ACRO BIOTECH, INC

S. pneumoniae and L. pneumophila Combo Rapid Test Cassette (Urine) Package Insert

REF ISLC-125	English

A rapid test for the qualitative detection of S. pneumoniae antigen and Legionella pneumophila antigen in human urine specimen.

For professional in vitro diagnostic use only.

INTENDED USE

The S. pneumoniae and L. pneumophila Combo Rapid Test Cassette (Urine) is a rapid chromatographic immunoassay for the qualitative detection of *Streptococcus pneumoniae* antigen and *Legionella pneumophila* antigen in human urine specimen.

SUMMARY

Streptococcus pneumoniae, or pneumococcus, is a Gram-positive, alpha-hemolytic (under aerobic conditions) or beta-hemolytic (under anaerobic conditions), facultative anaerobic member of the genus Streptococcus,[1] As a significant human pathogenic bacterium *S. pneumoniae* was recognized as a major cause of pneumonia in the late 19th century, and is the subject of many humoral immunity studies. *S. pneumoniae* resides asymptomatically in healthy carriers typically colonizing the respiratory tract, sinuses, and nasal cavity. However, in susceptible individuals with weaker immune systems, such as the elderly and young children, the bacterium may become pathogenic and spread to other locations to cause disease. It spreads by direct person-to-person contact via respiratory droplets and by autoinoculation in persons carrying the bacteria in their upper respiratory tract.[2] It can be a cause of neonatal infections.[3] *S. pneumoniae* is the main cause of community acquired pneumonia and meningitis in children and the elderly,[4] and of septicemia in those infected with HIV. The organism also causes many types of pneumococcal infections other than pneumonia. These invasive pneumococcal diseases include bronchitis, rhinitis, otitis media, conjunctivitis, meningitis, sepsis, osteomyelitis, septic arthritis, endocarditis, peritonitis, pericarditis, cellulitis, and brain abscess.[5]

Legionellosis is a serious pneumonia caused by bacteria of the genus Legionella assigned to the family Legionellaceae. This family now includes 48 species and over 60 serogroups. Approximately 20 species are implicated in human disease. The overwhelming majority of *Legionella* infections are caused by *Legionella pneumophila*. Legionnaires' disease is the major clinical manifestation of *Legionella* infection although extra-pulmonary infection and non-pneumonic disease like Pontiac fever occur. *Legionella pneumophila* is responsible for approximately 90% of infections, and of these, over 80% are due to a single serogroup, serogroup 1.[6]

Legionnaires' disease (LD) is not contagious. The disease is transmitted by aerosol, and there is no evidence for direct person-to-person transmission. Person at risk are those whose immune system is compromised, including transplant recipients, the elderly, cigarette smokers, or those showing chronic obstructive pulmonary disease or chronic renal disease.[7]

PRINCIPLE

The *S. pneumoniae* Rapid Test Cassette (Urine) is a qualitative, membrane based immunoassay for the detection of *Streptococcus pneumoniae* in urine specimen. During testing, *Sreptococcus pneumoiae (S. pneumoniae)* antigens, if present in the specimen react with *S.pneumoniae* antibody-conjugate in the reagent area. The conjugate-antigens complex thus formed will bind with Anti-*S.pneumoniae* antibodies coated on the membrane in case of a positive result. This would result in a dark red colored line in T line region in case of a positive result. In case of negative result, no conjugates would bind at Anti-*S. pneumoniae* coated in T line region and no line would form in T line region of the test membrane. The intensity of the lines will vary depending upon the amount of antigen present in the sample. A line in Control region should appear in all correctly performed cases. Absence of C line indicates an invalid test result.

The *L. pneumophila* Rapid Test Cassette (Urine) is a qualitative, membrane based immunoassay for the detection of *Legionella pneumophila antigen* in urine specimen. During testing, *Legionella pneumophila* antigen, if present in the specimen react with *Legionella pneumophila* antibody-conjugate in the reagent area. The conjugate-antigens complex thus formed will bind with Anti-*Legionella pneumophila* antibodies coated on the membrane in case of a positive result. This would result in a dark red colored line in T line region in case of a positive result. In case of negative result, no conjugates would bind at Anti- *Legionella pneumophila* coated in T line region and no line would form in T line region of the test membrane. The intensity of the lines will vary depending upon the amount of antigen present in the sample. A line in Control region should appear in all correctly performed cases. Absence of C line indicates an invalid test result.

