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THE STANDARDS INSTITUTION OF ISRAEL

No-step Strep A Test

The Instant Strep A Test for Rapid Diagnosis of Streptococcal Pharyngitis

Catalogue Number R-6009 (1 test)

Instruction for Use

INTENDED USE

No-step Strep A Test is a unique, instant immunochromatographic *in-vitro* diagnostic assay for the direct detection of beta-hemolytic group A streptococcus (GAS) antigen in throat swab specimens to aid in the diagnosis of streptococcal pharyngitis. The test utilizes Novamed's proprietary technology which facilitates minimal user involvement in the testing process. For ease, simplicity of procedure and rapidity of results, the No-step Strep A Test is recognized as a true Point-of-Care Test, allowing to correctly diagnose the streptococcal pharyngitis directly in doctors' offices, polyclinics and at home preventing antibiotic overuse and mishandling of the test.

SUMMARY

Rapid antigen testing to detect GAS infection provides important information for the antibiotic decision making for patients presenting with acute pharyngotonsillitis. The Infectious Diseases Society of America (IDSA) guideline 3 on streptococcal pharyngitis recommends using a rapid test in patients with a modest probability of GAS infection, treating those with a positive rapid test and withholding antibiotics in rapid test negative patients. The guideline recommends culturing rapid test negative children and treating patients with positive cultures; the guideline does not recommend culturing the rapid test negative adults given the lower prevalence and significantly reduced chance of non-suppurative complications of the disease in the adult population[3], unless the clinician wishes to increase diagnostic sensitivity.

PRINCIPLE

The No-step Strep A Test utilizes double antibodys sandwich immunoassay for the detection of GAS carbohydrate antigen. The test device consists of plastic housing containing a test membrane which has been pre-coated with anti-GAS antibody-dye conjugate on the test band region and anti-antibody on the control band region. When the device is activated, the specimen is absorbed into the membrane by capillary action, mixed with the antibody-dye conjugate, and flows across the pre-coated membrane. When the GAS antigen levels in specimens are at or above the target cutoff (the detection limit of the test), the antigen binds to the antibody-dye conjugate and are captured by anti-GAS antibody immobilized in the Test region (T) of the device. This produces a colored Test band and indicates a positive result. When the GAS antigen levels are zero or below the target cutoff, there is no a visible colored band in the Test region (T) of the device. This indicates a negative result. Unbound conjugate binds to the reagents in the control region (C), producing a colored band, indicating that the membrane and the reagents are functioning correctly.

KIT COMPONENTS

Each No-step Strep A Test contains everything required to perform 1 determination:

- 1 No Step Strep A Test device placed in a plastic case with desiccants.
- 1 sterile polyester swab / applicator.
- 1 leaflet of the Instruction for Use.

STORAGE AND STABILITY

Store as packaged in the sealed case either at refrigeration or room temperature (+2+30ºC). Do not freeze.

PRECAUTIONS

- *Do not use after expiration date.*
- *Do not use the test if case is damaged.*
- *The assay must be carried out within 2 hours of opening the sealed case.*

SPECIMEN COLLECTION

1. *Only use the sterile swab provided in the device.*
2. *For throat specimen collection, swab the posterior pharynx, tonsils and other inflamed areas. Avoid touching the tongue, cheeks and teeth with the swab.*
3. *Testing should be performed as soon as possible after the specimens have been collected. However, the swab specimens may be stored in a clean, dry plastic tube for up to 8 hours at room temperature.*

ASSAY PROCEDURE

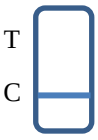
1. *Collect a throat swab specimen with the provided sterile swab and insert it into the hole on the blue cap until it stops.*

2. *Using two thumbs, press down the blue cap of the device, until it stops.* ***Notes:***
   - ***AFTER ABOUT 1 MINUTE, LIFT THE SWAB SLIGHTLY AND RE-INSERT IMMEDIATELY.***
   - *Do not move the device until the result is received.*

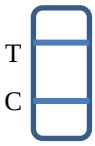
Read results 5 minutes after lifting and reinserting the swab.
Do not interpret thereafter.

INTERPRETATION OF RESULTS

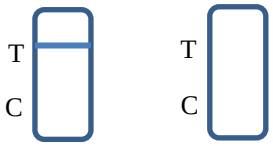
NEGATIVE: only one colored band appears at the control region (C). No band is visible at the test region (T).



POSITIVE: in addition to the control band (C), a distinguishable colored band appears at the test region (T) in the results window.



INVALID: if no control band (C) is visible, the test is inconclusive. The test should be repeated using a new device.



Notes on the interpretation of results

The intensity of the colored band at the test region (T) in the results window will vary depending on the concentration of antigens in the specimen. Any color intensity indicates a positive result.

PERFORMANCE CHARACTERISTICS

Analytical Sensitivity

To determine the analytical sensitivity of the No-step Strep A Test, the S. pyogenes microorganisms were grown in broth culture. The detection limits of the No-step Strep A Test was determined to be 2.5×10^5 CFU per test/swab.

