

Fite, *Nocardia sp.*, Artificial Control Slides – Technical Memo

CONTROL SLIDES:	Part 4215A	Part 4215B
	10 Slide/Set	98 Slide/Set

Fite, *Nocardia sp.*, Artificial Control Slides contain sections of positive staining rat lung and negative staining human lung.

PRODUCT DESCRIPTION:

The enclosed positive control slides are intended to be used to verify histological techniques and reagent reactivity. These slides 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 the 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.**

These Fite, *Nocardia sp.*, Artificial Control Slides were produced at the Newcomer Supply Laboratory under carefully controlled conditions. The positive control sections are not human tissue. The microorganisms were grown in pure culture, harvested, formalized and introduced in a freshly harvested rat lung. No infective process occurred. *Nocardia sp.* was used to produce these control slides, and was purchased from the American Type Culture Collection (ATCC® 700034™). 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, Fite Stain Kit:	Part 91013A	Individual Stain Solution
Solution A: Xylene/Peanut Oil, 2:1	500 ml	Part 1449
Solution B: Carbol Fuchsin Stain, Ziehl-Neelsen	250 ml	Part 1030
Solution C: Sulfuric Acid 1%, Aqueous	250 ml	Part 14012
Solution D: Methylene Blue Stain 0.5%, Working Aqueous	250 ml	Part 12402

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

APPLICATION:

Newcomer Supply Fite, *Nocardia sp.*, Artificial Control Slides are for the positive histochemical staining of *Nocardia sp.* and acid-fast bacilli in tissue sections.

METHOD:

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

Technique: Paraffin sections cut at 4 microns on Superfrost® Plus

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

NEWCOMER SUPPLY VALIDATION PROCEDURE:

1. Filter Solution B: Carbol Fuchsin Stain, Ziehl-Neelsen.
2. Deparaffinize slides in Solution A: Xylene/Peanut Oil, 2:1, two changes for 12 minutes each.
 - a. See Procedure Note #1.
3. Drain slides, wipe off excess oil, and blot to opacity taking care to remove residual oil.
 - a. See Procedure Note #2.
4. Stain slides in freshly filtered Solution B: Carbol Fuchsin Stain, Ziehl-Neelsen for 30 minutes at room temperature.
5. Wash in running tap water for 3 minutes.
6. Differentiate in Solution C: Sulfuric Acid 1%, Aqueous for 3 minutes.
7. Wash in running tap water for 3 minutes.
8. Counterstain lightly with Solution D: Methylene Blue Stain 0.5%, Working Aqueous for 5-10 seconds.
 - a. See Procedure Notes #3 and #4.
9. Rinse off excess Methylene Blue Stain in running tap water. Background should be a light sky blue.
10. Blot excess water from slide and air-dry completely.
11. Dip dried slides in xylene and coverslip with a compatible mounting medium.

RESULTS:

<i>Nocardia sp.</i>	Red
Acid-fast bacilli	Red
Red blood cells	Yellow-orange
Other tissue elements	Pale blue
Negative lung	Negative for <i>Nocardia sp.</i>

PROCEDURE NOTES:

1. Acid-fastness of the organisms is enhanced when the waxy capsule is protected by the mixture of xylene/peanut oil and the avoidance of dehydrating solutions.
2. It is important to blot well, residual oil may produce staining artifact.
3. If over-stained with methylene blue, organisms may be masked. Check microscopically before air drying. If over-stained, remove methylene blue with Acid Alcohol 1% (Part 10011); rinse thoroughly; repeat Step #8 with a shorter timing.
4. If laboratory tap water is generally acidic, the methylene blue stain may be pale. Adjust staining time accordingly.
5. A small percentage of *Nocardia sp.* organisms may resist taking the red stain and remain blue due to the growth phase of the individual organism.
6. If using a xylene substitute, closely follow the manufacturer's recommendations for coverslipping step.

REFERENCES:

1. Carson, Freida L., and Christa Hladik. *Histotechnology: A Self-Instructional Text*. 3rd ed. Chicago, Ill.: American Society of Clinical Pathologists, 2009. 228-229.
2. Fite, George, P.J. Cambre and M.H. Turner. "Procedure for Demonstrating Lepra Bacilli in Paraffin Sections". *Archives of Pathology* 43 (1947). 624-625.
3. Luna, Lee G. *Histopathologic Methods and Color Atlas of Special Stains and Tissue Artifacts*. Gaithersburg, MD: American Histolabs, 1992. 180-181.
4. Sheehan, Dezna C., and Barbara B. Hrapchak. *Theory and Practice of Histotechnology*. 2nd ed. St. Louis: Mosby, 1980. 237.
5. Modifications developed by Newcomer Supply Laboratory.