

## Trichrome, Liver Control Slides – Technical Memo

<b>CONTROL SLIDES:</b>	<b>Part 4690A</b>	<b>Part 4690B</b>
	10 Slide/Set	98 Slide/Set

### PRODUCT SPECIFICATIONS:

**Tissue:** Positive staining liver.

**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 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:

<b>With Trichrome, Gomori One-Step, Aniline Blue Stain Kit:</b>	<b>Part 9176A/B</b>	<b>Individual Stain Solution</b>
Solution A: Bouin Fluid	250/500 ml	Part 1020
Solution B: Ferric Chloride, Aqueous	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, Liver Control Slides are for the positive histochemical staining of connective tissue and to differentially demonstrate collagen and muscle fibers.

### PRESTAINING PREPARATION:

- Heat dry sections in oven according to your laboratory protocol.
- 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.
  - 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.
  - Skip Step #4 if tissue was originally Bouin fixed.

**Microwave Modification:** See Procedure Note #3.

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

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

### PROCEDURE NOTES:

- Drain slides after each step to prevent solution carry over.
- Do not allow sections to dry out at any point during procedure.
- 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.
- If Weigert Iron Hematoxylin is not completely washed from tissue sections, nuclear and cytoplasmic staining may be compromised.
- Trichrome, Liver 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.
- 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.
- Vacca, Linda L. *Laboratory Manual of Histochemistry*. New York: Raven Press, 1985. 308-310.
- Modifications developed by Newcomer Supply Laboratory.

