
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</tbody>
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PROCEDURE NOTES:

1. Drain slides after each step to prevent solution carry over.
2. Do not allow sections to dry out at any point during procedure.
3. The suggested microwave procedure has been tested at Newcomer Supply. This procedure is a guideline and techniques should be developed for use in your laboratory.
4. If using a xylene substitute, closely follow the manufacturer’s recommendations for deparaffinization and clearing steps.

REFERENCES:

4. Modifications developed by Newcomer Supply.