BIOSYNEX® S. pneumoniae

Réf: 1030002

Rapid test for the qualitative detection of Capsule Wall Polysaccharides (CWPS) in urine and CSF

CE

INTENDED USE

The **BIOSYNEX® S. pneumoniae** cassette test is a single use rapid immunoassay for the qualitative detection of Capsule Wall Polysaccharides (CWPS) in urine and CSF This kit is intended for use as an aid in the diagnosis of *Streptococcus pneumoniae*. The test is designed for professional *in vitro* diagnostic use only.

SUMMARY

Streptococcus pneumoniae is a Gram-positive bacterium visible as an isolated organism, in pairs or in small chains of bacteria. One of *Streptococcus pneumoniae* major characteristics is its ability to form a capsule, making it pathogenic. This capsule is mostly composed of polysaccharides with glucidic sub-units (CWPS) recognized by human antibodies, making them antigenic. *Streptococcus pneumoniae* is the most frequent cause of bacterial meningitis in the adult or child. It can also cause moderate otitis or pneumonia. Diagnostic is usually made by searching for the bacterium in the cerebrospinal fluid or for the soluble capsule antigens in the patient's urine. Screening for *Streptococcus pneumoniae* is likely to improve and prevent the detection of severe pneumonia at an early stage, reducing mortality.
The risk is 30-100 fold higher for HIV-infected individuals.

TEST PRINCIPLE

The Biosynex® S. pneumoniae test is based on an immunochromatographic technology detecting the Capsule Wall Polysaccharides (CWPS) in urine and CSF specimens.
A pair of monoclonal antibodies anti-CWPS- antigens is used for the Capsule Wall Polysaccharides (CWPS) detection. One is immobilized on the nitrocellulose membrane at the level of the test line: it corresponds to the capture antibody. Another one is labelled with colloidal gold for the subsequent revelation.
During the sample migration, CWPS- antigens, if present in the sample, will form antigen-antibody complexes with the labelled antibodies. These complexes will be captured by the capture antibodies on the test line, creating one purple coloured line generated by gold nanoparticles.

The presence of a purple internal control line in the upper part of the membrane indicates that the result is valid and that the followed procedure is correct.

MATERIAL PROVIDED

- Devices
- Plastic pipettes
- Instructions for use
- Positive Control Vial
- Dropper bottle containing diluent
- One card with barcode to identify the test lot and to check the compatibility of the SD card when used with the BIOSYNEX Reader.

MATERIAL REQUIRED BUT NOT PROVIDED

- Specimen collection container
- Disposable gloves
- Timer

STORAGE AND STABILITY

Store as packaged in the sealed pouch at either refrigerated or room temperature (2-30°C). The test is stable through the expiration date printed on the sealed pouch. The test must remain in the sealed pouch until use.

PRECAUTIONS

– For professional in vitro diagnostic use only
– For single use only
– Directions should be read and followed carefully
– Do not use components after stated expiration date (see pouch and box label)
– Do not use test if pouch is damaged
– Do not eat, drink or smoke in the area where the specimens or kits are handled
– Handle all specimens as if they contained infectious agents
– Observe established precautions for microbiological risks throughout all procedures and standard guidelines for appropriate disposal of specimens
– Wear protective clothing such as laboratory coats, disposable gloves and eye protection when specimens are being tested
– Humidity and high temperature can adversely affect results
– Do not use more than the required amount of liquid
– Do not spill the specimens into the reaction area
– Do not touch the reaction area of the device to avoid contamination
– The test device should remain in the sealed pouch until use
– Store and transport the test device always at 2-30°C
– Do not mix reagents from different lots. Do not interchange solution bottle caps.

SPECIMEN COLLECTION AND STORAGE

Urine samples

Samples should be collected in standard containers. Urine samples can be stored at room temperature (15-30°C) for up to 24 hours after collection. Alternatively, specimens may be stored at 2-8°Cfor up to 14 days or at -20°C for longer periods before testing. Boric acid may be used as a preservative.

