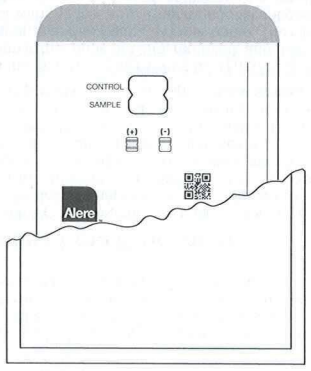
Alere™

Alere BinaxNOW®
Streptococcus pneumoniae Antigen Card

Materials Provided

1



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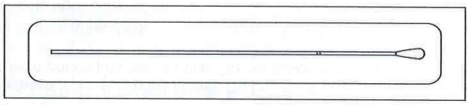

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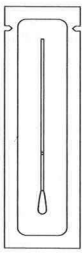


Intended Use

The Alere BinaxNOW® *Streptococcus pneumoniae* Antigen Card (Alere BinaxNOW® *Streptococcus pneumoniae)* is an *in vitro* rapid immunochromatographic (ICT) assay for the detection of *Streptococcus pneumoniae (S. pneumoniae)* antigen in the urine of patients with pneumonia and in the cerebral spinal fluid (CSF) of patients with meningitis. It is intended, in conjunction with culture and other methods, to aid in the diagnosis of both pneumococcal pneumonia and pneumococcal meningitis. The Alere BinaxNOW® *S. pneumoniae* Antigen Card can be read visually or used in conjunction with the Alere™ Reader.

Summary and Explanation of the Test

S. pneumoniae is the leading cause of community-acquired pneumonia and may be the most important agent in community-acquired pneumonia of unknown etiology.[1,2] Pneumococcal pneumonia has a mortality rate as high as 30%, depending on bacteremia, age, and underlying diseases.[1,3] When not properly diagnosed and treated, *S. pneumoniae* infection can lead to bacteremia, meningitis, pericarditis, empyema, purpura fulminans, endocarditis and/or arthritis.[4,5]

Pneumococcal meningitis, a condition that frequently leads to permanent brain damage or death, can occur as a complication of other pneumococcal infection or may arise spontaneously without any preceding illness.[6] It affects persons of all ages, but is most common in children under 5 years, teenagers and young adults, and in the elderly.[7] Progression from mild illness to coma can occur within hours, making immediate diagnosis and antimicrobial treatment critical. Twenty to thirty percent of all pneumococcal meningitis patients will die, often despite several days of appropriate antibiotic treatment.[6] Mortality is even higher among very young and very old patients.[6]

Alere BinaxNOW® *Streptococcus pneumoniae* provides a simple, rapid method for the diagnosis of pneumococcal pneumonia using a urine specimen that is conveniently collected, stored and transported. It also provides an immediate and highly accurate diagnosis of pneumococcal meningitis when CSF is tested.

Principles of the Procedure

Alere BinaxNOW® *Streptococcus pneumoniae* is an immunochromatographic membrane assay used to detect pneumococcal soluble antigen in human urine and CSF. Rabbit anti-*S. pneumoniae* antibody, the Sample Line, is adsorbed onto nitrocellulose membrane. Control antibody is adsorbed onto the same membrane as a second stripe. Both rabbit anti-*S. pneumoniae* and anti-species antibodies are conjugated to visualizing particles that are dried onto an inert fibrous support. The resulting conjugate pad and the striped membrane are combined to construct the test strip. This test strip and a well to hold the swab specimen are mounted on opposite sides of a hinged, book-shaped test card.

To perform the test (U.S. Patent Nos: 6,017,767; 6,548,309; 6,824,997), a swab is dipped into the specimen (either urine or CSF), removed, and then inserted into the test card. Reagent A, a buffer solution, is added from a dropper bottle. The card is then closed, bringing the specimen into contact with the test strip. Pneumococcal antigen present in the specimen reacts to bind anti-*S. pneumoniae* conjugated antibody. The resulting antigen-conjugate complexes are captured by immobilized anti-*S. pneumoniae* antibody, forming the Sample Line. Immobilized control antibody captures anti-species conjugate, forming the Control Line.

Test results are interpreted by the presence or absence of pink-to-purple colored lines. A positive test result, read in 15 minutes, will include the detection of both a Sample and a Control Line. A negative test result, read in 15 minutes, will produce only a Control Line, indicating that *S. pneumoniae* antigen was not detected in the specimen. Failure of the Control Line to appear, whether the Sample Line is present or not, indicates an invalid assay.

Reagents and Materials

Refer to illustrations on pull-out flap.

Materials Provided

❶ **Test Cards:** A membrane coated with rabbit antibody specific for *S. pneumoniae* antigen and with control antibody is combined with rabbit anti-*S. pneumoniae* antigen and anti-species conjugates in a hinged test card.

