HEALGEN®

Rapid Strep A Antigen Test

INSTRUCTIONS FOR USE

REF GCSTR-501Ca

CLIA Categorization: Waived

IVD | ℞ Only

A Certificate of Waiver is required to perform this test in a CLIA Waived environment. Laboratories with a Certificate of Waiver must follow the manufacturer's instructions for performing the test 42 CFR 493.15(e)(1) Failure to follow the instructions or modification to the test system instructions will result in the test no longer meeting the requirements for waived classification.

INTENDED USE

The Rapid Strep A Antigen Test is a rapid chromatographic immunoassay for the qualitative detection of Streptococcus pyogenes (Group A β-hemolytic Streptococcus, Strep A) antigen from throat swab specimens of symptomatic patients to aid in the diagnosis of Group A Streptococcus bacterial infection.

All negative test results should be confirmed by bacterial culture because negative results do not preclude infection with Group A Streptococcus and should not be used as the sole basis for treatment.

SUMMARY

Streptococcus pyogenes is non-motile gram-positive coccus, which contains the Lancefield group A antigen that can cause serious infections such as pharyngitis, respiratory infection, impetigo, endocarditis, meningitis, puerperal sepsis, and arthritis. Left untreated, these infections can lead to serious complications, including rheumatic fever and peritonsillar abscess.[1] Traditional identification procedures for Group A Streptococci infection involve the isolation and identification of viable organisms using techniques that require 24 to 48 hours or longer.[2,3]

The Rapid Strep A Antigen Test is a rapid test to qualitatively detect the presence of Group A Streptococcal antigen in throat swab specimens, providing results within 5 minutes. The test utilizes specific and sensitive antibodies reactive to the Rapid Strep A Antigen and is specific to group A with no cross-reactivity from other groups of Streptococci.

PRINCIPLE

The Rapid Strep A Antigen Test is a qualitative, lateral flow immunoassay that uses antibodies to detect carbohydrate antigen from Strep A in throat swabs. During testing, a throat swab is collected and added to buffer. The sample is added to the test. If Strep A antigen is present in the sample, a colored line will form at the test line at "T", which means the test is positive. If Strep A antigen is not in the sample, a test line will not appear at "T" which means the test is negative. A colored line will always appear at the control line "C" to indicate the test is working and proper test procedure has been followed.

MATERIALS PROVIDED

25	**Test Strips**
25	**Sterile swabs**
25	**Disposable extraction tubes** *One tube for each test strip*
1	**Reagent A*** *10mL; 2M Sodium Nitrite*
1	**Reagent B*** *10mL; 0.2M Acetic Acid*
1	**Positive Control** *1mL: Non-viable Strep A; 0.05% ProClin™ 300*
1	**Negative Control** *1mL: Non-viable Strep C; 0.05% ProClin™ 300*
2	**Tube Holders**
1	**Instructions for Use**
1	**Quick Reference Instructions**

** Reagent A and B are caustic. Avoid contact with eyes, sensitive mucous membranes, cuts, abrasions, etc. If these reagents contact the skin or eyes, flush with a large volume of water.*

MATERIALS REQUIRED BUT NOT PROVIDED

Timer, clock, or watch for specimen collection and test procedure.

WARNINGS AND PRECAUTIONS

- This kit is for prescription, *in vitro* diagnostic use only.
- Read all instructions carefully before performing the test. Failure to follow the instructions may result in inaccurate results.
- Do not use the test kit beyond the expiration date printed on the pouch.
- Do not use if any of the test kit contents or packaging is damaged.
- Do not interchange reagent bottle caps.
- Do not interchange external control solution bottle caps.
- Do not interchange or mix components from different kit lots.
- Swabs, tubes, and strips are for single use only. Do not re-use.
- Testing should only be performed using the swabs provided within the kit. Do not touch the swab tip.
- Do not eat, drink or smoke in the area where the specimens and kits are handled.
- Test strips must remain sealed in the pouch until just prior to use. Do not open the pouch until you are ready to perform the test.
- Reagent A and Bare caustic. Avoid contact with eyes, sensitive mucous membranes, cuts, abrasions, etc.

If these reagents contact the skin or eyes, flush with a large volume of water.