Sensitivity and Specificity

In total, six hundred eighteen (n=618) throat swab specimens were collected from outpatients with clinical suspicion of streptococcal throat infections visiting the municipal hospital in Russia between March and October 2016 (n=488) and public outpatient reference lab in Israel in March 2017 (n=130). The samples were collected during a visit of outpatients in the microbiological lab at the hospital or the polyclinic as directed by the physician. Two swab samples were collected simultaneously from each of 488 patients, one for immediate testing by No-step Strep A Test and second for immediate inoculation onto the blood agar plates. Each swab sample collected from 130 outpatients visiting the polyclinic was placed into a tube with Amies transport medium and sent to a central reference lab. Upon admission to the lab, the swab was firstly inoculated onto the blood agar plate and then returned to the Amies transport medium and stored at room temperature for 24 hours. After observation of the results of growth on blood agar, each swab was tested using No-step Strep A Test. Subsequently, the results of both tests were compared.

Culture and identification

TSA supplemented with 5% sheep blood plates were incubated at 35±2°C at 5-10% CO_2 atmosphere and examined at 24 hours for presence of beta-hemolytic colonies. The negative culture plates were incubated for an additional 18-24 hours. The typical beta-hemolytic and catalase-negative colonies were tested for the presence of pyroglutamate aminopeptidase (PYR) by using the

StrepSwift test (REF: BI-601, Novamed Ltd.) and finally were confirmed with a commercially available REF: R6202 PathoDx® Strep Grouping latex agglutination assay (Remel, USA).

Results

Overall, both throat culture and No-step Strep A Test were identically negative in 438 (70.9%) specimens and positive in 174 (28.2%) specimens. Of the 618 total specimens, 440 (71.2%) were confirmed to be negative and 178 (28.8%) were confirmed to be positive by culture.

Table1. Total results

		Throat Culture		
		Positive	Negative	Total
No-step Strep A Test	Positive	174	2	176
	Negative	4	438	442
	Total	178	440	618

95% (CI) Confidence Intervals

Sensitivity = 97.8%; Specificity = 99.7%; Positive Predictive Value =98.9%; Negative Predictive Value =99.1 %; Diagnostic Accuracy = 99 %.

Discussion

In patients with GAS confirmed pharyngotonsillitis, the positive predictive value of the No-step Strep A Test (the chance of a positive culture if the No-step Strep A Test is positive) was very high at 98. 9% (174/178). The negative predictive value (the chance of a negative culture if the No-step Strep A Test is negative) for the whole patients, was extremely high at over 99.1% (438/440). Furthermore, the No-step Strep A Test sensitivity was very high at 97.8%.

Conclusion

No-step Strep A Test is an extremely simple, quick, and reliable test. It should be incorporated in the initial assessment of patients with fever and URTI/pharyngotonsillitis. Its use will allow the practicing of a 'no initial antibiotics' policy safely, with a marked reduction in antibiotic use in these children.

QUALITY CONTROL

Internal Quality Control

Internal procedural control is included in the test. A colored line appearing in the control line region (C) is an internal positive procedural control. It confirms sufficient specimen volume, adequate membrane wicking and correct procedural technique.

Cross-Reactivity

The following organisms were tested at 1×10^6- 1×10^8 CFU per test and were all found to be negative when tested with the No-step Strep A Test.

Streptococcus Group B	*Streptococcus mutans*	*Pseudomonas aeruginosa*
Enterococcus faecalis	*Neisseria meningitidis*	*Escherichia coli*
Streptococcus Group C	*Neisseria gonorrhoeae*	*Klebsiella pneumoniae*
Streptococcus Group D	*Neisseria sicca*	*Serratia marcescens*
Streptococcus Group F	*Neisseria subflava*	*Haemophilus influenzae*
Streptococcus Group G	*Staphylococcus aureus*	*Candida albicans*
Streptococcus pneumoniae	*Staphylococcus epidermidis*	*Bordetella pertussis*
Streptococcus sanguis	*Corynebacterium diphtheriae*	*Branhamella catarrhalis*

LIMITATIONS

This test will only indicate the presence of GAS antigen in the specimen from both viable and non-viable Group A Streptococcus bacteria.

Bibliography:

1. The Current Evidence for the Burden of Group Streptococcal Diseases. Department of Child and Adolescent Health and Development World Health Organization. WHO/FCH/CAH/05.07. World Health Organization 2005.
2. Shulman ST, Bisno AL, Clegg HW, Gerber MA, Kaplan EL, et al. (2012) Clinical practice guideline for the diagnosis and management of group A streptococcal pharyngitis: 2004 update by the Infectious Diseases Society of America. Clin Infect Dis 55: 1279–1282. [PubMed]
3. Manual of Clinical Microbiology. 2011. 10th Edition.

SYMBOLS FOR IVD COMPONENTS AND REAGENTS

	Manufacturer	IVD	For *in vitro* diagnostic use only
EC REP	Authorized representative		Consult instructions for use
REF	Catalogue Code		Temperature limitation
LOT	Lot Number		Use by
	28 Pierre Köenig St., P.O.B 53231, Jerusalem 91531, Israel. Tel. 972-2-6781561; Fax: 972-2-6781852	EC REP	MedNet GmbH, Borkstrasse 10, D-48163 Münster, Germany. Tel. 49-251-32266-0; Fax: 49-251-32266-22
REF	R-6009		