

Amyloid, Animal Control Slides – Technical Memo

CONTROL SLIDES:	Part 4031A	Part 4031B
	10 Slide/Set	98 Slide/Set

Amyloid, Animal Control Slides contain a section of positive staining animal organ.

PRODUCT DESCRIPTION:

The enclosed positive control slides are intended to be used to verify histological techniques and reagent reactivity. They are to be used for the qualitative purpose of determining positive or negative results, and are not intended to be used for any quantitative purpose. The first serial section within this control box is stained and provided for your reference. **Before using the unstained slides, review the enclosed stained slide with your pathologist to ensure that this tissue source is acceptable. Newcomer Supply will not accept a return with missing slides in the series. Newcomer Supply guarantees reactivity of these control slides for one year from the date of receipt. Revalidate after one year to verify continued reactivity. Store at 15-30°C in a light deprived and humidity controlled environment.**

The positive control sections were produced from animal tissues under carefully controlled conditions. Reasonable measures are used to deliver quality control slides that are as consistent as possible. However, characteristics of quality control slides may be dissimilar due to variations in the reagents, stains, techniques, laboratory conditions, and tissue sources used. Newcomer Supply Laboratory uses a manual method of performing quality control procedures, specifically avoiding automation, in order to provide reactive control slides for even less aggressive methods of staining that our customers may be using.

CONTROL SLIDE VALIDATION:

With Amyloid, Bennhold Congo Red Stain Kit:	Part 9103A	Individual Stain Solution
Solution A: Congo Red Stain 1%, Aqueous	250 ml	Part 1038
Solution B: Alkaline Alcohol	250 ml	Part 1038
Solution C: Hematoxylin Stain, Mayer Modified	250 ml	Part 1202

For storage requirements and expiration date refer to individual product labels.

APPLICATION:

Newcomer Supply Amyloid, Animal Control Slides are for the positive histochemical staining of extraneous protein deposits in amyloidosis.

METHOD:

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

Technique: Paraffin sections cut at 10 microns on Superfrost® Plus

Solutions: All solutions are manufactured by Newcomer Supply, Inc.

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.
- Place slides in Solution A: Congo Red Stain 1%, Aqueous for 1 hour.

Microwave Modification: See Procedure Note #3.

 - Place slides in a plastic Coplin jar (Part 5184) containing Solution A: Congo Red Stain 1%, Aqueous and microwave at 70°C for 3 minutes.
- Rinse in two to three changes of tap water; rinse in distilled water.
- Differentiate in Solution B: Alkaline Alcohol, 5 to 30 seconds, agitating constantly until slide background is cleared of Solution A: Congo Red Stain 1%, Aqueous.
- Rinse in two to three changes of tap water; rinse in distilled water.
- Counterstain with Solution C: Hematoxylin Stain, Mayer Modified, 3 to 5 minutes, depending on preference of nuclear stain intensity.
- Wash in running tap water for 5 to 10 minutes.
- 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:

Light Field Microscopy:

Amyloid	Pink to red
Nuclei	Blue

Polarized Light:

Amyloid fluorescence	Apple green
----------------------	-------------

PROCEDURE NOTES:

- Drain staining rack/slides after each step to prevent solution carry over.
- Do not allow sections to dry out at any point during staining procedure.
- The suggested microwave procedure has been tested at Newcomer Supply using an "EB Sciences", 850 watt microwave oven with temperature probe and agitation tubes. This procedure is reproducible in our laboratory. It is nonetheless a guideline and techniques should be developed for your laboratory which meet the requirements of your situation. Microwave devices should be placed in a fume hood or vented into a fume hood, according to manufacturer's instructions, to prevent exposure to chemical vapors.
- For optimal results sections should be cut at 8-10 microns. This will provide more intense staining and allow smaller amyloid deposits to be identified. Sections cut too thin may show faint staining and sections cut thicker than 8-10 microns may display yellow birefringence.
- If using a xylene substitute, closely follow the manufacturer's recommendations for deparaffinization and clearing steps.

REFERENCES:

- Luna, Lee G. *Histopathologic Methods and Color Atlas of Special Stains and Tissue Artifacts*. Gaithersburg, MD: American Histolabs, 1992. 366-367.
- Sheehan, Dezna C., and Barbara B. Hrapchak. *Theory and Practice of Histotechnology*. 2nd ed. St. Louis: Mosby, 1980. 177-178.
- Modifications developed by Newcomer Supply Laboratory.

