

STANDARD F Legionella Ag FIA

STANDARD F Legionella Ag FIA

PLEASE READ INSTRUCTIONS CAREFULLY BEFORE YOU PERFORM THE TEST



EXPLANATION AND SUMMARY

[Introduction]

The genus *Legionella* is a pathogenic group of gram-negative bacteria. The genus *Legionella* contains more than 50 species, of which at least 24 have been associated with human infection. The best characterized member of the genus, *Legionella pneumophila*, is the major causative agent of Legionellosis. *Legionella pneumophila* is commonly found in lakes, rivers, creeks, hot springs and other bodies of water where it is symbiotically present in aquatic amoebae. Most people become infected when they inhale microscopic water droplets containing the bacteria. Legionellosis is a generic term describing the pneumonic and non-pneumonic forms of infection with *Legionella*. The non-pneumonic form, Pontiac disease fever, is an acute, self-limiting influenza-like illness usually lasting 2-5 days. No deaths are associated with this type of infection. The pneumonic form, Legionnaires' disease, has an incubation period of 2-10 days. The severity of disease ranges from a mild cough to a rapidly fatal pneumonia. Death occurs through progressive pneumonia with respiratory failure or shock and multi-organ failure. Early diagnosis of Legionnaires' disease is important, as the rate of mortality from untreated Legionnaires' disease ranges from 10 to 50%. STANDARD F Legionella Ag FIA, containing a highly specific and sensitive antibody, provides significantly fast, easy and accurate system to identify the *Legionella* Ag from human urine specimen.

[Intended use]

STANDARD F Legionella Ag FIA is a fluorescence immunoassay for the qualitative detection of *Legionella pneumophila* serogroup 1 antigen present in urine samples from patients with symptoms of pneumonia. This test is for *in vitro* diagnostic use and intended for professional use, only for an initial screening test. STANDARD F Legionella Ag FIA should be used with the appropriate analyzer, STANDARD F Analyzers, manufactured by SD BIOSENSOR.

[Test principle]

STANDARD F Legionella Ag FIA is based on immunofluorescence technology with STANDARD F Analyzer to detect *Legionella pneumophila* serogroup 1 antigen. STANDARD F Legionella Ag FIA has control line and test line which is coated with monoclonal anti-PPR and rabbit anti-*Legionella* each. The patient's sample is applied into the sample well of the test device and the sample migrates through the membrane. If *Legionella* antigen is present, it will react with europium conjugated monoclonal anti-*Legionella* on the conjugation pad and form antibody-antigen fluorescence particle complexes. These complexes move along to the membrane to be captured by the anti-*Legionella* on the test line and make fluorescence signal. The intensity of the fluorescence light generated on the membrane is scanned by the STANDARD F Analyzer manufactured by SD BIOSENSOR. STANDARD F Analyzer can analyze the presence of the analyte in the clinical specimen by processing the results using programmed algorithms and display the test result on the screen.

[Kit contents]

- 1 Test device
- 2 Fixed volume pipette (100µl)
- 3 Positive control
- 4 Negative control
- 5 Control extraction buffer
- 6 Instructions for use

[Materials required but not provided]

- STANDARD F Analyzer
- Timer

KIT STORAGE AND STABILITY

Store the kit at 2-30°C / 36-86°F, out of direct sunlight. Kit materials are stable until expiration date printed on the outer box. Do not freeze the kit.

WARNINGS AND PRECAUTIONS

1. Do not re-use the test kit.
2. Use the STANDARD F Legionella Ag FIA at 15-32°C / 59-90°F and 10-90%RH.
3. Do not use the test kit if the pouch is damaged or the seal is broken.
4. Do not smoke, drink or eat while handling specimen.
5. Wear personal protective equipment, such as gloves and lab coats when handling kit reagents. Wash hands thoroughly when afterwards.
6. Clean up spills thoroughly using an appropriate disinfectant.
7. Handle all specimens as if they contain infectious agents.
8. Observe established precautions against microbiological hazards throughout testing procedures.
9. Dispose of all specimens and materials used to perform the test as bio-hazard waste. Laboratory chemical and bio-hazard waste must be handled and discarded in accordance with all local, state, and national regulations.
10. Silica gel in foil pouch is to absorb moisture and keep humidity from affecting products. If the moisture indicating silica gel beads change from yellow to green, the test device in the pouch should be discarded.
11. Immediately use the test device after taking out of a foil pouch.
12. If the test result with positive/negative control is abnormal, do not use the kit.
13. The bar code of the test device is used by analyzer to identify the type of test being run and to identify the individual test device so as to prevent to a second read of the test device by the same analyzer.
14. Once a test device has been successfully scanned by analyzer, do not attempt to scan the test device again in the same analyzer.
15. As the detection reagent is a fluorescent compound, no visible results will form on the test device.
16. Improper specimen collection, handling or transport may yield inaccurate results.
17. Do not write on the bar code or damage the bar code of the test device.
18. Do not use unspecified specimens in this instructions for use.

