

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

PNEU	English
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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 asymptotically 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, *Streptococcus pneumoniae* (*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°C). 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°C) if assayed within 24 hours of collection. Alternatively, specimens may be stored at 2-8°C for up to 14 days or at -10°C to -20°C for longer periods before testing. When necessary, urine specimens should be shipped in leak-proof containers at 2-8°C or frozen. Allow all specimens to equilibrate to room temperature before testing.

[MATERIALS]

- Materials Provided

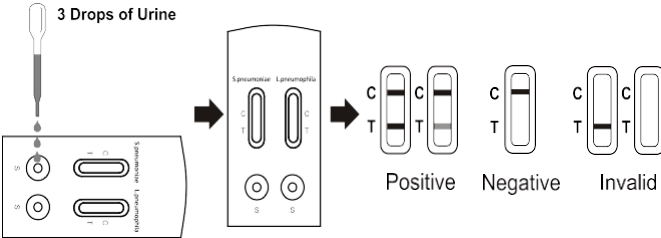
- Test Cassettes
 - Droppers
 - Package Insert
- Materials Required But Not Provided

- Specimen collection containers
 - Timer

[DIRECTIONS FOR USE]

Allow the test, specimen and/or controls to reach room temperature (15-30°C) 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.120ul) 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 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
<i>S. pneumoniae</i> 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. pneumophila* 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
<i>L. pneumophila</i> 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):

<i>Legionella pneumophila</i>	<i>Candida albicans</i>
<i>Helicobacter pylori</i>	<i>Clostridium difficile</i>
<i>Chlamydia</i>	<i>Neisseria gonococcus</i>
Cross-reactivity to urines spiked with the following pathogens were found negative when tested with the <i>L. pneumophila</i> Rapid Test Cassette (Urine).	
<i>Adenovirus</i>	<i>Clostridium difficile</i>
<i>Aspergillus niger</i>	<i>E. coli</i> (different strains)
<i>Candida albicans</i>	<i>Enterobacter cloacae</i>
<i>Haemophilus influenzae</i>	<i>Enterococcus faecalis</i>
<i>Influenza A</i>	<i>Escherichia hermanni</i>
<i>Influenza B</i>	<i>Helicobacter pylori</i>
<i>Moraxella catarrhalis</i>	<i>Klebsiella pneumoniae</i>
<i>Mycoplasma pneumonia</i>	<i>Legionella bozemanii</i> (sg1)
<i>Nocardia asteroides</i>	<i>Legionella longbeachae</i>
<i>Parainfluenzae</i>	<i>Neisseria meningitidis</i>
<i>Rhinovirus</i>	<i>Proteus mirabilis</i>
<i>RSV</i>	<i>Salmonella enteritidis</i>
<i>Staphylococcus aureus</i>	<i>Shigella flexneri</i>
<i>Streptococcus pneumonia</i>	<i>Staphylococcus epidermidis</i>
<i>Streptococcus pyogenes</i>	<i>Yersinia enterocolitica</i> (types 3,9)
<i>Campylobacter jejuni</i>	<i>Streptococcus</i> (Group B,C, F, G)
	<i>Pseudomonas aeruginosa</i>
	<i>Shigella sonnei</i>
	<i>Campylobacter coli</i>
	<i>S. typhimurium</i>
	<i>Vibrio parahemolyticus</i>
	<i>Neisseria meningitidis</i> (sg C)
	<i>Mycoplasma hominis</i>

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.

[BIBLIOGRAPHY]

1. Ryan KJ, Ray CG, eds. (2004). Sherris Medical Microbiology. McGraw Hill. ISBN 0-8385-8529-9.

2. "Transmission". cdc.org. Retrieved 24 Oct 2017.

3. Baucells, B.J.; MercadalHally, M.; Álvarez Sánchez, A.T.; FiguerasAloy, J. (2015). "Asociaciones de probióticos para la prevención de la enterocolitis necrosante y la reducción de la sepsis tardía y la mortalidad neonatal en recién nacidos pretérmino de menos de 1.500g: una revisión sistemática". Anales de Pediatría. doi:10.1016/j.anpedi.2015.07.038. ISSN 1695-4033.

4. van de Beek, Diederik; de Gans, Jan; Tunkel, Allan R.; Wijdicks, Eelco F.M. (5 January 2006). "Community-Acquired Bacterial Meningitis in Adults". New England Journal of Medicine. 354 (1): 44–53. doi:10.1056/NEJMra052116. ISSN 0028-4793.PMID 16394301.Retrieved 15 February 2017.

5. Siemieniuk, Reed A.C.; Gregson, Dan B.; Gill, M. John (Nov 2011). "The persisting burden of invasive pneumococcal disease in HIV patients: an observational cohort study". BMC Infectious Diseases. 11:314.doi:10.1186/1471-2334-11:314.

6. J.H. Helbig et al.; Pan-European study on culture-proven Legionnaires' Disease; Eur. J. Clin. Microbiol. Infect. Dis. 2002 21:710-716, 2002

7. B.S. Fields et al.; Legionella and Legionnaires'Disease : 25 years of investigation; Clin. Microbiol. Rev. 2002 15: 506-526, 200

Index of Symbols

	Consult instructions for use or consult electronic instructions for use		Contains sufficient for <n> tests		Temperature limit
IVD	<i>In vitro</i> diagnostic medical device	LOT	Batch code	REF	Catalogue number
	Authorized representative in the European Community/European Union		Use-by date		Do not re-use
	Do not use if package is damaged and consult instructions for use		Manufacturer		Caution

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