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Part 4691 Revised January 2020

# Trichrome, Kidney Control Slides – Technical Memo

CONTROL SLIDES:

Part 4691A 10 Slide/Set Part 4691B 98 Slide/Set

## PRODUCT SPECIFICATIONS:

**Tissue:** Positive staining kidney. **Fixation:** Formalin 10%, Phosphate Buffered (Part 1090).

Section/Glass: Paraffin sections cut at 4 microns on Superfrost™ Plus slides.

Quality Control Stain: Gomori One-Step Aniline Blue guality 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 Trichrome, Gomori One-Step, Aniline Blue Stain Kit:	Part 9176B/A	Individual Stain Solution
Solution A: Bouin Fluid	250/500 ml	Part 1020
Solution B: Ferric Chloride, Acidified	125/250 ml	Part 1409
Solution C: Hematoxylin 1%, Alcoholic	125/250 ml	Part 1409
Solution D: Trichrome Stain, Gomori One-Step, Aniline Blue	250/500 ml	Part 1403
Solution E: Acetic Acid 0.5%, Aqueous	250/500 ml	Part 100121

#### APPLICATION:

Newcomer Supply Trichrome, Kidney Control Slides are for the positive histochemical staining of connective tissue and to differentially demonstrate collagen and muscle fibers.

#### **PRESTAINING PREPARATION:**

- 1. Heat dry sections in oven according to your laboratory protocol.
- 2. Preheat Solution A: Bouin Fluid to 56-60°C in oven or water bath. (*Skip if using overnight method or microwave procedure.*)

#### 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.
   a. See Procedure Notes #1 and #2.
- Mordant in preheated Solution A: Bouin Fluid (Step #2) for one hour at 56-60°C or overnight at room temperature. Cool at room temperature for 5-10 minutes.

a. Skip Step #4 if tissue was originally Bouin fixed.

- Microwave Modification: See Procedure Note #3.
  - b. Place slides in a <u>plastic</u> Coplin jar containing Solution A: Bouin Fluid and microwave for 5 minutes at 60°C.
- 5. Wash well in running tap water; rinse in distilled water.
- 6. Prepare fresh Weigert Iron Hematoxylin; combine and mix well.
  - a. Solution B: Ferric Chloride, Acidified 20 ml
    - b. Solution C: Hematoxylin 1%, Alcoholic 20 ml
  - Stain in fresh Weigert Iron Hematoxylin for 10 minutes.
- Wash in running tap water for 10 minutes; rinse in distilled water.
  a. See Procedure Note #4.
- 9. Stain with Solution D: Trichrome Stain, Gomori One-Step, Aniline Blue for 20 minutes.
- 10. Differentiate in Solution E: Acetic Acid 0.5%, Aqueous; 2 minutes.
- 11. Rinse quickly in distilled water.
- 12. 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:**

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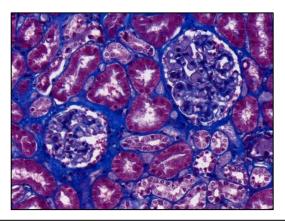
Collagen and mucin	Blue
Muscle fibers, cytoplasm and keratin	Red
Nuclei	Blue/black

### 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 Weigert Iron Hematoxylin is not completely washed from tissue sections, nuclear and cytoplasmic staining may be compromised.
- Trichrome, Kidney Control Slides are validated with Trichrome Stain Kit, Gomori One-Step, Aniline Blue but can be used as positive controls with any preferred Trichrome procedure.
- 6. If using a xylene substitute, closely follow the manufacturer's recommendations for deparaffinization and clearing steps.

#### **REFERENCES:**

- Brown, Richard. Histologic Preparations: Common Problems and Their Solutions. Northfield, Ill.: College of American Pathologists, 2009. 95-101.
- Carson, Freida L., and Christa Hladik. *Histotechnology: A Self-Instructional Text.* 3rd ed. Chicago, Ill.: American Society of Clinical Pathologists, 2009. 165-166.
- Sheehan, Dezna C., and Barbara B. Hrapchak. *Theory and* Practice of Histotechnology. 2nd ed. St. Louis: Mosby, 1980. 191-192.
- 4. Vacca, Linda L. *Laboratory Manual of Histochemistry*. New York: Raven Press, 1985. 308-310.
- 5. Modifications developed by Newcomer Supply Laboratory.



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