Results are as follows:

CLIAwaived™ Inc. Rapid Strep A Test				
		+	-	Total
Culture	+	46	1	47
	-	4	142	146
Total		50	143	193

Sensitivity: 97.9% (95% CI, 88.7% to 99.9%)
Specificity: 97.3% (95% CI, 93.1% to 99.2%)
Overall Agreement: 97.4 (95% CI, 94.1% to 99.1%)

EXPECTED VALUES

It is believed that approximately 19% of all upper respiratory tract infections are caused by Group A Streptococci.[5] Infection is most prevalent in winter and early spring, with most cases arising in patients living in highly populated areas.

LIMITS OF DETECTION

Group A Streptococcus organisms were grown and tested at different concentrations. The test was capable of detecting 1.5 x 10^5 organisms per test.

SPECIFICITY

To confirm the specificity of the CLIAwaived™ Inc. Rapid Strep A Test, bacterial cultures likely to be found in the respiratory tract were tested at 3.0x10^5 to 2.8x10^9 organisms/test and all yielded negative results. The organisms tested are listed below:

Branhamella catarrhalis
Haemophilus influenzae
Enterococcus faecalis
Streptococcus mutans
Neisseria subflava
Streptococcus pneumoniae
Neisseria gonorrhaeae
Staphylococcus aureus
Pseudomonas aeruginosa
Candida albicans
Neisseria mucosa
Neisseria meningitides
Group B Streptococcus
Group C Streptococcus
Group F Streptococcus
Group G Streptococcus
Ungrouped Streptococcus

To further confirm the specificity, the following eleven strains of Group A Streptococcus were tested and positive results were detected at 1.5 x 10^5 organisms/test.

SS-091, SS-482, SS-633, SS-635, SS-754, BRS0023A, SS-410, SS-496, SS-634, SS-721, SS-799

Physician Office Lab Study
The CLIAwaived™ Inc. Rapid Strep A Test was evaluated at three different physician's offices using a panel of five samples. Physician office personnel with diverse educational backgrounds performed the testing. The sample panel consisted of two negative, one low positive, one medium positive and one high positive. One hundred percent (100%) of the forty-five (45) samples tested produced the expected results.

Lay Person User Study
Individuals having diverse educational backgrounds evaluated the CLIAwaived™ Inc. Rapid Strep A Test at three different sites. Each site tested a coded panel consisting of a negative, low positive and high positive. There was greater than ninety-seven percent (97%) agreement (175/180) of the expected results.

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4. Finegold, S.M. and Martin, W.J. 1982. Microorganisms Encountered in Respiratory Tract Infections, p. 66, Diagnostic Microbiology, 6th Edition. The C.V. Mosby Co. St. Louis.
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	Do not reuse		Use by YYY-MM-DD or YYYY-MM
LOT	Batch Code	SN	Serial number
	Caution, consult accompanying documents	REF	Catalog number
	Manufacturer		Temperature limitation
	Contains sufficient for <n> tests		Keep away from sunlight
IVD	*In vitro* diagnostic medical device		Keep away from moisture
	Consult instructions for use	CE	CE Mark
	Recycle	EC REP	Authorized representative in the European Community

CLIA*waived*™ Inc.

Rapid Strep A Test

CLIA COMPLEXITY - WAIVED

Intended for health care professional use only, not for self-testing.
A certificate of CLIA waiver is required to perform the testing in a waived setting.

EC REP

Emergo Europe
Molenstraat 15
2513 BH, The Hague
The Netherlands

Tel: 31-0-70-345-8570
Fax: 31-0-70-346-7299

IVD

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Distributed by:
CLIAwaived, Inc., San Diego, CA 92121

P/N: CLIA-ST-501
Rev.: CLIA.11.16.11

Revised November 2011

Made in the U.S.A. CE

CLIA*waived*™ Inc.

Rapid Strep A Test CE

CLIA COMPLEXITY – WAIVED

For the presumptive *in vitro* qualitative detection of Group A Streptococcal antigen from throat swab specimens

FOR TECHNICAL ASSISTANCE
Call 1-888-882-7739

OUTSIDE THE USA
Call 1-848-481-5031

For Professional *In Vitro* Diagnostic Use Only
Store at 2ºC to 30ºC

PLEASE READ ALL INSTRUCTIONS BEFORE BEGINNING ASSAY

INTENDED USE

The CLIAwaived™ Inc. Rapid Strep A Test is a rapid, visual test for the presumptive in vitro qualitative detection of Group A Streptococcal antigen from throat swab specimens. The test is for professional use only.

SUMMARY AND EXPLANATION

Group A Streptococcus *(S. pyogenes)* is the principal cause of respiratory tract infection in humans. Streptococcal pharyngitis, which primarily affects children and young adults, can lead to serious complications such as rheumatic fever or acute glomerulonephritis. The rapid and accurate detection of Group A Streptococcus is important for the early initiation of antibiotic therapy in the treatment of Streptococcal pharyngitis. Traditional diagnostic tests have relied on overnight culture together with confirmation by serological or biochemical methods.[1-3] Newer methods based on immunochemical detection of Streptococcal antigen have been developed, which do not require growth of the organism, nor do these methods require viable organisms for detection of the antigen.