- The positive and negative controls contain ProClin™ 300 as a preservative.
- Do not use the kit to evaluate patient samples if either the positive control swab or negative control swab fails to give the expected result.
- Do not read test results before 5 minutes or after 10 minutes. Results read before 5 minutes or after 10 minutes may lead to a false positive, false negative, or invalid result.
- All specimens should be treated as potentially infectious diseases. Wear appropriate personal protection equipment and gloves when handling patient samples and running each test.

KIT STORAGE AND STABILITY

Store as packaged in the original sealed pouch either at room temperature or refrigerated (2-30°C/36-86°F). **DO NOT FREEZE** any of the test kit components (below 0°C/32°F). The test strip is stable through the expiration date printed on the sealed pouch. Do not use the test device or reagents after the expiration date. The test strip must remain in the sealed pouch until use.

SPECIMEN COLLECTION AND PREPARATION

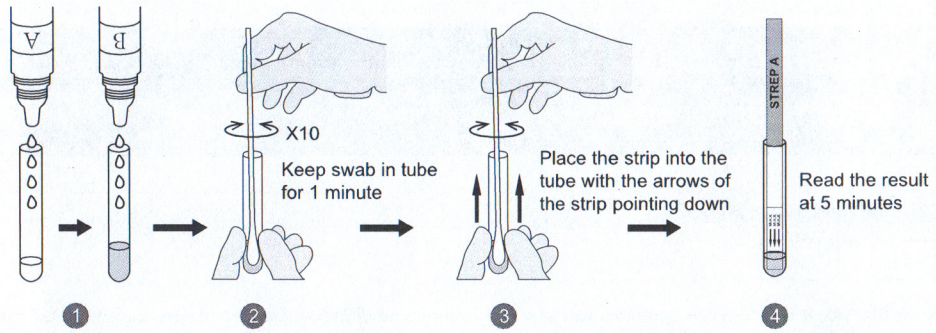
Acceptable specimen type for testing is direct throat swab. Inadequate specimen collection and/or handling may yield inaccurate results.

- Only use the sterile swabs and reagents provided in the kit.
- Collect the throat swab specimen with the sterile swab that is provided in the kit. Swab the posterior pharynx, tonsils and other inflamed areas. Avoid touching the tongue, cheeks and teeth with the swab.[4]
- For best results, throat swabs should be tested immediately after collection. If immediate testing is not possible, a direct throat swab may be stored in a clean, dry plastic tube for 2-4 hours at room temperature (15-30°C/59-86°F) or 24 hours at 2-8°C (36-46°F).

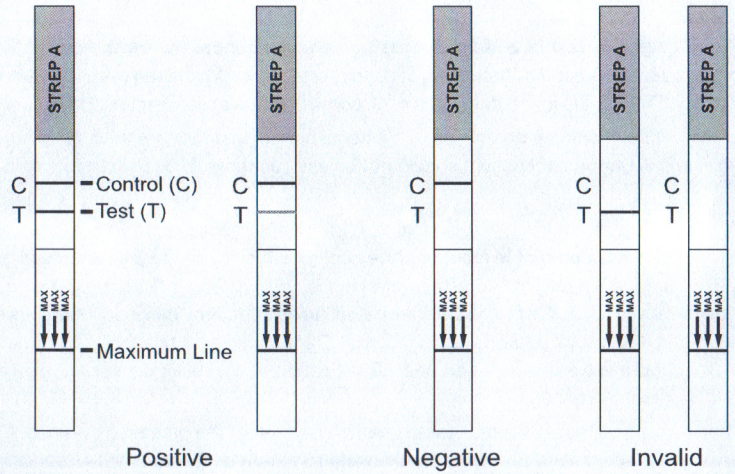
TEST PROCEDURE

The test should be performed at room temperature (15-30°C/59-86°F). Allow the test materials to reach room temperature prior to use. Do not open the Test Strip until you are ready to use.

1. Place an empty extraction tube in the tube holder. Hold the Reagent A bottle vertically over the empty extraction tube and add 4 full drops of Reagent A. Reagent A is light red in color.
2. Hold the Reagent B bottle vertically over the same tube and add 4 full drops to the tube. Reagent B is colorless. Mix the solution by gently swirling the extraction tube.
3. Immediately place the throat swab into the extraction tube. Mix well by rolling the swab in a circular motion at least 10 times. Press the swab tip against the bottom and sides of the tube while rolling. Leave the swab in the tube for 1 minute.
4. Remove the swab while pressing the swab against the side of the tube and squeezing the bottom of the tube as the swab is withdrawn. Discard the swab.
5. Remove the test strip from the foil pouch. Do not touch the bottom of the strip. Place the test strip into the tube with the arrows of the strip pointing down. Start a timer for 5 minutes. Do not handle or move the strip until the test is complete and ready for reading.
6. After 5 minutes, read the tests results visually in the results window, labeled as "C" and "T" on the test strip. Do not read the results before 5 minutes or after 10 minutes.