SPECIMEN COLLECTION AND PREPARATION

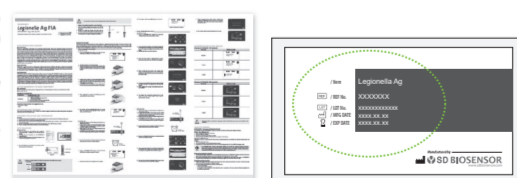
[Urine]

1. Urine specimens should be collected in a clean and dry container and collected at any time of the day may be used.
2. Urine specimen may be stored at room temperature (15-30°C/59-86°F) for up to 3 days or at 2-8°C/36-46°F for up to 2 weeks prior to testing.
3. For prolonged storage, specimens may be frozen and stored at -40°C/ -40°F. The frozen specimens are stable up to 3 months at -40°C/ -40°F.
4. Frozen urine specimens should be thawed and fully mixed before testing.
5. If the urine specimen exhibits visible precipitates, it should be centrifuged, filtered, or allowed to settle to obtain a clear supernatant for testing.

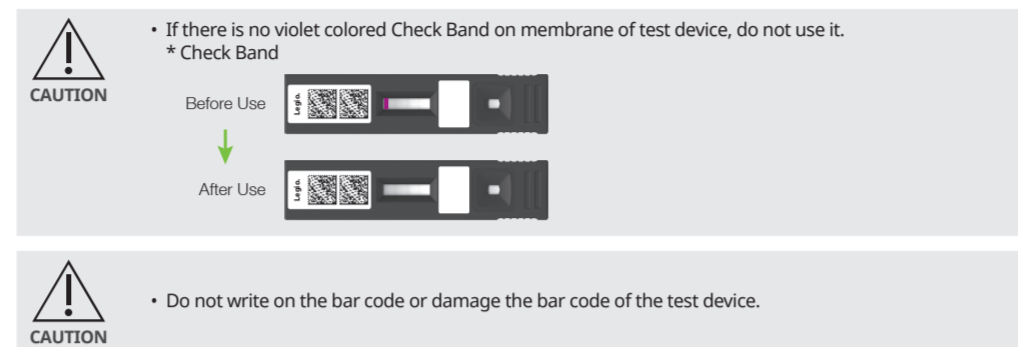
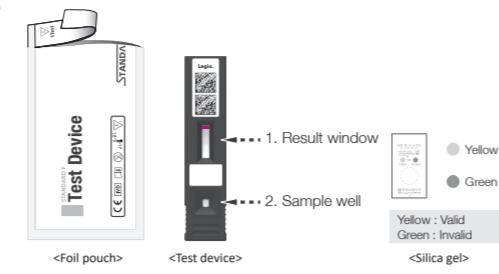
TEST PROCEDURE

[Preparation]

1. Allow kit components and collected urine sample to room temperature (15-30°C/59-86°F) a minimum of 30 minutes prior to testing.
2. Carefully read instructions for the STANDARD F Legionella Ag FIA.
3. Check the expiry date at the back of the foil pouch. Use another lot, if expiry date has passed.



4. Open the foil pouch, and check the test device and the silica gel pack in the foil pouch.



• Preparation of the positive/negative control sample

1. Insert the positive/negative control swab in the kit into the control extraction buffer tube. Swirl the swab at least five times.
2. Remove the swab and dispose the used swab in accordance with your biohazard waste disposal protocol.
3. To analyze of the external quality control sample, follow instructions for 'Analysis of sample' stated below.