❷ **Reagent A:** Citrate / Phosphate buffer with sodium lauryl sulfate, Tween® 20, and sodium azide.

❸ **Specimen Swabs:** Designed for use with Alere BinaxNOW® *Streptococcus pneumoniae*. Do not use other swabs.

❹ **Positive Control Swab:** Inactivated *S. pneumoniae* antigen dried onto swab.

❺ **Negative Control Swab:** *S. pneumoniae* negative swab.

Materials Recommended But Not Provided

Clock, timer, or stopwatch; standard urine collection containers, or CSF transport tubes; Alere™ Reader.

Accessory Item

Alere BinaxNOW® *Streptococcus pneumoniae* Control Swab Pack (catalog number 710-010) containing 5 positive and 5 negative control swabs.

Precautions

Control swabs require six (6) drops of Reagent A. Patient specimens require three (3) drops of Reagent A.

1. **INVALID RESULTS**, indicated by no Control Line, can occur when an insufficient volume of Reagent A is added to the test card. To ensure delivery of an adequate volume, hold vial vertically, ½ - 1 inch above the swab well, and slowly add free falling drops.
2. For *in vitro* diagnostic use.
3. If the kit is stored in a refrigerator, allow all kit components to equilibrate to room temperature (15-30°C) before use.
4. The test card is sealed in a protective foil pouch. Do not use if pouch is damaged or open. Remove test card from pouch just prior to use. Do not touch the reaction area of the test card.
5. Do not use kit past its expiration date.
6. Do not mix components from different kit lots.
7. Swabs in the kit are approved for use with Alere BinaxNOW® *Streptococcus pneumoniae*. **Do not use other swabs.**
8. Solutions used to make the control swabs are inactivated using standard methods. However, patient specimens, controls, and test cards should be handled as though they could transmit disease. Observe established precautions against microbial hazards.
9. Clean catch urine is not necessary for Alere BinaxNOW® *Streptococcus pneumoniae*. Therefore, urine specimens used for this test may not be appropriate for bacteriological culture.
10. Once the Alere™ swab is dipped into CSF specimen, the specimen is no longer sterile and may not be appropriate for culture. If CSF specimen will be cultured, either perform culture first or split CSF specimen.
11. Refer to the Alere™ Reader User Manual, INLFR000, for operating instructions.

Storage and Stability

Store kit at 36-86°F (2-30°C). The Alere BinaxNOW® *Streptococcus pneumoniae* card and reagents are stable until the expiration dates marked on their outer packaging and containers. Do not use the kit beyond its labeled expiration date.

Quality Control

Daily Quality Control:

Alere BinaxNOW® *Streptococcus pneumoniae* contains built-in positive and negative procedural controls. The manufacturer's minimum recommendation for daily quality control is to document these procedural controls for the first specimen tested each day.

Positive Procedural Control

The pink-to-purple line at the "Control" position can be considered an internal positive procedural control. If capillary flow has occurred and the functional integrity of the card was maintained, this line will always appear.

Negative Procedural Control

The clearing of background color in the result window provides a negative background control. The background color in the window should be light pink to white within 15 minutes and should not interfere with the reading of the test result.

External Positive and Negative Controls:

Good laboratory practice suggests the use of positive and negative controls to ensure that:

- test reagents are working, and
- the test is correctly performed.

Alere BinaxNOW® *Streptococcus pneumoniae* kits contain Positive and Negative Control Swabs. These swabs will monitor the entire assay. Test these swabs with each new shipment received. Other controls may be tested in order to conform with:

- local, state and/or federal regulations,
- accrediting groups, and/or,
- your lab's standard Quality Control procedures.

Refer to 42 CFR 493.1256 for guidance on proper QC practices (U.S. customers only).

If the correct control results are not obtained, do not report patient results. Contact Alere™ Technical Service during normal business hours.

Specimen Collection

Allow all specimens to equilibrate to room temperature (59-86°F, 15-30°C) before testing in Alere BinaxNOW® *Streptococcus pneumoniae*. Just before testing, mix specimen by swirling gently.

URINE (for diagnosis of pneumonia)

Collect urine specimens in standard containers. Store at room temperature (59-86°F, 15-30°C) if assayed within 24 hours of collection. Alternatively, store urine at 2-8°C, or frozen for up to 14 days, before testing. Boric acid may be used as a preservative.

When necessary, ship urine specimens in leakproof containers at 2-8°C or frozen.

CSF (for diagnosis of meningitis)

Collect CSF according to standard procedures and store at room temperature (59-86°F, 15-30°C) for up to 24 hours before testing. Alternatively, properly collected CSF may be refrigerated (2-8°C) or frozen (-20°C) for up to 1 week before testing.