PRINCIPLE OF THE TEST

The CLIAwaived™ Inc. Rapid Strep A Test begins with an extraction of Group A Streptococcal antigen from the throat swab. After the extraction has been completed, the test dipstick is placed into the extraction mixture and observed for the formation of colored lines. The specimen is absorbed and migrates via capillary action through a membrane that contains dried gold conjugated antibody that is specific for Group A Streptococcal antigen. If Group A Streptococcal antigen is present, a "half-sandwich" immunocomplex is formed. The membrane contains immobilized antibody to Group A Streptococcal antigen, which binds the "half sandwich" complex. Thus, in the presence of Group A Streptococcal antigen, a "whole sandwich" immuno-complex is formed and a visible, pink-colored line develops in the specimen zone of the dipstick. In the absence of Group A Streptococcal antigen, a "sandwich" immunocomplex is not formed and a negative result is indicated. To serve as a procedural control, a pink colored control line will always appear in the control zone regardless of the presence or absence of Group A Streptococcal antigen.

REAGENTS & MATERIALS PROVIDED

1. Test dipsticks-containing gold conjugated with anti-strep antibody specific for Group A Streptococcal antigen.
2. Reagent A (7 ml)-Sodium Nitrite (2.0 M), avoid contact.
3. Reagent B (7 ml)-Acetic Acid (0.5 M), avoid contact.
4. Positive control (1 ml)-inactivated Group A Streptococcus in solution with 0.1% Sodium Azide. Mix well before use.
5. Negative control (1 ml)-inactivated Group B Streptococcus in solution with 0.1% Sodium Azide. Mix well before use.
6. Collection Swabs.
7. Extraction Tubes.
8. Package Insert.

MATERIALS REQUIRED BUT NOT PROVIDED

Timer.

STORAGE

The kit and components can be stored at room temperature (15º - 30ºC) or refrigerated at 2º - 8ºC. If refrigerated, the components must be brought to room temperature before use.

PRECAUTIONS

1. For in vitro diagnostic use only.
2. Do not mix reagents or dipsticks from different lots.
3. Swabs from other suppliers have not been validated. Using swabs other than those provided with the kit may affect performance of the kit.
4. Do not use beyond the expiration date.
5. Reagents A & B contain a mild irritant. Avoid contact with eyes, skin or mucous membranes. If accidental contact occurs, flush thoroughly with water and seek appropriate medical attention.
6. The test dipsticks must remain in the closed canister until ready for use. Record the date the canister is first opened in the place provided on the label. The test strips should not be used beyond 90 days after opening the canister. Dipsticks from a pouch must be used immediately after the pouch is opened.
7. Be careful not to touch the tongue, cheeks, or lips with the swab. Sampling in these areas, as well as excess saliva, may affect the performance of the kit.
8. In accordance with the principles of Good Laboratory Practice it is strongly recommended that all specimens be treated as potentially infectious and handled with all necessary precautions.
9. The Positive & Negative controls included in the kit contain sodium azide as a preservative, which may react with lead or copper in plumbing to form potentially explosive metal azides. Upon disposal, always flush with a large volume of water to prevent azide buildup in drains.
10. If the laboratory modifies the test system instructions, then the test is considered high complexity and subject to all applicable CLIA requirements.

SPECIMEN COLLECTION & PREPARATION

1. Only sterile rayon collection swabs, supplied with the kit, should be used. Swabs from other suppliers have not been validated. Using swabs other than those provided with the kit may affect performance of the kit. Do not use cotton, wooden shaft or calcium alginate swabs.
2. A throat swab should be obtained using a standard collection method such as outlined by Finegold and Martin in the sixth edition of Diagnostic Microbiology.[4]
3. Depress the patient's tongue with a blade or spoon and rub the swab firmly over the back of the throat, over both tonsils, and any areas of redness. Be careful not to touch the tongue, cheeks, or lips with the swab.
4. Swabs may be transported using modified Stuart's media or equivalent. Do not use transport media containing gelatin or charcoal.
5. Test specimen swabs immediately after collection. If the swab is not to be tested immediately, it may be stored in a clean, dry, capped tube for up to 72 hours at 2º - 8ºC.
6. If culture results are required, gently streak swab on appropriate medium before performing the test. The extraction procedure will cause bacteria to become inactive, therefore not allowing the organism to be cultured. Alternatively, a second swab may be collected for culture.