When necessary, urine specimens should be shipped in leak-proof containers at 2-8°C or frozen. Bring all specimens to room temperature before testing.

CSF samples

It is recommended to freeze **CSF samples** for long storage to prevent contamination or antigen degradation.
However, CSF can be stored at room temperature (15-30°C) for up to 24 hours, at 2-8°C for one week. CSF samples can also be frozen at -20°C or -80°C for prolonged storage.

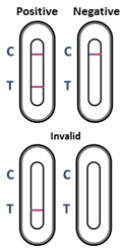
PROCEDURE

The protocol described here is validated on both urine and cerebrospinal fluid.
1. Bring all components of the kit and sample to room temperature.
2. Homogenize the sample and take a fraction of it with the help of the provided disposable pipet.
3. Dispense 1 drop (30µl) -with the included plastic pipette- of the sample into the cassette sample well. Avoid air bubbles and splatters on the reading window.
4. Add immediately 3 drops of diluent.
5. Start the timer. A purplish liquid will appear and move along the membrane.
6. Read the result after 15 minutes. A strong positive signal can appear before that time. **Do not interpret any test band appearing 20 minutes after the sample is dropped in the cassette.**
7. Discard all used components following the infection waste procedure

RESULTS INTERPRETATION

The test result can be read visually or with the help of the ***BIOSYNEX Reader***.

➢ Visual reading:



POSITIVE:
Two distinct colored lines visible: one control line appearing next to the C line marking and one, even faint, next to the T line marking.

NEGATIVE:
Only one colored line visible in the control line zone (C). No line in the test line zone (T).

INVALID:
No colored line visible in the control line zone, whatever the result in the test line zone.

➢ Reading with the reader

- The BIOSYNEX® S. *pneumoniae* test is compatible with the BIOSYNEX Reader, in combination with the SD card 'BIOSYNEX® S. *pneumoniae*'.
  To read results with the reader, please refer to the reader Instructions for use.
- The barcode printed on the supplied card should be scanned to identify the test lot and to check the compatibility of the SD card.

BIOSYNEX
12, rue Ettoré Bugatti – CS28006 Tél. : +33 3 88 77 57 00 info@biosynex.com
67038 STRASBOURG Cedex – France Fax : +33 3 59 81 21 74 www.biosynex.com

QUALITY CONTROL

- Internal procedural controls are included in the test. A coloured line appearing in the control zone (C) ensures that sufficient specimen volume has been loaded and that the correct procedure has been followed.
- Good Laboratory Practices recommend the use of positive and negative controls to check the correct test operating. A positive control that will monitor the entire assay is provided in the kit. Positive controls should be tested once for each new test kit opened and as otherwise required by your laboratory's standard quality control procedures.

LIMITATIONS

1. As with all diagnostic tests, the test result must always be correlated with clinical findings.
2. The results of test are to be interpreted within the epidemiological, clinical and therapeutic context. When it seems indicated, other techniques of reference should be considered.

EXPECTED VALUES

The detection limit is of 0.25 ng/mL of CWPS (reference 3459 from SSI).

PERFORMANCE CHARACTERISTICS.

➢ **Sensitivity and specificity**

A side-by-side study has been done to compare BIOSYNEX® S. pneumoniae with another rapid test on the market. The results are presented in the following table:

		Other rapid test		
		Negative	Positive	
Biosynex® *S.peumoniae*	Negative	113	4	117
	Positive	5	56	61
		118	60	178

Relative sensitivity	93 %	[84-98 %]
Relative specificity	96 %	[90-99 %]
VPP	92 %	[82-97%]
VPN	97 %	[91-99%]
Correlation	95 %	[91-98%]

➢ **Analytical sensitivity:**

Detection limit of the Biosynex® S. pneumoniae is 0.25 ng/mL of recombinant antigen using the reference 3459, SSI.
No hook effect is observed at 10 µg/mL of recombinant CWPS antigen.

