**ΝΕΨζΟΜΕΙ<u>Έ</u>ΛΡΡΓ**Υ°

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Part 3746 Revised January 2025

# Spirochete Treponema, Artificial Control Slides – Technical Memo

CONTROL SLIDES:	Part 3746A	Part 3746B
	10 Slide/Set	98 Slide/Set

# PRODUCT SPECIFICATIONS:

Tissue: Positive staining rat lung and negative staining human lung. Fixation: Formalin 10%, Phosphate Buffered (Part 1090). Section/Glass: Paraffin sections cut at 4 microns on Superfrost™ Plus slides. Quality Control Stain: Spirochete 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.

## **APPLICATION:**

Newcomer Supply Spirochete Treponema, Artificial Control Slides are for the positive immunohistochemical staining of spirochetes, the causative agent of a variety of diseases such as; syphilis, bejel, pinta, yaws and Lyme.

Treponema hyodysenteriae purchased from American Type Culture Collection is used to produce the positive control tissue.

# **NEWCOMER SUPPLY VALIDATION PROCEDURE:**

- 1. Heat dry sections in oven according to your laboratory protocol.
- Deparaffinize sections thoroughly in three changes of xylene, 3 2. minutes each. Hydrate through two changes each of 100% and 95% ethyl alcohols, 10 dips each. Wash well with distilled water. See Procedure Note #1. a.
- Proceed with an epitope/antigen retrieval technique approved for 3. use in your laboratory.
- Rinse in distilled water; tap off excess water. 4
- 5. Circle sections with Pap Pen Liquid Blocker (Part 6505, 6506 or 6507) to reduce reagent usage and ensure tissue coverage.
- Block endogenous peroxidase with freshly made 3% Hydrogen 6. Peroxide. Incubate for 5 minutes.
  - See Procedure Note #2. a.
- Wash slides gently in distilled water. Rinse in two changes of Tris 7. Buffered Saline. a.
  - See Procedure Note #3.
- Tap off excess buffer; apply Treponema pallidum primary antibody. 8. Incubate at room temperature for 30 minutes.
- Rinse slides in two changes of buffer. 9
- Tap off excess buffer; apply Amplifier. Incubate for 10 minutes. 10.
- Rinse slides in two changes of buffer. 11.
- Tap off excess buffer; apply HRP Polymer. Incubate for 10 minutes. 12.
- Rinse slides in two changes of buffer. 13.
- Prepare required quantity of DAB substrate/chromogen. 14.
- Tap off excess buffer; apply DAB. Incubate for 5 minutes. 15.
- Rinse slides in four changes of distilled water. 16.
- Counterstain with Hematoxylin Stain, Gill I (Part 1180); 1-10 dips. 17.
- Rinse slides in warm tap water to blue sections. 18.
- Dehydrate in two changes each of 95% and 100% ethyl alcohol. 19. Clear in three changes of xylene, 10 dips each; coverslip with compatible mounting medium.

#### **RESULTS:**

Spirochete positive expression	
Negative lung	
Nuclei	

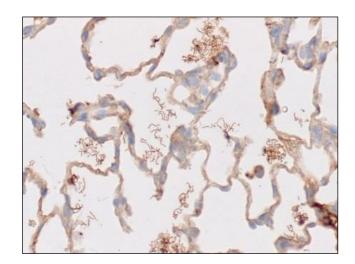
Brown Negative for spirochetes Blue

# PROCEDURE NOTES:

- Do not allow sections to dry out at any point during procedure. 1.
- Dilute Hydrogen Peroxide 30%, Aqueous (Part 1206) with distilled 2. water to a 3% (1/10) solution prior to use.
- Dilute Tris Buffered Saline 0.05M, pH 7.6, 10X (Part 140304) with 3. distilled water to a 1/10 solution prior to use for all buffer rinses.
- 4. Biocare Treponema pallidum Polyclonal is the primary antibody used along with Cell Marque detection and ancillary reagents.
- 5. If using a xylene substitute, follow manufacturer's recommendation for deparaffinization and clearing steps.

## **REFERENCES:**

- Biocare Treponema pallidum Antibody datasheet. 1.
- Modifications developed by Newcomer Supply Laboratory. 2.



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