Test Procedure using Visual Interpretation

Urine Specimens, CSF Specimens and Liquid Controls

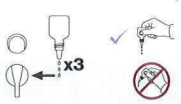
Use a **URINE** specimen when testing for **PNEUMOCOCCAL PNEUMONIA** and a **CSF** specimen when testing for **PNEUMOCOCCAL MENINGITIS**.

***Note**: Use 3 drops of Reagent A when testing liquid specimens.*

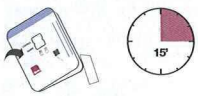
Refer to illustration on pull-out flap. Allow reagents and cards to equilibrate to room temperature (15-30°C) before testing.

1. Bring patient specimen(s) and/or liquid control(s) to room temperature (59-86°F, 15-30°C), then swirl gently to mix. Remove card from its pouch just before use and lay flat.
2. Dip an Alere™ swab into the specimen to be tested, completely covering the swab head. If the swab drips, touch swab to side of collection container to remove excess liquid.
3. There are two holes on the inner right panel of the card. Insert swab into the **BOTTOM** hole (swab well). Firmly push upwards so that the swab tip is fully visible in the top hole. **DO NOT REMOVE SWAB**.

4. Hold Reagent A vial vertically, ½ to 1 inch above the card. Slowly add **three (3)** free falling drops of **Reagent A** to the **BOTTOM** hole.



5. Immediately peel off adhesive liner from the right edge of the test card. Close and securely seal the card. Read result in window 15 minutes after closing the card. Results read beyond 15 minutes may be inaccurate. However, some positive patients may produce a visible specimen line in less than 15 minutes.



***Note**: For convenience, the swab shaft has been scored and may be snapped off **after** closing the card. Avoid dislodging the swab from the well when doing so.*

Test Procedure using the Alere™ Reader

Urine Specimens, CSF Specimens and Liquid Controls

Use a **URINE** specimen when testing for **PNEUMOCOCCAL PNEUMONIA** and a **CSF** specimen when testing for **PNEUMOCOCCAL MENINGITIS**.

***Note**: Use 3 drops of Reagent A when testing liquid specimens.*

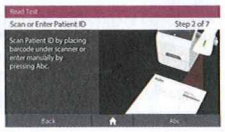
Refer to the Quick Reference Guide. Allow reagents and cards to equilibrate to room temperature (15-30°C) before testing.

1. Bring patient specimen(s) and/or liquid control(s) to room temperature (59-86°F, 15-30°C), then swirl gently to mix. Remove card from its pouch just before use and lay flat.
2. Turn on the Alere™ Reader by pressing the power button. Wait for approximately 10 seconds for the instrument start up sequence. For full instructions on using the Alere™ Reader please refer to Manual and Quick Start Guide. ***Note**: Ensure the correct tray for use with the Alere™ BinaxNOW S. pneumoniae test is in place in the drawer of the Reader.*

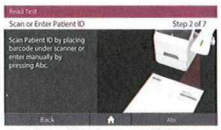
3. Enter Operator ID by placing Operator ID barcode under scanner or entering manually with the keyboard. Enter Operator Password and confirm by pressing 'OK'.

4. Select 'Read Test' on the Alere™ Reader menu. Pressing 'Read Test' on the display will start the reading process.

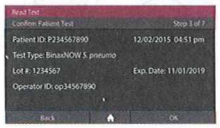
5. Remove card from the foil pouch just prior to testing and lay flat on work bench. Enter Test Device ID by scanning the barcode on the foil pouch, or manually enter the numerical number written below the barcode using the electronic keyboard by pressing Abc.



6. Enter Patient ID by scanning Patient ID barcode under scanner or entering manually with the keyboard.

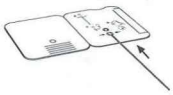


7. Confirm the data entry of User ID, Patient ID and Test Device ID on the screen and press 'OK' to confirm.

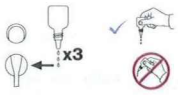


8. Dip an Alere™ swab into the specimen to be tested, completely covering the swab head. If the swab drips, touch the swab to the side of the collection container to remove excess liquid.

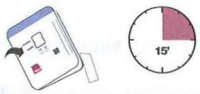
9. There are two holes on the inner right panel of the card. Insert swab into the **BOTTOM** hole (swab well). Firmly push upwards so that the swab tip is fully visible in the top hole. **DO NOT REMOVE SWAB.**



10. Hold Reagent A vial vertically, ½ to 1 inch above the card. Slowly add **three (3)** free falling drops of **Reagent A** to the **BOTTOM** hole.

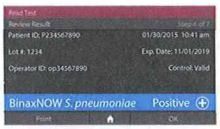


11. Immediately peel off adhesive liner from the right edge of the test card. Close and securely seal the card. Gently snap off swab shaft which is purposely scored. Avoid dislodging the swab from the well when doing so.



