

TECHNICAL DATA SHEET 482

Fungi-Fluor[®]Pneumocystis Kit

Catalog No. 22363

DESCRIPTION

Pneumocystis carinii is a unicellular, eukaryotic organism which is present in the lungs of many mammalian species and can cause Pneumocystis carinii pneumonia (PCP) in severely immunocompromised individuals.^{1,2} It is suggested that most children experience mild or subclinical infection with Pneumocystis carinii and that the organism remains in a latent state until immunosuppression triggers reactivation.¹⁻⁵ Prior to the AIDS epidemic, those primarily at risk have been premature infants, patients with immunodeficiency disorders and patients receiving immunosuppressive treatment. ^{1, 2, 4-11} Since 1981, the prevalence of PCP has significantly increased due to the susceptibility of individuals with AIDS. PCP develops in 60-85% of AIDS patients and is the most common opportunistic infection of the group.^{12, 13} Rapid clinical diagnosis of Pneumocystis carinii is very important since the organism quickly infiltrates lung tissue causing dyspnea, fever, and cough²

SPECIMEN TYPES

Broncho alveolar lavage (BAL), Bronchial wash, Bronchial brush, Lung Imprint

There are three life forms of Pneumocystis carinii: the cyst, the trophozoite, and the sproozoite^{-1,9} The Fungi-Fluor[®] Kit Pneumocystis Kit has been designed to detect cysts, which have these characteristics:

- 5-8µm in diameter, thick-walled
- Most easily recognized form of Pneumocystis carinii
- Cluster in masses within an extra cellular matrix
- Intense staining of thickenings in cell walls gives "double parenthesis" appearance

SPECIMEN PREPARATION: The specimen or smear should be fixed in absolute methanol for 1 to 5 minutes. This may vary with the thickness of the smear. The fixation step will help adhere the specimen to the slide and prevent loss of material during staining. Frozen sections or specimens should be fixed in absolute methanol for 5 to 10 minutes depending on section thickness or laboratory protocol. Slides can be stained immediately after rinsing gently with distilled or deionized water.

STAINING PROCEDURE CONTROL: Use the positive control slide provided with the kit to assure staining results with the patient specimen. Control slides may be dipped in deionized water prior to staining to aid in penetration of the solution.

1. Place slide in a horizontal position and apply a few drops of Fungi Fluor[®] Pneumocystis stain directly on the specimen. The entire specimen should be covered or flooded. Stain for 1 minute.

2. Drain the solution from the slide and rinse very gently with running distilled or deionized water. Slides can be gently dipped in two changes of distilled or deionized water.

3. Slides may be air dried or viewed directly as a wet mount. Slides may also dehydrate through 2 changes of 95% ethanol, absolute ethanol, and then xylene followed by coverslipping with PolyMount (Cat. # 08381) Coverslipping Media for a permanent slide



TECHNICAL DATA SHEET 482

MICROSCOPE FILTER REQUIREMENTS: Fluorescence microscope filter descriptions can vary with the type of microscope used. We suggest you discuss filter options with your specific microscope manufacturer to assure proper excitation levels for viewing the specimen. The microscope manufacturer should have a list of filters with common descriptions to match your needs.

POSITIVE STAINING: Specimens containing two typical cysts with either the apple-green or blue fluorescence can be considered positive as for P. carinii. P. carinii cysts appear round, uniform in size and non-budding with a diameter of 5-8µm. The highly characteristic staining pattern of the peripheral cyst wall with intense internal staining of the "double parenthesis" structure should be noted. Free "double parenthesis" forms can be seen and are presumably degenerating cysts. Cysts are found in clusters.

 Methenamine silver nitrate Toluidine Blue O 	 Stains cyst wall only Time consuming procedure Requires high level of expertise for interpretation May require noxious chemical use Appearance: purple-brown to black, lavender to blue-violet
• Wright Giemsa Gram Staining	 Stain trophozoites and sporozoites only High level of expertise required Extensive time necessary for interpretation Appearance: deep blue to purple
• Direct/Indirect Fluorescent Immunoassay	 Long staining times - up to 1 hour or more High background with staining of extracellular matrix Incubation steps required Appearance: bright apple-green fluorescence
• Fungi-Fluor® Pneumocystis Kit	 Minimal prep 3 minutes for stain and analysis Easy to use Distinct fluorescent staining pattern Low cost per slide Appearance: bright apple-green fluorescence or blue depending on the filter used

COMPARATIVE TECHNIQUES FOR DETECTION OF PNEUMOCYSTIS CARINII TECHNIQUE

TECHNICAL DATA SHEET 482

PRECAUTIONS AND STORAGE:

Fungi-Fluor[®] Solution is an irritant. Exercise normal care in handling. Fungi-Fluor[®] Solution should be stored protected from light at room temperature. Do not freeze.

REFERENCES

- 1. Thorax, 40:461-570(1985).
- 2. Ann. Intern. Med., 80:83-93 (1974).
- 3. Pediatrics, 66:56-62 (1980).
- 4. Rev. Infect. Dis., 8(6):1001-1011(1986)
- 5. Ann. Intern. Med., 100:663-671 (1984).
- 6. J. Pediat., 82:404-415 (1973).
- 7. J. Clin. Microbiol., 20:887-890 (1984).
- 8. J. Infect. Dis., 140:143-147 (1987).
- 9. J. Respir. Dis., June, 83-94 (1987).
- 10. Blood, 39:759-770 (1972).
- 11. Pediatrics, 19:543-565 (1957)

America, Asia, Oceania info@polysciences.com www.polysciences.com (P) 1 (800) 523-2575 (F) 1 (800) 343-3291

Europe

info@polysciences.de www.polysciences.de (P) +(49) 6201 845 20 0 (F) +(49) 6201 845 20 20

Taiwan

info@polysciences.tw www.polysciences.tw (P) (886) 2 8712 0600 (F) (866) 2 8712 2677

