VITASSAY

Strep. pneumoniae

Rapid test for the qualitative detection of *Streptococcus pneumoniae* in human urine samples.

IUE-7355047 Ed02 February 2018

CE IVD EN

Vitassay

For professional *in vitro* diagnostic use only.

INTENDED USE

Vitassay Strep. pneumoniae is a rapid one step immunochromatographic assay for the qualitative detection of *Streptococcus pneumoniae* in human urine samples.

Simple, non-invasive and highly sensitive screening assay to make a presumptive diagnosis of *pneumoniae* caused by *Streptococcus pneumoniae* in infected humans.

INTRODUCTION

Streptococcus pneumoniae is a potent human pathogen. Infections leads to common diseases such as otitis media, meningitis and pneumonia, which affect several million people and is responsible for significant infant death in developing countries. Worldwide each year, there are over 14 million serious *Streptococcus pneumoniae* infections in children <5 years of age leading to over 800.000 deaths.

There are at least 95 capsular serotypes, but only a few causes the majority of disease. The organism produces a range of colonization and virulence factors, including the polysaccharide capsule, surface proteins and the toxin pneumolysin (PLY).

PRINCIPLE

Vitassay Strep. pneumoniae is a qualitative immunochromatographic assay for the detection of *Streptococcus pneumoniae* in human urine samples.

The test line zone of the nitrocellulose membrane is pre-coated with monoclonal antibodies against *Streptococcus pneumoniae*.

During the process, the sample reacts with the antibodies against *Streptococcus pneumoniae*, forming conjugates. The mixture moves upward on the membrane by capillary action. If the sample is positive, antibodies present on the membrane (test line) capture the conjugate complex and a red line will be visible. Although the sample is positive or negative, the mixture continues to move across the membranes and the blue control line always appears.

The presence of this blue line (in the control zone (C)) indicates that sufficient volume is added; proper flow is obtained and serves as an internal control for the reagents.

PRECAUTIONS

- For professional *in vitro* use only.
- Do not use after expiration date.
- Do not use the test if its pouch is damaged.
- Specimens should be considered as potentially hazardous and handle in the same manner as an infectious agent. A new test must be used for each sample to avoid contaminations errors. Single use device.
- Tests should be discarded in a proper biohazard container after testing.
- Reagents contain preservatives. Avoid any contact with the skin or mucous membrane. Consult safety data sheet, available on request.
- Components provided in the kit are approved for use with the **Vitassay Strep. pneumoniae**. Do not use any other commercial kit component.
- Follow Good Laboratory Practices, wear protective clothing, use disposal gloves, goggles and mask. Do not eat, drink or smoke in the working area.

STORAGE AND STABILITY

Store as packaged in the sealed pouch either at refrigerated or room temperature (2-30ºC/36-86ºF).

The test is stable until the expiration date printed on the sealed pouch.

The test must remain in the sealed pouch until use.

Do not freeze.

MATERIALS

MATERIAL PROVIDED	MATERIAL REQUIRED BUT NOT PROVIDED
▪ 25 Tests/kits **Vitassay Strep. pneumoniae.** ▪ Instructions for use. ▪ 25 Plastic pipettes. ▪ Positive Control: Inactivated S. pneumoniae swab + pipette + testing tube. ▪ Negative Control: S. pneumoniae negative swab + pipette + testing tube. ▪ Reagent (sample and controls diluent) ▪ 25 Testing tubes	▪ Specimen collection container. ▪ Disposable gloves. ▪ Timer.

SPECIMEN COLLECTION

Urine specimens should be collected in standard containers. The samples can be stored at room temperature (15-30ºC/59-86ºF) if assayed within 24 hours of collection. Alternatively, specimens may be stored at 2-8ºC (35.6-46.4ºF) for up to 14 days or at -10ºC to -20ºC (14ºF to -4ºF) for longer periods before testing.

When necessary, urine specimens should be shipped in leak-proof containers at 2-8ºC (35.6-46.4ºF) or frozen.

Vitassay
Parque Tecnológico WALQA
Ctra. N.330, Km.566
22197-Cuarte (Huesca, SPAIN)
www.vitassay.com

SPECIMEN PREPARATION

Allow all specimens to equilibrate to room temperature before testing.