REAGENTS

The *S. pneumoniae and L. pneumophila* Combo Rapid Test Cassette (Urine) contains anti-*S.pneumoniae* antibody conjugated gold particles, anti-*S.pneumoniae* antibody coated on the membrane, anti-*Legionella* particles and anti-*Legionella* coated on the membrane

PRECAUTIONS

1. For professional *in vitro* diagnostic use only. Do not use after the expiration date.
2. Do not eat, drink or smoke in the area where the specimens or kits are handled.
3. Handle all specimens as if they contain infectious agents. Observe established precautions against microbiological hazards throughout all procedures and follow the standard procedures for proper disposal of specimens.
4. Wear protective clothing such as laboratory coats, disposable gloves and eye protection when specimens are assayed.
5. The used test should be discarded according to local regulations.
6. Humidity and temperature can adversely affect results.
7. Caution must be taken at the time of specimen collection. Inadequate volume of specimen may lead to lower sensitivity.

STORAGE AND STABILITY

The kit can be stored at room temperature or refrigerated (2-30℃). The test cassette is stable through the expiration date printed on the sealed pouch. The test cassette must remain in the sealed pouch until use. **DO NOT FREEZE**. Do not use beyond the expiration date.

SPECIMEN COLLECTION AND PREPARATION

The *S. pneumoniae and L. pneumophila* Combo Rapid Test Cassette (Urine) can be performed using urine. Urine specimens should be collected in standard containers. The sample can be stored at room temperature (15-30℃) if assayed within 24 hours of collection. Alternatively, specimens may be stored at 2-8℃ for up to 14 days or at -10℃ to -20℃ for longer periods before testing. When necessary, urine specimens should be shipped in leak-proof containers at 2-8℃ or frozen. Allow all specimens to equilibrate to room temperature before testing.

MATERIALS

Materials Provided

- Test Cassettes
- Droppers
- Package Insert

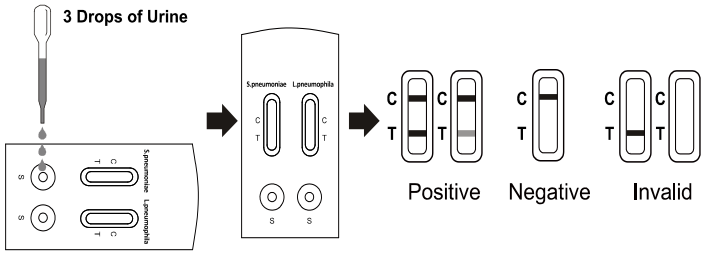
Materials Required But Not Provided

- Specimen collection containers
- Timer

DIRECTIONS FOR USE

Allow the test and specimen to reach room temperature (15-30℃) prior to testing.

1. Remove the test cassette from the foil pouch and use it within one hour. Best results will be obtained if the test is performed immediately after opening the foil pouch.
2. Place the cassette on a clean and level surface.
3. Absorb the urine specimen with a dropper, add **3 full drops** (approx.120 μL) specimen into the sample well (S) of test cassette vertically.
4. Wait for the colored line(s) to appear. **Read results at 15 minutes**. Do not interpret the result after 20 minutes.



INTERPRETATION OF RESULTS

(Please refer to the illustration above)

POSITIVE: * Two colored lines appear. One colored line should be in the control line region (C) and the other colored line should be in the test line region (T). A positive result indicates that *S.pneumoniae* antigen and/or *Legionella pneumophila* antigen are present in the specimen.

***NOTE:** The intensity of the color in the test line region (T) will vary depending on the concentration of *S.pneumoniae* antigen and/or *Legionella pneumophila* antigen present in the specimen. Therefore, any shade of color in the test line region (T) should be considered positive.

NEGATIVE: One colored line appears in the control line region (C). No line appears in the test line region (T). A negative result indicates that *S.pneumoniae* antigen or *Legionella pneumophila* antigen is not present in the specimen, or is present below the detectable level of the test.

INVALID: Control line fails to appear. Insufficient specimen volume or incorrect procedural techniques are the most likely reasons for control line failure. Review the procedure and repeat the test with a new test. If the problem persists, discontinue using the test kit immediately and contact your local distributor.

QUALITY CONTROL

Internal procedural controls are included in the test. A colored line appearing in the control region (C) is an internal procedural control. It confirms sufficient specimen volume and correct procedural technique.

Control standards are not supplied with this kit; however, it is recommended that positive and negative controls be tested as a good laboratory practice to confirm the test procedure and to verify proper test performance.

LIMITATIONS

1. *The S. pneumoniae* and *L. pneumophila* Combo Rapid Test is for *in vitro diagnostic* use only. The test should be used for the detection of *S.pneumoniae* antigen and/or *L. pneumophila* antigens in urine specimens only. Neither the quantitative value nor the rate of increase in *S.pneumoniae* antigen and/or *L. pneumophila* antigen concentration can be determined by this qualitative test.
2. A negative result should be confirmed by culture. A negative result may be obtained, if the concentration of the *S.pneumoniae* antigen or *L. pneumophila* antigen present in the urine is not adequate or is below the detectable level of the test.
3. As with all diagnostic tests, all results must be interpreted together with other clinical information available to the physician.