INTERPRETATION OF RESULTS



Positive Result

If the Control (C) line and the Test (T) line are visible, the test is positive. Any visible faint red or pink test (T) line with a visible control (C) line should be read as positive.

Negative Result

If the Control (C) line is visible, but the Test (T) line is not visible, the test is negative. A negative test result indicates that Strep A was not detected in the sample.

NOTE: Negative results are presumptive and should be confirmed by bacterial culture.

Invalid Result

If a control (C) line is not visible, the test is not valid. Invalid tests should be repeated with a new test.

NOTE: Insufficient specimen volume, incorrect operation procedure, or the use of expired tests are the most likely reasons for control band failure.

LIMITATIONS

- The Rapid Strep A Antigen Test is for the detection of Group A Streptococcal antigen in throat swab specimens only.
- This is a his is a qualitative test. The line intensity is not indicative of the quantity of bacteria in the sample
- This test will only indicate the presence of Group A Streptococcal antigen in the specimen from both viable and non-viable Group A Streptococcus bacterium. This test cannot rule out diseases cause by other bacterial or viral agents.
- Positive and negative predictive values are dependent upon prevalence. The performance was established for the 2019 – 2025 season. Performance may vary depending on the prevalence and the population tested.
- False negative results may occur if bacteria are present at levels below the test's limit of detection.
- False negative results may occur if mutations are present in the regions targeted by the test.
- Test performance has not been evaluated for patients without signs and symptoms of Strep A infection.
- A negative result must be confirmed by culture. A negative result may be obtained if the concentration of the Group A Streptococcal antigen present in the throat swab is not adequate or is below the detectable level of the test.
- Additional follow-up testing using the culture method is required if the result is negative and clinical symptoms persist, or in the event of an acute rheumatic fever (ARF) outbreak.
- The sterile swabs provided with this test must be used for specimen collection. Other swabs have not been validated with this test.
- As with all diagnostic tests, all results must be interpreted together with other clinical information available to the physician.

QUALITY CONTROL

Built-in Procedural Control Features

Internal procedural controls are included in the test. A color line appearing in the control region (C) is an internal positive procedural control. It confirms sufficient specimen volume, adequate membrane wicking and correct procedural technique. If a color band is not visible in the control region (C), the test is invalid.

External Quality Control

In addition to your laboratory's standard quality control procedures, it is recommended that positive and negative external controls be tested at least once per kit lot number and by each new untrained operator. This will verify that the reagents and test strips are working properly, and the operator is able to correctly perform the test procedure.

Procedure for External Quality Control Testing

1. Add 4 full drops of Reagent A and 4 full drops of Reagent B into an extraction tube. Mix the solution by gently swirling the extraction tube.
2. Add 1 full drop of positive or negative control solution into the tube, holding the bottle vertically.
3. Place a clean swab into the extraction tube. Mix well by rolling the swab in a circular motion at least 10 times. Press the swab tip against the bottom and sides of the tube while rolling. Leave the swab in the tube for 1 minute.
4. Remove the swab while pressing the swab against the side of the tube and squeezing the bottom of the tube as the swab is withdrawn. Discard the swab.
5. Continue with Step 5 in the **TEST PROCEDURE** Section. If the controls do not yield the expected results, do not use the test results. Repeat the test or contact Technical Services or Customer Support.

PERFORMANCE CHARACTERISTICS

Analytical Sensitivity: Limit of Detection (LoD)

LoD studies determine the lowest detectable concentration of Strep A at which approximately 95% of all (true positive) replicates test positive. *Streptococcus pyogenes* isolate ATCC 19615 was spiked into negative clinical matrix. Serial dilutions were tested with the Rapid Strep A Antigen Test and the LoD was confirmed by testing 20 replicates.

The limit of detection of the test is 7.2×10^3 CFU/mL.