PROCEDURE

Allow tests, urine samples and controls to reach room temperature (15-30ºC/59-86ºF) prior to testing.

Do not open pouches until the performance of the assay.

Patient samples:

- Use a separate testing tube or vial for each sample. Add 3 drops of urine sample. (figure 1).
- Add 1 drop of Reagent into the testing tube or vial and mix (figure 2). Homogenize the sample.
- Remove the **Vitassay Strep. pneumoniae** from its sealed bag just before using it.
- Use a separate pippette and device for each sample or control. Dispense 3 drops from the testing tube in the circular window marked with the letter S (figure 3).
- Read the results at **15 minutes**. Do not read the test results later than 15 minutes.

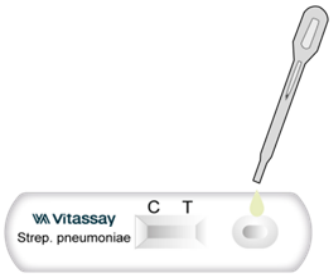
1

Add 3 drops of urine sample.

2

Add 1 drop of Reagent and mix.

3



Dispense 3 drops in the circular window marked with the letter S.

Positive and negative Swabs controls:

- Hold Reagent vertically. Add slowly 13 free falling drops of the Reagent into the testing tube (figure 1b)
- Remove the Positive Control Swab from the pouch and put the swab into the testing tube with reagent (figure 2b), mix 1 minute and extract as much liquid possible from the swab, squeezing the sides of the tube as the swab is withdrawn. Discard the swab (figure 3b)
- Remove the **Vitassay Strep. pneumoniae** from its sealed bag just before using.
- Use a separate pipette and device for each sample or control. Dispense 3 drops from the testing tube, into the circular window marked as the letter S (figure 4b).
- Read the results at **15 minutes**. Do not read the test results later than 15 minutes.

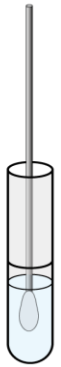
Repeat the procedure for Negative Control swab using the Reagent, same used for test and for positive control swab.

Positive and negative controls should be tested once for each new test kit opened and as otherwise required by your laboratory's standard quality control procedures.

1b

Add 13 drops of Reagent into a testing tube.

2b



Put the swab into the testing tube with Reagent.

3b

Mix and extract as much liquid possible from the swab.

4b

Dispense 3 drops in the circular window marked with the letter S.

Vitassay
Parque Tecnológico WALQA
Ctra. N.330, Km.566
22197-Cuarte (Huesca, SPAIN)
www.vitassay.com

INTERPRETATION OF THE RESULTS

	**NEGATIVE** Only one blue line in the control zone **(C)**	There is no *Streptococcus pneumoniae* presence. No infection caused by *Streptococcus pneumoniae.* Negative control result.
	**POSITIVE** In addition to the blue line (**control line C**), another red line appears, **test line(T)**	There is presence of *Streptococcus pneumoniae*. Infection caused by *Streptococcus pneumoniae.* Positive control result.
ANY OTHER RESULTS		Invalid result, we recommend repeating the assay using the sample with another test. **Note:** Wrong procedural techniques, deterioration of the reagents or insufficient specimen volume are mostly the main reasons for control line failure. If the symptoms or situation persist, discontinue using the test kit and contact your local distributor.

Notes: The intensity of the red colored test line in the result line zone (T) will vary depending on the concentration of antigens in the specimen.

QUALITY CONTROL

Internal procedural control is included in **Vitassay Strep. pneumoniae**. Blue line appearing in the results window is an internal control, which confirms sufficient specimen volume and correct procedural technique.

External Positive and Negative Controls are included in the kit. The use of positive and negative controls is recommended to assure functionality of reagents and proper performance of assay procedure.