12. At 15 minutes open the Reader drawer, insert test device into product specific tray insert and close drawer. A result will be displayed in approximately 15 seconds. **DO NOT OPEN THE DRAWER** until Test Results appear on the screen. ***Note**: Do not read Test Results before or after 15 minutes as they may not be correct.*

13. To print test result press 'Print'.



14. Open drawer, discard test device and close drawer. **DO NOT REINSERT TEST DEVICE ONCE A RESULT HAS BEEN OBTAINED.**

Procedure for Alere BinaxNOW® Swab Controls

***Note**: Use 6 drops of Reagent A for Control Swabs.*

Procedure Using Visual Interpretation

Do not remove card from pouch until test specimen has reached room temperature.

1. Allow reagents and cards to equilibrate to room temperature (15-30°C) before testing. Remove card from the pouch just before use. Lay card flat.
2. There are two holes on the inner right panel of the card. Insert swab into the **BOTTOM** hole. Firmly push upwards so that the swab tip is fully visible in the top hole. **DO NOT REMOVE SWAB.**
3. Hold Reagent A vial vertically, 1/2 to 1 inch above the card. Slowly add **six (6)** free falling drops of Reagent A to the **BOTTOM** hole.
4. Immediately peel off adhesive liner from the right edge of the test card. Close and securely seal the card. Gently snap off swab shaft which is purposely scored. Avoid dislodging the swab from the well when doing so. Read result in window 15 minutes after closing the card. Results read beyond 15 minutes may be inaccurate. However, the Positive Control Swab Sample line may be visible in less than 15 minutes.

***Note:** For convenience, the swab shaft has been scored and may be snapped off **after** closing the card. Avoid dislodging the swab from the well when doing so.*

Procedure Using the Alere™ Reader

1. Allow reagents and cards to equilibrate to room temperature (15-30°C) before testing.
2. Turn on the Alere™ Reader by pressing the power button. Wait for approximately 10 seconds for the instrument start up sequence. For full instructions on using the Alere™ Reader please refer to Manual and Quick Start Guide.
3. Enter Operator ID by placing Operator ID barcode under scanner or entering manually with the keyboard. Enter Operator Password and confirm by pressing 'OK'.
4. Select 'Read QC Test' on the Alere™ Reader menu. Pressing 'Read QC Test' on the display will start the reading process.
5. Remove card from the foil pouch just prior to testing and lay flat on work bench. Enter Test Device ID by scanning the barcode on the foil pouch or manually enter the numerical number written below the barcode using the electronic keyboard by pressing Abc.
6. Select whether a positive or negative control is to be tested and press "OK" to continue.
7. Confirm the data entry of User ID, Test type, Control type and Test Device ID on the screen and press 'OK' to confirm.
8. Lay card flat and run test as follows:
   a) There are two holes on the inner right panel of the card. Insert swab into the **BOTTOM** hole. Firmly push upwards so that the swab tip is fully visible in the top hole. **DO NOT REMOVE SWAB.**
   b) Hold Reagent A vial vertically, ½ to 1 inch above the card. Slowly add **six (6)** free falling drops of **Reagent A** to the **BOTTOM** hole.
   c) Immediately peel off adhesive liner from the right edge of the test card. Close and securely seal the card. Gently snap off swab shaft which is purposely scored. Avoid dislodging the swab from the well when doing so.
9. At 15 minutes open the Reader drawer, insert test device and close drawer. A result will be displayed in approximately 15 seconds. DO NOT OPEN THE DRAWER until QC Test Results appear on the screen. ***Note**: Do not read Test QC Results before or after 15 minutes as they may not be correct.*
10. To print test result press 'Print'.
11. Open drawer, discard test device and close drawer. **DO NOT REINSERT TEST DEVICE ONCE A RESULT HAS BEEN OBTAINED.**

Visual Result Interpretation

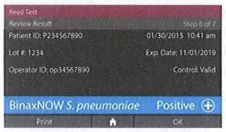
A **negative specimen** will give a single pink-to-purple colored Control Line in the top half of the window, indicating a presumptive negative result. This Control Line means that the detection part of the test was done correctly, but no *S. pneumoniae* antigen was detected.

A **positive specimen** will give two pink-to-purple colored lines. This means that antigen was detected. Specimens with low levels of antigen may give a faint patient line. Any visible line is positive.

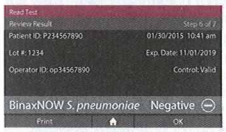
If no lines are seen, or if just the Sample Line is seen, the assay is **invalid**. Invalid tests should be repeated. If the problem persists, contact Alere™ Technical Service.

Alere™ Reader Result Interpretation:

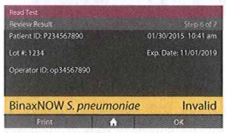
Results will be automatically displayed on the Alere™ Reader screen within 15 seconds of closing the test drawer. Results will be interpreted as positive or negative for *S. pneumoniae* antigen in addition to the procedural control line status.