➢ **External clinical study**

32 CFS samples have been tested by academic laboratories by Identification via cell culture. 13 CFS samples have also been tested using another rapid test, an agglutination test and PCR.
All obtained results have been compared to the BIOSYNEX® S. pneumoniae rapid test. Results are showed in the below tables

		Bacterial culture		
		Positive	Negative	Total
Cassette	Positive	13	2	15
	Negative	0	17	17
	Total	13	19	32

		Agglutination/rapid test		
		Positive	Negative	Total
Cassette	Positive	11	0	11
	Negative	0	2	2
	Total	11	2	13

		PCR		
		Positive	Negative	Total
Cassette	Positive	11	0	11
	Negative	2	0	2
	Total	13	0	13

The global correlation between results obtained with the Biosynex cassettes in comparison with bacterial culture, rapid test and agglutination test is of 100% on the available samples.
Only the PCR method is more sensitive when compared to Biosynex test.

➢ **Interference**

Albumin (from 0.68 to 1.06 g/L) or bilirubin (at 120 or 300 mg/L) does not influence the test performances.
A presence of 10 µL/mL of blood does not influence results but a presence of 100 µL/mL can create false-negative results on weak positives due to difficulty of reading the results with a reddish background noise.

REFERENCES

1. Ehara N., Fukushima K., Kakeya H., Mukae H., Akamatsu S., Kageyama,A., Saito A., and Kohno S., A novel method for rapid detection of *Streptococcus pneumoniae* antigen in sputum and its application in adult respiratory tract infections, Journal of Medical Microbiology (2008), 57, 820–826
2. Murdoch, D., Laing R., Mills G., Karalus N., Town G.I., Mirrett S., and Reller B., Evaluation of a Rapid Immunochromatographic test for detection of a *Streptococcus pneumonia* antigen in urine samples from adults with community-acquired pneumonia, J. Clin. Microbiol. 2001, 39(10):3495.
3. Statens Serum Institut, Testbook en Serotyping, Virulence Factors and Enzyme-linked Immunosorbent Assay (ELISA) for measuring Pneumococcal antibodies, 1st edition April 2013
4. AlonsoDeVelasco E. Verheul, A., Verhoef J., and Snippe H., *Streptococcus pneumoniae*: Virulence Factors, Pathogenesis, and Vaccines, Eijkman-Winkler Institute of Medical and Clinical Microbiology, University Hospital, Utrecht, The Netherlands, Microbiological reviews, D*ec. 1995, p. 591–603 Vol. 59, No. 4*
5. Clothilde Manzano. Structural and functional characterization of pilus components of *Streptococcus pneumoniae*: toward a better understanding of the biogenesis of pili.. Biomolecules. Universit_e Joseph-Fourier - Grenoble I, 2009.
6. Lumin Zhang, Zihai Li, Zhuang Wan, Andrew Kilby, J Michael Kilby and Wei Jiang, Humoral Immune Responses to *Streptococcus pneumoniae* in the Setting of HIV-1 Infection, in Vaccine. 2015 Aug 26; 33(36): 4430–4436. Department of microbiology and immunology, Medical University of South Carolina, Charleston
7. Bles P., de Mast Q., van der Gaast-de Jongh C., Kinabo G., Kibiki G., van de Ven A. and de Jonge M., Antibiotic resistance of *Streptococcus pneumoniae* colonising the nasopharynx of HIV-exposed Tanzanian infants, Tropical Medicine and International Health, 2015, vol 00 N° 00

SYMBOLS

	Attention, see instructions for use	LOT	Lot number
IVD	For *in vitro* diagnostic use only		Manufacturer
2°C 30°C	Store between 2-30°C		Do not reuse
Σ	Tests per kit	REF	Catalog number
	Expiry	DIL	Diluent

Version 02 EN 08/2016

Distributed by

BIOMEDICA
www.bmgrp.eu

BIOSYNEX
12, rue Ettoré Bugatti – CS28006 Tél. : +33 3 88 77 57 00 info@biosynex.com
67038 STRASBOURG Cedex – France Fax : +33 3 59 81 21 74 www.biosynex.com