Clinical Sensitivity and Specificity

Clinical performance of the Rapid Strep A Antigen Test was established in 533 throat samples prospectively collected from symptomatic subjects between July 2019 and January 2025 at three clinical point of care sites. Two throat swab samples were collected from sequentially enrolled subjects presenting with symptoms of pharyngitis using standard collection methods. One swab was tested with the Rapid Strep A Antigen Test and the other swab was sent to a central lab for reference testing. Results obtained from the Rapid Strep A Antigen Test were compared to the clinical reference standard (culture on blood agar). Testing was performed by operators who had no prior experience in the laboratory and were representative of the intended users. Operators used only the QRI to conduct testing.

Of the 533 samples, 114 were found to be positive by culture and 419 were found to be negative. For Rapid Strep A Antigen Test results, the positive percent agreement (PPA) was 96.5% and the negative percent agreement (NPA) was 99.5% (see tables).

Table 1 Clinical Performance: Rapid Strep A Antigen Test vs. Culture

Rapid Strep A Antigen Test Results	Reference Culture Results	
	Positive	Negative
Positive	110	2
Negative	4	417
Total	114	419

Sensitivity: 96.5% (95% C.I. = 91.3 – 98.6%)
Specificity: 99.5% (95% C.I. = 98.3 – 99.9%)

Table 2. Clinical Performance Stratified by Age

Age	Sensitivity	95% CI	Specificity	95% CI
0 - 5	100% (8/8)	67.6% - 100%	100% (28/28)	87.9% - 100.0%
>5 – 21	94.3% (49/52)	84.4% - 98.0%	99.1% (119/120)	95.4% - 99.9%
>21	98.2% (53/54)	90.2% - 99.7%	99.6% (270/271)	97.9% - 99.9%
All	96.5% (110/114)	91.3% - 98.6%	99.5% (417/419)	98.3% - 99.9%

Cross-Reactivity

Organisms likely to be found in the respiratory tract were tested for cross-reactivity and microbial interference with the Rapid Strep A Antigen Test in triplicate. The tested concentration of each microorganism is documented in the following table. No cross-reactivity and no microbial interference were found for each microorganism at the listed concentration in table 3.

Table 3. Results of Microorganisms Tested for Cross-Reactivity and Microbial Interference

