

## Acid Fast Bacteria (AFB), Animal Control Slides – Technical Memo

<b>CONTROL SLIDES:</b>	<b>Part 4011A</b>	<b>Part 4011B</b>
	10 Slide/Set	98 Slide/Set

Acid Fast Bacteria (AFB), Animal Control Slides contain sections of positive staining animal gastrointestinal tract and negative staining human lung.

### 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 and the negative control sections from human surgical or autopsy 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 AFB, Ziehl-Neelsen Stain Kit:	Part 9101A	Individual Stain Solution
Solution A: Carbol Fuchsin Stain, Ziehl-Neelsen	250 ml	Part 1030
Solution B: Acid Alcohol 1%	250 ml	Part 10011
Solution C: Methylene Blue Stain 0.14%, Alcoholic	250 ml	Part 12401

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

### APPLICATION:

Newcomer Supply Acid Fast Bacteria (AFB), Animal Control Slides are for the positive histochemical staining of acid-fast mycobacteria in tissue sections.

### METHOD:

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

**Technique:** Paraffin sections cut at 5 microns on Superfrost® Plus

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

### NEWCOMER SUPPLY VALIDATION PROCEDURE:

- Filter Solution A: Carbol Fuchsin Stain, Ziehl-Neelsen before use.
- 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.
- Stain in freshly filtered Solution A: Carbol Fuchsin Stain, Ziehl-Neelsen for 60 minutes at room temperature. Keep solution covered.
- Rinse in running tap water for 2 to 3 minutes.
- Differentiate in Solution B: Acid Alcohol 1% until color no longer runs off the slide and sections are pale pink; 3 to 10 rapid dips.
- Wash in running tap water 3 to 5 minutes; rinse in distilled water.
- Counterstain in Solution C: Methylene Blue Stain 0.14%, Alcoholic.
  - Dip slides a few times in counterstain; rinse in tap water, followed by a distilled water rinse and check microscopically. Sections should be pale blue.
  - See Procedure Notes #3 and #4.
- Wash in running tap water for 1 minute; rinse 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:

Acid-fast bacilli	Bright red
Background	Pale blue
Lung	Negative for AFB

### 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.
- It is important not to over-counterstain, as the organisms may be masked. If section is over-stained, remove methylene blue with acid alcohol, rinse thoroughly, and repeat methylene blue step (Step #7).
- If laboratory tap water is generally acidic, the methylene blue stain may be pale. Adjust staining time accordingly.
- If using a xylene substitute, closely follow the manufacturer's recommendations for deparaffinization and clearing steps.

### REFERENCES:

- Carson, Freida L., and Christa Hladik. *Histotechnology: A Self-Instructional Text*. 3rd ed. Chicago, Ill.: American Society of Clinical Pathologists, 2009. 226-227.
- Sheehan, Dezna C., and Barbara B. Hrapchak. *Theory and Practice of Histotechnology*. 2nd ed. St. Louis: Mosby, 1980. 236-237.
- Modifications developed by Newcomer Supply Laboratory.