LIMITATIONS

- **Vitassay Strep. pneumoniae** must be carried out within 2 hours of opening the sealed bag.
- The use of other samples different from human urine samples has not been established.
- Positive results determine the presence of *Streptococcus pneumoniae* in urine samples; nevertheless, a positive result should be followed up with additional laboratory techniques to confirm the results. A confirmed infection should only be made by a physician after all clinical and laboratory findings have been evaluated and must be based in the correlation of the results with further clinical observations.
- Negative results should not be considered as conclusive; it is possible that the concentration of antigens is lower than the detection limit value. If symptoms or situation still persist, a *Streptococcus pneumoniae* determination should be carried out from a culture or other methods.
- Excretion of *Streptococcus pneumoniae* antigen in urine may vary depending on the individual patient. Antigen excretion may begin as early as 3 days after onset of symptoms and persist for up to 1 year afterwards. A positive result can occur due to current or past infection and therefore is not definitive for infection without other supporting evidence.
- The test is compatible with acid boric used as a preservative up to a percentage of 2%.

EXPECTED VALUES

Streptococcus pneumoniae is a common cause of invasive disease and respiratory tract infections in more and less developed countries. Risk groups for diseases caused by pneumococci such as meningitis, sepsis and pneumonia; include young children, elderly people and patients with immunodeficiencies. Each year, 1 million children younger than 5 years old die from pneumonia and invasive diseases. In USA, the annual number of fatal pneumococcal infections is 40000.

PERFORMANCE CHARACTERISTICS

Analytical sensitivity (detection limit)

Detection limit value of **Vitassay Strep. pneumoniae** is: 0.25 ng/mL of CWPS.

Clinical sensitivity and specificity

An evaluation was performed, with urine samples, comparing **Vitassay Strep. pneumoniae** and another commercial test (BinaxNOW®*Streptococcus pneumoniae* Antigen Card, Alere).

Results were as follows:

		BinaxNOW®*Streptococcus pneumoniae* Antigen Card (Alere)		
		Positive	**Negative**	**Total**
Vitassay Strep. pneumoniae	**Positive**	33	1	34
	Negative	4	73	77
	Total	37	74	111

Vitassay Strep. pneumoniae vs BinaxNOW® *Streptococcus pneumoniae* Antigen Card		
	Mean Value	**95% confidence interval**
Sensitivity	89.2%	74.6-97.0%
Specificity	98.6%	92.7-100.0%
PPV	97.1%	84.7-99.9%
NPV	94.8%	87.2-98.6%

The results showed that **Vitassay Strep. pneumoniae** has a high sensitivity and specificity to detect *Streptococcus pneumoniae*.

Cross reactivity

No cross reactivity was detected against other pathogens that are occasionally present in urine:

Legionella pneumophila

Reproducibility Study

Evaluation studies were performed to determine reproducibility of the **Vitassay Strep. pneumoniae**, including inter-day, inter-laboratory, inter and intra lot, showing high reproducibility in all cases.

REFERENCES

1. WOLFMEIER, H.; RADECKE, J,; SCHOENAUER, R.; ET. AL. "Active release of pneumolysin prepores and pores by mammalian cells undergoing a Streptococcus pneumoniae attack". BIOCHIMICA ET BIOPHYSICA ACTA-GENERAL SUBJECTS 2016; 1860(11): 2498-2509.

2. MITCHELL, A.M.; MITCHELL, T.J. "Streptococcus pneumoniae: Virulence factors and variation". CLINICAL MICROBIOLOGY AND INFECTION 2010; 16(5): 411-418.

3. MIERNYK, KAREN M.; BULKOW, LISA R.; CASE, SAMANTHA L.; ET AL. "Population structure of invasive Streptococcus pneumoniae isolates among Alaskan children in the conjugate vaccine era, 2001 to 2013". DIAGNOSTIC MICROBIOLOGY AND INFECTIOUS DISEASE 2016; 86(2): 224-230.

4. BOGAERT, D; DE GROOT, R; HERMANS, PWM "Streptococcus pneumoniae colonization: the key to pnemococcal disease". Lancet Infectious Diseases 2004; 4: 144-154.

SYMBOLS FOR IVD COMPONENTS AND REAGENTS

IVD	*in vitro* diagnostic device		Keep dry
	Consult instructions for use		Temperature limitation
	Use by		Manufacturer
LOT	Batch code		Contains sufficient for <n> test
DIL	Sample diluent	REF	Catalogue number
Control +	Positive Control	Control -	Negative Control

Parque Tecnológico WALQA
Ctra. N.330, Km.566
22197-Cuarte (Huesca, SPAIN)
www.vitassay.com