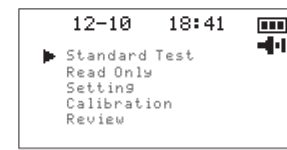


[Analysis of sample]

• Using a STANDARD F100 Analyzer

- 'Standard Test' mode

1. Prepare a STANDARD F100 Analyzer and set the 'Standard Test' mode according to the analyzer's manual.
2. Take the test device out of the foil pouch.
3. Insert the test device to the Test Slot of the analyzer. The analyzer automatically reads the information of the bar code on the test device and releases the test device for adding sample.



4. Compress the top bulb of a fixed volume pipette (100µl) and place the tip of the pipette into the prepared sample.
5. Slowly release the top bulb of the pipette dipping the tip of the pipette into the sample.
6. Completely squeeze the sample into the sample well of the test device pressing hard the top bulb of the pipette.

7. After applying the sample, immediately press the center button to start the test.

8. The analyzer will automatically display the test result within 15 minutes. Strong positive results may be observed after 5 minutes.

- 'Read Only' mode

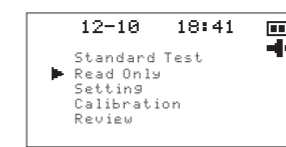
1. Take the test device out of the foil pouch and place it on a flat and dry surface.
2. Compress the top bulb of a fixed volume pipette (100µl) and place the tip of the pipette into the prepared sample.
3. Slowly release the top bulb of the pipette dipping the tip of the pipette into the sample.
4. Completely squeeze the sample into the sample well of the test device pressing hard the top bulb of the pipette.



5. Leave the test device for 15 minutes. Notice that the test device should not leave for 30 more minutes.



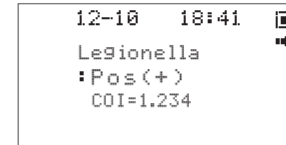
6. Prepare a STANDARD F100 Analyzer and set the 'Read Only' mode according to analyzer's manual.



7. Insert the test device to the Test Slot of the analyzer.



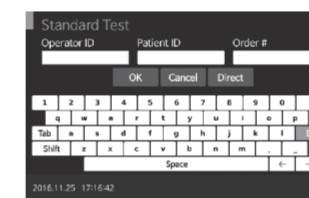
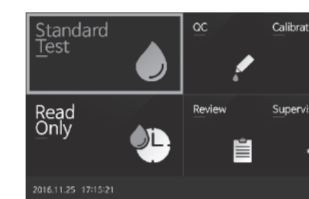
8. The analyzer will automatically display the test result.



• Using a STANDARD F200 Analyzer

- 'Standard Test' mode

1. Prepare a STANDARD F200 Analyzer and select the 'Standard Test' on the analyzer's screen.
2. Input operator ID, patient ID, and order #. If patient ID is not input into the analyzer by touching the 'Direct' item, the analyzer will regard the test as that of the guest.
3. Take the test device out of the foil pouch.

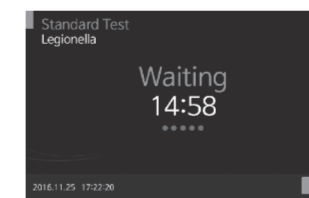


4. Once the 'Insert Device' is displayed in the screen, insert the test device into the Test Slot of the analyzer.

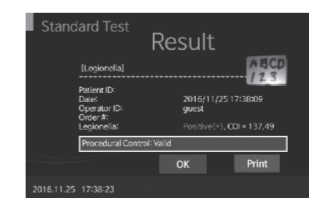


6. Apply 100µl of prepared sample to the sample well in the test device with a fixed volume pipette (100µl).
7. After applying the sample, immediately press the start button.

8. The analyzer allows the test device to develop for 15 minutes.

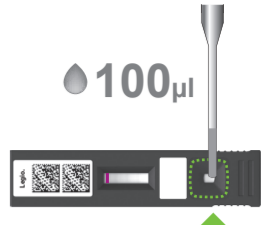


9. The analyzer will automatically display the test result on the screen. Strong positive results may be observed after 5 minutes.



- 'Read only' mode

1. Take the test device out of the foil pouch and place it on a flat and dry surface.
2. Compress the top bulb of a fixed volume pipette (100µl) and place the tip of the pipette into the prepared sample.
3. Slowly release the top bulb of the pipette dipping the tip of the pipette into the sample.
4. Completely squeeze the sample into the sample well of the test device pressing hard the top bulb of the pipette.