S. pneumoniae antigen positive result



S. pneumoniae antigen negative result



Invalid test result

Reporting of Results

Result	Recommended Report
Positive Urine	Positive for pneumococcal pneumonia.
Negative Urine	Presumptive negative for pneumococcal pneumonia, suggesting no current or recent pneumococcal infection. Infection due to *S. pneumoniae* cannot be ruled out since the antigen present in the specimen may be below the detection limit of the test.
Positive CSF	Positive for pneumococcal meningitis.
Negative CSF	Presumptive negative for pneumococcal meningitis. Infection due to *S. pneumoniae* cannot be ruled out since the antigen present in the specimen may be below the detection limit of the test.

Limitations

Alere BinaxNOW® *Streptococcus pneumoniae* has been validated using urine and CSF specimens only. Other specimens (e.g. plasma or other body fluids) that may contain *S. pneumoniae* antigen have not been evaluated.

A negative Alere BinaxNOW® *Streptococcus pneumoniae* result does not exclude infection with *S. pneumoniae*. Therefore, the results of this test as well as culture results, serology or other antigen detection methods should be used in conjunction with clinical findings to make an accurate diagnosis.

Alere BinaxNOW® *Streptococcus pneumoniae* has not been evaluated on patients taking antibiotics for greater than 24 hours or on patients who have recently completed an antibiotic regimen. The effects of over-the-counter drugs have not been determined on persons with pneumococcal meningitis.

Streptococcus pneumoniae vaccine may cause false positive results in urine with Alere BinaxNOW® *Streptococcus pneumoniae* in the 48 hours following vaccination. The effect of vaccination has not been determined on persons with pneumococcal meningitis. Hence, it is recommended that Alere BinaxNOW® *Streptococcus pneumoniae* not be administered within 5 days of receiving the *S. pneumoniae* vaccine.

The accuracy of Alere BinaxNOW® *Streptococcus pneumoniae* in urine has not been proven in young children. Performance on CSF in young children, on the other hand, is established (see Performance Data - CSF).

Performance Data - Urine

Analytical Sensitivity

Serotype Evaluation

Forty-four (44) isolates, representing the 23 *S. pneumoniae* serotypes responsible for at least 90% of serious pneumococcal infection in the United States and worldwide, were grown in culture and found to be positive in Alere BinaxNOW® *Streptococcus pneumoniae* at concentrations of 10^5 cells/ml.

Limit of Detection

Alere BinaxNOW® *Streptococcus pneumoniae* limit of detection (LOD), defined as the dilution of positive urine that produces positive Alere BinaxNOW® *Streptococcus pneumoniae* results approximately 95% of the time, was identified by preparing multiple dilutions of a known positive patient urine and running these dilutions with Alere BinaxNOW® *Streptococcus pneumoniae*.

Five (5) different operators each interpreted 20-40 cards run at each dilution for a total of 100-200 determinations per dilution. The following results identify a 1:250 dilution of this particular patient urine as the Alere BinaxNOW® *Streptococcus pneumoniae* LOD.

Urine Dilution	Positive Results per Cards Run	Overall Detection
1:200	100/100	100%
1:250	**95/100**	**95%**
1:300	160/200	80%
1:400	44/100	44%
1:600	8/100	8%

Clinical Sensitivity and Specificity (Retrospective Study)

As part of the retrospective study, urine specimens from 35 blood culture positive pneumococcal pneumonia patients and 338 presumed *S. pneumoniae* negative patients (373 total patients) were collected at 3 different facilities and evaluated in Alere BinaxNOW® *Streptococcus pneumoniae*. Alere BinaxNOW® *Streptococcus pneumoniae* performance was

calculated using standard methods. Sensitivity was 86%, specificity was 94%, and overall accuracy was 93%. Ninety-five percent (95%) confidence intervals are listed below.

		Blood Culture +	Blood Culture −
Alere BinaxNOW® Result	+	30	21
	−	5	317

Sensitivity = 86% (71% - 94%)
Specificity = 94% (91% - 96%)
Accuracy = 93% (90% - 95%)

Clinical Sensitivity and Specificity (Prospective Study)

In a separate seven-center prospective study, Alere BinaxNOW® *Streptococcus pneumoniae* was used to evaluate urine specimens collected from 215 hospitalized patients and outpatients presenting with lower respiratory symptoms or sepsis and from patients otherwise suspected of pneumococcal pneumonia. Patients were considered positive for pneumococcal pneumonia if diagnosed by positive blood culture.

Alere BinaxNOW® *Streptococcus pneumoniae* performed equivalently on both outpatients and hospitalized patients. Ninety-five percent (95%) confidence intervals are listed below.