PERFORMANCE CHARACTERISTICS

Clinical Sensitivity, Specificity and Accuracy

***S. pneumoniae* Rapid Test**

The performance of the *S. pneumoniae* Rapid Test Cassette (Urine) has been evaluated with 103 clinical specimens collected from the patient symptomatic and asymptomatic in comparison with other rapid test method. The results show that the relative sensitivity of the *S. pneumoniae* Rapid Test Cassette (Urine) is 90.0% and the relative specificity is 98.9%.

***S. pneumoniae* Rapid Test Cassette vs. Other Rapid Test**

Method		Other Rapid Test		Total Results
***S. pneumoniae* Rapid Test Cassette (Urine)**	**Results**	Positive	Negative	
	Positive	9	1	10
	Negative	1	92	93
Total Results		10	93	103

Relative Sensitivity: 90.0% (95%CI*: 55.5%~99.7%);
Relative Specificity: 98.9% (95%CI*: 94.2%~>99.9%);
Overall Accuracy: 98.1% (95%CI*: 93.2%~99.8%). *Confidence Intervals

***Legionella pneumophila* Rapid Test**

The performance of the *L. pneumophi*la Rapid Test Cassette (Urine) has been evaluated with 105 clinical specimens collected from the patient symptomatic and asymptomatic in comparison with other rapid test method. The results show that the relative sensitivity of the *L.pneumophila* Rapid Test Cassette (Urine) is 97.0% and the relative specificity is 98.6%.

***L. pneumophila* Rapid Test Cassette vs. Other Rapid Test**

Method		Other Rapid Test		Total Results
***L. pneumophila* Rapid Test Cassette (Urine)**	**Results**	Positive	Negative	
	Positive	32	1	33
	Negative	1	71	72
Total Results		33	72	105

Relative Sensitivity: 97.0% (95%CI*: 84.2%~99.9%);
Relative Specificity: 98.6% (95%CI*: 92.5%~>99.9%);
Overall Accuracy: 98.1% (95%CI*: 93.3%~99.8%). *Confidence Intervals

Precision

Intra-Assay

Within-run precision has been determined by using 3 replicates of same positive and negative urine specimens with the same production batch in the same experimental conditions. The specimens were correctly identified >99% of the time.

Inter-Assay

Between-run precision has been determined by 3 independent assays on the same positive and negative urine specimens. Three different lots of the *S. pneumoniae* and *L. pneumophila* Combo Rapid Test Cassette (Urine) have been tested using these specimens. The specimens were correctly identified >99% of the time.

Cross-reactivity

Cross reactivity with following organisms has been studied at 1.0E+07 organisms/mL. The following organisms were found negative when tested with the *S.pneumoniae* Rapid Test Cassette (Urine):

Legionella pneumophila	Candida albicans
Helicobacter pylori	Clostridium difficile
Chlamydia	Neisseria gonococcus

Cross-reactivity to urines spiked with the following pathogens were found negative when tested with the *L. pneumophila* Rapid Test Cassette (Urine).

Adenovirus	Clostridium difficile	HMPV
Aspergillus niger	E.coli (different strains)	Streptococcus mutans
Candida albicans	Enterobacter cloacae	Vibrio parahemolyticus
Haemophilus influenzae	Enterococcus faecalis	Ureaplasma urealyticum
Influenza A	Escherichia hermanni	Mycobacterium avium
Influenza B	Helicobacter pylori	Mycobacterium intracellulare

Moraxella catarrhalis	Klebsiella pneumoniae	Mycobacterium tuberculosis
Mycoplasma pneumonia	Legionella bozemanii (sg1)	Serratia marcescens
Nocardia asteroides	Legionella longbeachae	Pseudomonas aeruginosa
Parainfluenzae	Neisseria meningitidis	Shigella sonnei
Rhinovirus	Proteus mirabilis	Campylobacter coli
RSV	Salmonella enteritidis	S. typhimurium
Staphylococcus aureus	Shigella flexneri	Vibrio parahemolyticus
Streptococcus pneumonia	Staphylococcus epidermidis	Neisseria meningitidis(sg C)
Streptococcus pyogenes	Yersinia enterocolitica(types 3,9)	Mycoplasma hominis
Campylobacter jejuni	Streptococcus (Group B,C, F, G)	

The blood naturally present in urine (microhematuria conditions) doesn't affect test performances. However, bloody specimens (at 0.1% whole blood) may fail to flow properly causing smears and inconclusive test results.

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Index of Symbols

	Consult instructions for use	Σ	Tests per kit	EC REP	Authorized representative
IVD	For *in vitro* diagnostic use only		Use by		Do not reuse
2°C 30°C	Store between 2-30°C	LOT	Lot number	REF	Catalog #
	Do not use if package is damaged		Manufacturer		

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