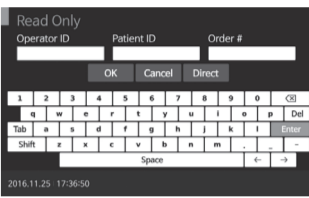
5. Leave the test device for 15 minutes. Notice that the test device should not leave for 30 more minutes.



6. Prepare a STANDARD F200 Analyzer and select the 'Read Only' on the analyzer's screen.



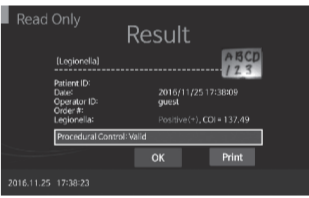
7. Input operator ID, patient ID, and order #. If patient ID is not input into the analyzer by touching the 'Direct' item, the analyzer will regard the test as that of the guest.



8. Once the 'Insert Device' is displayed in the screen, insert the test device to the Test Slot of the analyzer.



10. The analyzer will automatically display the test result.



• The mark on the label between sample well and result window is scanned by the STANDARD F200 Analyzer and displayed on the screen.

INTERPRETATION OF TEST RESULTS

[Displays of STANDARD F100 Analyzer]

Test result	Window example
Negative	
Positive	
Invalid	

[Displays of STANDARD F200 Analyzer]

Test result	Window example	
Negative		
Positive		
Invalid		

CAUTION The test result of a sample is given either as Positive(+)/Pos(+) or Negative(-)/Neg(-) with a COI (cutoff index) value. COI is calculated that a measured signal is divided by an appropriate cutoff value.

- Test results of a COI ≥ 1.00 are considered positive for *Legionella* Antigen.
- Test results of a COI < 1.00 are considered negative for *Legionella* Antigen.

QUALITY CONTROL

[Calibration Set Test]

The calibration set test of STANDARD F Analyzers should be conducted according to the analyzer's manual.

When to use calibration set

1. Before using the analyzer for the first time
2. When you drop the analyzer
3. Whenever you do not agree with your result
4. When you want to check the performance of an analyzer and test device

How to use calibration set

Calibration set test is a required function that ensures optimal performance by checking the internal analyzer optics and functions.

1. Select the 'Calibration' menu.
2. The specific calibration set is included with the analyzer.
3. Insert the CAL-1 first, and then insert the CAL-2 for UV-LED testing and the CAL-3 for RGB-LED testing in order.

NOTE The STANDARD F Analyzer automatically calibrate and identify the optical performance through measuring the membrane of the test device whenever the test is conducted in 'Standard Test' mode. If 'EEE' message displays on the screen, it means that the analyzer has a problem, so check with CAL devices. Contact the SD BIOSENSOR local distributor if the 'EEE' message still appears.

[Internal procedural control]

1. The internal procedural control zone is on the membrane of the test device. STANDARD F Analyzers read the fluorescence signal of the internal procedural control zone and decide whether the result is valid or invalid.
2. The invalid result denotes that the fluorescence signal is not within the pre-set range. If the screen of STANDARD F Analyzer shows 'Invalid Device', turn off and turn of the analyzer again and re-test with a new test device.

[External quality control]

1. Positive and negative controls are supplied with each kit and these controls are provided as a means of additional quality control to demonstrate a positive or negative reaction.
2. It is recommended that positive and negative controls be run:
 - once for each new lot.
 - once for each untrained operator.
 - as required by test procedures in this instruction and in accordance with local, state and federal regulations or accreditation requirements.

LIMITATION OF THE TEST

1. The test procedure, precautions and interpretation of results for this test must be followed strictly when testing.
2. This test detects the presence of *Legionella pneumophila* serogroup 1 antigen in the specimen and should not be used as the sole criteria for the diagnosis of *Legionella* infection.
3. Test results must be considered with other clinical data available to the physician.
4. For more accuracy of immune status, additional follow-up testing using other laboratory methods is recommended.
5. Neither the quantitative value nor the rate of *Legionella pneumophila* serogroup 1 antigen concentration can be determined by this qualitative test.
6. Failure to follow the test procedure and interpretation of test results may adversely affect test performance and/or produce invalid results.
7. Rheumatoid Factor does not interfere at concentrations ≤14.84 IU/mL with the STANDARD F *Legionella* Ag FIA test.