Outpatient Performance

		Blood Culture +	Blood Culture −
Alere BinaxNOW® Result	+	19	25
	−	2	90

Sensitivity = 90% (70% - 97%)
Specificity = 78% (70% - 85%)
Accuracy = 80% (72% - 86%)

Hospitalized Patient Performance

		Blood Culture +	Blood Culture −
Alere BinaxNOW® Result	+	9	20
	−	1	49

Sensitivity = 90% (60% - 98%)
Specificity = 71% (59% - 80%)
Accuracy = 73% (62% - 82%)

Cross-Reactivity:

Urine Testing

Two hundred seventy (270) different organisms were isolated from the 338 negative patients tested as part of the above retrospective study. Of the 165 organisms isolated from patients with urinary tract infections, 15 (9%) produced positive results. These were 2/2 *Enterobacter cloacae*, 1/2 *Staphylococcus aureus*, 1/1 *Streptococcus* (non A,B), 1/1 *Streptococcus* (non D), 1/17 *Streptococcus* (Group D), 1/3 *Providencia stuartii*, 5/78 *Escherichia coli* and 3 with no identified pathogen. Of the 59 organisms isolated from patients with pneumonia, 3 (5%) were positive, including 1/3 *Mycobacterium kansasii* and 2/15 *Mycobacterium tuberculosis*. One of the 41 (2%) organisms isolated from bacteremic patients, *Proteus mirabilis*, was positive. There was no cross-reactivity with the 5 empyema isolates. Lastly, 4/100 urine specimens collected from people with no known infection were positive.

Due to the retrospective nature of this study, only a limited number of patients with each infection were available for testing and the complete clinical history of each is not known. Therefore, the presence of *S. pneumoniae* co-infection cannot be ruled out. When tested in pure culture (data below), these organisms do not cross-react in Alere BinaxNOW® *Streptococcus pneumoniae*.

Whole Organism Testing

To determine the analytical specificity of Alere BinaxNOW® *Streptococcus pneumoniae*, a panel of 144 potential cross-reactants was compiled, including organisms associated with pneumonia and those likely to be found in the urogenital tract as normal flora or as a result of urinary tract infection. All were evaluated in Alere BinaxNOW® *Streptococcus pneumoniae* at concentrations of 10^6 to 10^9 CFU/mL. Alere BinaxNOW® *Streptococcus pneumoniae* does not cross-react with 143 of the 144 organisms. The single positive organism, *Streptococcus mitis*, is an expected cross-reactant as it shares the antigen against which Alere BinaxNOW® *Streptococcus pneumoniae* is directed. *Streptococcus mitis* is associated with endocarditis, not pneumonia, and is not likely to appear with any frequency in the population intended to be tested with Alere BinaxNOW® *Streptococcus pneumoniae*.[8] The following organisms were tested and produced negative results. When more than one strain was tested, the number is listed in parenthesis.

Acinetobacter sp. (4)	*Mycoplasma* sp.* (3)
Adenovirus* (2&3 pooled)	*Neisseria cinerea*
Alcaligenes faecalis	*Neisseria gonorrhoeae (3)*
Bacillus subtilis	*Neisseria lactamica*
*Blastomyces dermatitidis**	*Neisseria meningitidis*
Bordetella pertussis	*Neisseria polysaccharea*
Branhamella catarrhalis	*Neisseria subflava*
Candida albicans (3)	*Nocardia farcinia**
Candida stellatoides	*Paracoccidiodes brasiliensis**
*Coccidiodes immitis**	Parainfluenzae* (2)
Corynebacterium sp. (3)	*Proteus mirabilis (2)*
Enterobacter cloacae (4)	*Proteus vulgaris (2)*
Enterococcus avium ◊	*Providencia stuartii*
Enterococcus durans ◊	*Pseudomonas sp. (7)*
Enterococcus faecalis ◊ *(6)*	Respiratory Syncitial Virus*
Escherichia coli (8)	Rhinovirus*
Escherichia hermannii (2)	*Salmonella sp. (4)*

Flavobacterium sp. (2)	*Serratia marcescens*
Gardnerella vaginalis	*Sphingobacterium multivorum*
Haemophilus influenzae (10) (types a-f & nontypeable)	*Staphylococcus aureus (6)*
	Staphylococcus sp. (8)
Haemophilus parainfluenzae	*Stenotrophomonas maltophilia*
Histoplasma capsulatum (2)*	*Streptococcus anginosus ◊•*
Klebsiella oxytoca (2)	*Streptococcus bovis ◊*
Klebsiella pneumoniae (3)	*Streptococcus Group A • (2)*
Lactobacillus sp. (5)	*Streptococcus Group B • (8)*
Legionella pneumophila	*Streptococcus Group C ◊•*
Listeria monocytogenes	*Streptococcus Group F ◊•*
Micrococcus luteus (2)	*Streptococcus Group G ◊•*
Moraxella osloensis	*Streptococcus mutans ◊•*
Morganella morganii	*Streptococcus parasanguis ◊•*
Mycobacterium kansasii	*Streptococcus sanguis ◊•*
Mycobacterium tuberculosis	*Trichomonas vaginalis (2)*
* Pure cultures from CDC believed to be in high concentration.	
◊ *Streptococcus* Non A, B (Total number of strains is 16)	
• *Streptococcus* Non D (Total number of strains is 17)	