LIMITATIONS OF THE PROCEDURE

1. The quality of the swab sample is extremely important to the accuracy of the test. False negatives may result from improper collection or storage. When there is suspicion of having Strep A pharyngitis, additional testing may be done using cultures.
2. The CLIAwaived™ Inc. Rapid Strep A Test should only be used with properly collected throat swabs. The use of specimens from other sites or the use of other samples such as saliva, sputum or urine has not been established.
3. This test will not distinguish between a carrier state and those individuals with an active disease state. Pharyngitis may be caused by organisms other than Group A Streptococcus. In rare cases, specimens heavily colonized with Staphylococcus aureus can give false positive results. If test results are inconsistent with clinical presentation, a follow-up throat swab should be obtained for culture confirmation. All results should be evaluated in conjunction with all clinical and laboratory findings.
4. A negative test result means there is no detectable amount of extracted Group A Streptococcal antigen on the throat swab. A negative result may be obtained if no Group A Streptococcal antigen is present in the specimen or if the amount of extracted antigen is below the detection limit of the test.
5. The American Academy of Pediatrics (1994 Redbook, p. 443) recommends that cultures be performed on throat swab specimens with negative antigens results.
6. Some Group D and Group C strains of Streptococci gave false positive results in clinical trials.

TEST PROCEDURE

Step 1
Add 3 drops of Reagent A to the Extraction Tube.

Step 2
Add 3 drops of Reagent B to the Extraction Tube.

Step 3
Add the specimen swab into the extraction tube. Mix the solution thoroughly by swirling the swab vigorously in the extraction tube several times. Allow the swab to soak for at least 1 minute but no longer than 15 minutes.

Step 4
Remove all liquid from the swab by rolling it against the side of the extraction tube and then squeezing the swab between the flexible sides. Discard the swab in a biohazard container.

Step 5
Place the dipstick into the extraction tube. Gently stir for 10-15 seconds.

Step 6
Allow the dipstick to remain in the tube to absorb as much of the extraction liquid as possible. Read results after 5 minutes. Do not interpret results after 10 minutes.

INTERPRETATION OF RESULTS

Negative Results

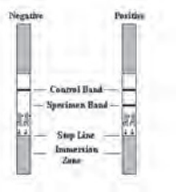
The test is negative if only one pink colored line appears in the control zone (see illustration). A negative test means that the assay did not detect any antigen to Group A Streptococcus in the specimen or the levels of antigen are below the detection level of the assay. The specimen should be cultured for up to 48 hours to rule out absence of Group A Streptoccus infection. If clinical symptoms are not consistent with results, obtain another specimen for culture.

Positive Results

The test is positive if two pink colored lines appear. One pink colored line will appear in the specimen zone and one in the control zone (see illustration). **Any pink colored line** in the specimen zone should be considered positive. The pink colored line in the control zone may be lighter or darker than the specimen line. A positive test means that the assay detected antigen to Group A Streptococcus in the specimen.

Invalid Results

The test is invalid if no pink colored line appears in the control zone even if a pink colored line appears in the specimen zone.



QUALITY CONTROL

Internal Controls

Each test device includes 3 levels of internal procedural controls.

- Reagent A contains a pink dye. When combined with Reagent B, the color changes from pink to light yellow. This color change is an internal extraction reagent control indicating that the two reagents have been mixed and are functioning properly.
- The pink band in the control area of the dipstick is a positive procedural control. This shows that the dipstick has absorbed enough sample and that the test is functioning properly. If the colored band fails to appear in the C (control) area, the test result is invalid and should be repeated.
- A clear background is an internal negative procedural control. The background color should be white to light pink and should not interfere with the reading of the test result. An intensely red background may interfere with the ability to read the test result, therefore the test should be repeated.

External Controls

Each kit contains a set of Positive and Negative controls to be used for external quality testing of the reagents and dipsticks.

External controls should be used according to individual laboratory guidelines. If the controls do not work as expected, repeat the test or contact Technical Assistance.

Quality Control Testing Procedure

1. Follow instructions in the test procedure for preparation of Extraction Reagents.
2. Add 1 drop of control.
3. Place a clean swab in the test tube.
4. Follow instructions in test procedure (steps 3 through 6) to complete test.

PERFORMANCE CHARACTERISTICS

Comparison Study

A multi-site evaluation of the CLIAwaived™ Inc Rapid Strep A Test was carried out to determine the clinical performance characteristics of the test relative to another commercially available test. Two throat swabs were collected from patients presenting with symptoms of pharyngitis. A total of 104 patients were tested.

CLIAwaived™ Inc. Rapid Strep A Test				
		+	-	**Total**
Other Commercially Available Test	+	28	2	**30**
	-	5	69	**74**
Total		**33**	**71**	**104**

The CLIAwaived™ Inc. Rapid Strep A Test had an overall agreement of 93.3% to that of the other commercially available test. The four (4) samples that were found to be CLIAwaived™ Inc. Rapid Strep A Test positive (+) and other commercially available test negative (-) were confirmed positive (+) by culture. The sample that was other commercially available test positive (+) and CLIAwaived™ Inc. Rapid Strep A Test (-) was found to be positive by culture.

Correlation Study

A multi-site evaluation of the CLIAwaived™ Inc. Rapid Strep A Test was carried out to determine the clinical performance characteristics of the test relative to standard culture techniques. Throat swabs were collected from patients presenting with symptoms of pharyngitis. A total of 193 patients were tested. Swabs were first used to streak blood agar plates before testing. All cultures were confirmed for the presence of Group A Streptococcus using serological grouping methods.