PERFORMANCE CHARACTERISTICS

1. Analytical Reactivity: The strains of *Legionella pneumophila* serogroup 1, 3, 5, 6 and 8 were tested by STANDARD F *Legionella* FIA. All of the strains were detected at the concentrations listed in the below table.

No.	Strain	Limit of Detection (CFU/mL)*
1	<i>L. pneumophila</i> serogroup 1 Knoxville	4.0 x 10 ³
2	<i>L. pneumophila</i> Serogroup 1 Camperdown	8.5 x 10 ⁴
3	<i>L. pneumophila</i> Serogroup 1 Philadelphia	4.5 x 10
4	<i>L. pneumophila</i> Serogroup 3 ESC1	9.0 x 10 ⁴
5	<i>L. pneumophila</i> Serogroup 5 Dallas 1E	8.5 x 10 ⁴
6	<i>L. pneumophila</i> Serogroup 6 Chicago 2	9.5 x 10 ⁴
7	<i>L. pneumophila</i> Serogroup 8 Concord 3	8.0 x 10 ⁴

* Colony forming unit (CFU) is a unit used to estimate the number of viable bacteria in a sample.

2. Clinical sensitivity: Total 39 samples were evaluated for sensitivity. The STANDARD F *Legionella* Ag FIA Test got a high correlation with other FIA test.

Reference	STANDARD F <i>Legionella</i> Ag		Total Result
	Positive	Negative	
FIA	37	2	39
	0	0	0
Total Result	37	0	39
Sensitivity	37/39 (94.9%)		

3. Clinical specificity: Total 40 samples were evaluated for specificity. The STANDARD F *Legionella* Ag FIA Test got a high correlation with other FIA test.

Reference	STANDARD F <i>Legionella</i> Ag		Total Result
	Positive	Negative	
FIA	0	0	0
	0	40	40
Total Result	0	40	40
Specificity	40/40 (100%)		

BIBLIOGRAPHY

1. Berdal B P, Farshy C E, Feely J C. Detection of *Legionella pneumophila* antigen in urine by enzyme-linked immunospecific assay. J Clin Microbiol. 1979;9:575-578.
2. Kohler R B, Zimmerman S E, Wilson E, Allen S D, Edelstein P H, Wheat L J, White A. Rapid radioimmunoassay diagnosis of Legionnaires' disease: detection and partial characterization of urinary antigen. Ann Intern Med. 1981;94:601-605.
3. Tang P W, Toma S. Broad-spectrum enzyme-linked immunosorbent assay for detection of *Legionella* soluble antigens. J Clin Microbiol. 1986;24:556-558.
4. Tilton R C. Legionnaires' disease antigen detected by enzyme-linked immunosorbent assay. Ann Intern Med. 1979;90:697-698.
5. Bibb, W.F., P.M. Arnow, L. Thacker, and R.M. McKinney. Detection of soluble *Legionella pneumophila* antigens in serum and urine specimens by enzyme-linked immunosorbent assay with monoclonal and polyclonal antibodies. J. Clin. Microbiol. 1984;20:478-482.

Product Disclaimer

Whilst every precaution has been taken to ensure the diagnostic ability and accuracy of this product, the product is used outside of the control of the SD BIOSENSOR and distributor and the result may accordingly be affected by environmental factors and/or user error. A person who is the subject of the diagnosis should consult a doctor for further confirmation of the result.

Warning

The SD BIOSENSOR and distributors of this product shall not be liable for any losses, liability, claims, costs or damages whether direct or indirect of consequential arising out of or related to an incorrect diagnosis, whether positive or negative, in the use of this product.



Manufactured by **SD BIOSENSOR**
 Head office : C-4th&5th, 16, Deogyong-daero 1556beon-gil, Yeongtong-gu, Suwon-si, Gyeonggi-do, 16690, REPUBLIC OF KOREA
 Manufacturing site : 74, Osongsaengmyeong 4-ro, Osong-eup, Heungdeok-gu, Cheongju-si, Chungcheongbuk-do, 28161, REPUBLIC OF KOREA
 Authorized Representative
MT Promed Consulting GmbH
 Altenhofstrasse 80 D-66386 St. Ingbert Germany
 Phone : +49 6894 581020, Fax : +49 6894 581021

Any inquiries regarding instructions provided should be addressed to: sales@sdbiosensor.com or you can also contact us through www.sdbiosensor.com

L28LEG1ENR4
 Issued date: 2018.03