Interfering Substances

Urine specimens with elevated white blood cells (including loaded per low power field), red blood cells* (including loaded per low power field), protein (including 500 mg/dl), glucose (including >2000 mg/dl), and turbidity (including turbid) were evaluated in Alere BinaxNOW® *Streptococcus pneumoniae* and found not to affect test performance.

**Note that one urine with elevated red blood cells produced an invalid result due to extreme coloration of the test membrane which masked line development.*

Reproducibility Study:

A blind study of Alere BinaxNOW® *Streptococcus pneumoniae* was conducted at 3 separate point of care settings using a panel of blind coded specimens containing negative, low positive, moderate positive, and high positive specimens. Specimens both with and without boric acid were tested. Participants tested each specimen multiple times on 3 different days. Three hundred fifty-seven (357) of the 359 total specimens tested (99.4%) produced the expected result.

Performance Data - CSF

Analytical Sensitivity

Limit of Detection

Alere BinaxNOW® *Streptococcus pneumoniae* limit of detection (LOD) was identified by testing multiple *S. pneumoniae* dilutions in Alere BinaxNOW® *Streptococcus pneumoniae*.

Ten (10) different operators each interpreted 10 cards run at each dilution for a total of 100 determinations per dilution. The following results identify 5 x 10^4 cells per milliliter as Alere BinaxNOW® *Streptococcus pneumoniae* LOD.

Concentration of *S. pneumoniae*	Positive Results per Cards Run	Overall Detection
7.5 x 10^4 cells/ml	100/100	100%
5 x 10^4 cells/ml	**100/100**	**100%**
3 x 10^4 cells/ml	91/100	91%
1.5 x 10^4 cells/ml	44/100	44%
0 cells/ml	0/100	0%

Serotype Evaluation

The four (4) serotypes (6, 14, 19, 23) most commonly associated with pneumococcal invasive disease were grown in culture, diluted to 5 x 10^4 cells/ml in CSF and run in Alere BinaxNOW® *Streptococcus pneumoniae*. Fourteen (14) operators each interpreted 10 cards per serotype for a total of 140 determinations per serotype. All four (4) serotypes were detected 100% of the time at the test LOD (5 x 10^4 cells/ml).

Clinical Sensitivity and Specificity

In a multi-center (4) prospective study, Alere BinaxNOW® *Streptococcus pneumoniae* was used to evaluate CSF specimens collected from 590 hospitalized patients and outpatients presenting with symptoms of meningitis or from patients on whom a lumbar puncture was otherwise indicated. Patients were considered positive for pneumococcal meningitis if diagnosed by positive CSF culture.

Alere BinaxNOW® *Streptococcus pneumoniae* performance was calculated using standard methods. Specificity was 99% (557/560), with a 95% confidence interval of 98% to 100%. Sensitivity was 97% (29/30), with a 95% confidence interval of 84% to 100%. The single culture positive specimen not detected in Alere BinaxNOW® *Streptococcus pneumoniae* was reported as producing only 2 colonies.

		CFS Culture	
		+	–
Alere BinaxNOW® Result	+	29	3
	–	1	557

Sensitivity = 97% (84% - 100%)
Specificity = 99% (98% - 100%)
Accuracy = 99% (98% - 100%)

Cross-Reactivity

CSF Testing

Either enterovirus or bacteria was isolated from 61 of the *S. pneumoniae* negative CSF specimens tested as part of the above prospective study. Sixty (60) of these specimens tested negative in Alere BinaxNOW® *Streptococcus pneumoniae* for a specificity of 98%. The single positive specimen contained Enterococci. However, a second clinical CSF containing Enterococci tested negative in Alere BinaxNOW® *Streptococcus pneumoniae* as did the cultured whole organism (see Whole Organism Testing on the next page).

Bacteria/Virus Isolated From CSF	Specimens Tested	Specificity
Enterovirus	24	100%
Acinetobacter	3	100%
Cryptococcus neoformans	1	100%
C. diphtheriae	1	100%
Enterobacter	2	100%
Enterococci	2	50%
Escherichia coli	2	100%
Haemophilus influenzae type B	1	100%
Klebsiella pneumoniae	2	100%
Morganella morganii	1	100%
Neisseria meningitidis	3	100%
Staphylococcus coagulase negative	9	100%
Staphylococcus coagulase positive	2	100%
Staphylococcus epidermidis	2	100%
Streptococcus Group A	1	100%
Streptococcus Group B	1	100%
Streptococcus viridans	4	100%
Overall Specificity	**61**	**98%**

Whole Organism Testing

In addition to the bacterial and viral infections encountered as part of the prospective study, Alere compiled a panel of potential cross-reactants, including the most common bacterial and viral agents of meningitis. All bacteria were evaluated in Alere BinaxNOW® *Streptococcus pneumoniae* at concentrations ranging from 10^7 to 10^8 CFU/mL. Viruses were tested at 10^5 I.U./mL or greater. Alere BinaxNOW® *Streptococcus pneumoniae* demonstrated 100% specificity, producing negative results for all viruses and bacteria tested.

Burkitt's Lymphoma (Epstein Barr)	*Haemophilus influenzae*, non-typeable (35891)
Coxsackie A7 Virus	Herpes Simplex Virus Type 1
Coxsackie B3 Virus	Herpes Simplex Virus Type 2
Echovirus	*Listeria monocytogenes* (19115)
Enterococcus faecium	*Listeria monocytogenes* (19424)
Haemophilus influenzae A	*Neisseria meningitidis* serogroup A
Haemophilus influenzae B	*Neisseria meningitidis* serogroup B
Haemophilus influenzae C	*Neisseria meningitidis* serogroup C
Haemophilus influenzae D	*Neisseria meningitidis* serogroup D
Haemophilus influenzae E	*Neisseria meningitidis* serogroup L
Haemophilus influenzae F	*Streptococcus oralis* (35037)
Haemophilus influenzae, non-typeable (51997)	

Interfering Substances

CSF specimens with elevated white blood cells (1 x 10^4 cells/ml), red blood cells (30 cells/µl), protein (3 g/dl) and bilirubin (100 µg/ml) were evaluated in Alere BinaxNOW® *Streptococcus pneumoniae* and found not to affect test performance.

Reproducibility Study

A blind study of Alere BinaxNOW® *Streptococcus pneumoniae* was conducted in 3 separate laboratories using a panel of blind coded specimens containing negative, low positive and moderate positive specimens. Participants tested each specimen multiple times on 3 different days. One hundred percent (100%) of the 270 specimens produced the expected result.

Ordering and Contact Information

Reorder numbers:

#710-012: Alere BinaxNOW® *Streptococcus pneumoniae* Antigen Card (12 test kit)
#710-100: Alere BinaxNOW® *Streptococcus pneumoniae* Antigen Card (22 test kit)
#710-010: Alere BinaxNOW® *Streptococcus pneumoniae* Control Swab Pack
#LFR-000: Alere™ Reader

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Technical Support
Advice Line

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References

1. Plouffe, J., S. Moore, R. Davis, R. Facklam. Serotypes of *Streptococcus pneumoniae* blood culture isolates from adults in Franklin County, Ohio. J. Clin. Microbiology 1994; 32:1606-1607.
2. A. Ruiz-Gonzalez, MD, M. Falguera, MD, A. Nogues, MD, M. Rubio-Caballeroa, MD. Is *Streptococcus pneumoniae* the leading cause of pneumonia of unknown etiology? A microbiologic study of lung aspirates in consecutive patients with community-acquired pneumonia. Am. J. of Med. 1999; 106:385-390.
3. Holmberg, H., A. Krook, A. Sjogren. Determination of antibodies to pneumococcal C polysaccharide in patients with community-acquired pneumonia. J. Clin. Microbiology 1985; 22:808-814.
4. Johnston, Jr., R. Pathogenesis of pneumococcal pneumonia. Rev. of Infect. Diseases 1991; 13(Suppl 6):S509-S517.
5. Robbins, J. B., R. Austrian, C. J. Lee, S. C. Rastogi, G. Schiffman, J. Henrichsen, P. H. Makela, C. V. Broome, R. R. Facklam, R. H. Tiesjema, J. C. Parke, Jr. Considerations for formulating the second-generation pneumococcal capsular polysaccharide vaccine with emphasis on the cross-reactive types within groups. J. Infect. Diseases 1983; 148:1136-1159.
6. Wiselka, Martin. Specialists view on pneumococcal meningitis. www.eclipse.co.uk/miningitis.information/text/medic-guide/pm.htm.
7. Cetron, Martin MD, J. Butler MD, D. Jernigan MD, MPH, M. Alexander MA, MPH, S. Roush MT, MPH and R. Breiman MD. Chapter 9: Pneumococcal Disease. Meningitis Foundation of America. www.musa.org/pneumococ.htm.
8. Howard, G.J., Clinical and Pathogenic Microbiology, 2nd ed. 1994. Mosby-YearBook, Inc., St. Louis, MO, p. 267.

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CE

+	Positive
−	Negative
INV	Invalid

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