Microorganism	Concentration Tested	Microorganism	Concentration Tested
Arcanobacterium haemolyticum	2.6×10^8 CFU/mL	*Staphylococcus epidermidis*	2.1×10^8 CFU/mL
Bordetella pertussis	7.5×10^8 CFU/mL	*Staphylococcus marcescens*	1.5×10^8 CFU/mL
Candida albicans	9.5×10^8 CFU/mL	*Staphylococcus haemolyticus*	1.58×10^8 CFU/mL
Corynebacterium diphtheria	5.37×10^8 CFU/mL	*Streptococcus agalactiae* (Group B)	7.9×10^7 CFU/mL
Enterococcus faecalis	2.3×10^8 CFU/mL	*Streptococcus dysgalactiae* (Group C)	1.43×10^5CFU/mL
Enterococcus faecium	4.4×10^8 CFU/mL	*Streptococcus sp. (bovis II)* Group D	5.6×10^8 CFU/mL
Enterovirus (VR-28 Human Coxsackievirus)	1.6×10^8 $TCID_{50}$/mL	*Streptococcus sp.* Strain H60R (Group F)	1×10^6 CFU/mL
Escherichia coli	1.1×10^8 CFU/mL	*Streptococcus anginosus* (Group G)	4.2×10^7 CFU/mL
Fusobacterium necrophorum	7.3×10^8 CFU/mL	*Streptococcus pneumoniae*	4.2×10^6 CFU/mL
Haemophilus parahaemolyticus	1.3×10^8 CFU/mL	*Streptococcus salivarius*	8.7×10^8 CFU/mL
Haemophilus influenzae	4.5×10^8 CFU/mL	*Streptococcus mitis*	5.9×10^8 CFU/mL
Haemophilus parainfluenzae	1.6×10^8 CFU/mL	*Streptococcus mutans*	4.7×10^8 CFU/mL
Human metapneumovirus (HMPV-27 A2)	3.55×10^5 $TCID_{50}$/mL	*Streptococcus oralis*	6.4×10^8 CFU/mL
Human coronavirus OC43	1.7×10^5 $TCID_{50}$/mL	*Streptococcus sanguis*	1.5×10^8 CFU/mL
Klebsiella pneumoniae	3.1×10^8 CFU/mL	*Yersinia enterocolitica*	2.0×10^8 CFU/mL
Legionella pneumophila	1×10^4 bacteria/mL	*Adenovirus Type I*	3.09×10^8 $TCID_{50}$/mL
Lactobacillus sp. (Lactobacillus casei)	6.5×10^8 CFU/mL	*Adenovirus Type II*	3.9×10^7 $TCID_{50}$/mL
Mycobacterium tuberculosis	1×10^3 bacteria/mL	*Adenovirus 3*	1.5×10^8 $TCID_{50}$/mL
Moraxella lacunata	1.95×10^8 CFU/mL	*Adenovirus 7*	2.8×10^6 $TCID_{50}$/mL
Moraxella (Branhamella) catarrhalis	4.8×10^8 CFU/mL	*Cytomegalovirus*	1.6×10^5 $TCID_{50}$/mL
Mycobacterium tuberculosis (avirulent strain)	2.3×10^8 CFU/mL	*Epstein Barr Virus*	7.85×10^7 copies/mL
Neisseria gonorrhoeae	3.8×10^8 CFU/mL	*HSV Type 1 MacIntyre strain*	1.6×10^5 $TCID_{50}$/mL
Neisseria lactamica	1.19×10^8 CFU/mL	*Human parainfluenza Type 1*	1.6×10^5 $TCID_{50}$/mL
Neisseria meningitides	7.5×10^8 CFU/mL	*Human parainfluenza Type 2*	1.6×10^5 $TCID_{50}$/mL
Neisseria mucosa	3.25×10^8 CFU/mL	*Human parainfluenza Type 3*	1.6×10^5 $TCID_{50}$/mL
Neisseria sicca	8.5×10^8 CFU/mL	*Human rhinovirus 26*	5×10^6 $TCID_{50}$/mL
Neisseria subflava	3.27×10^8 CFU/mL	*Measles Virus*	8.9×10^5 $TCID_{50}$/mL
Proteus vulgaris	2.9×10^8 CFU/mL	*Mumps virus*	1.38×10^7 $TCID_{50}$/mL
Pseudomonas aeruginosa	5.1×10^8 CFU/mL	*Respiratory syncytial virus Type A*	5.5×10^7 PFU/mL
Serratia marcescens	2.1×10^8 CFU/mL	*Respiratory syncytial virus Type B*	2.8×10^5 $TCID_{50}$/mL
Staphylococcus aureus	3.2×10^8 CFU/mL		

Interference Substances

The following substances, naturally present in respiratory specimens or that may be artificially introduced into the upper respiratory tract, were evaluated with the Rapid Strep A Antigen Test at the concentrations listed in table 4 and were found not to affect test performance.

Table 4. Results of Potential Interfering Substances

Interfering Substance	Concentration Tested	Interfering Substance	Concentration Tested
Endogenous			
Blood (human)	20%(vol/vol)	Mucin	1 mg/mL
OTC Mouthwashes			
Colgate Total Pro-Shield, Spearmint	20%(vol/vol)	Crest Pro-Health Clean Mint	20%(vol/vol)
Crest Pro Health Multi Protection Clean Mint	20%(vol/vol)	Listerine Antiseptic Cool Mint	20%(vol/vol)
OTC Lozenges			
Cepacol Extra Strength Sore Throat & Cough Drop Lozenges, Cherry	5 mg/mL	Sucrets Sore Throat Lozenges Cherry	5 mg/mL
Halls Mentho-Lyptus Drops Cherry	5 mg/mL	Sucrets Sore Throat & Cough Lozenges, Honey Lemon,	5 mg/mL
Halls Cough Suppressant Cherry Triple Soothing Action	5 mg/mL		
OTC Throat Sprays			
Cepacol Dual Relief	20%(vol/vol)	Chloraseptic Max	20%(vol/vol)
OTC Cough Syrups			
Basic Care Tussin DM, Cough Suppressant & Expectorant	10%(vol/vol)	Robitussin Nighttime Cough	10%(vol/vol)
Children's Dimetapp Cold & Flu	10%(vol/vol)	Robitussin (Guaifenesin Syrup)	10%(vol/vol)
Children's Dimetapp Cold & Cough	10%(vol/vol)	Tylenol Cough and Sore Throat	10%(vol/vol)
Active Ingredients			
Acetaminophen (Tylenol)	5 mg/mL	Doxylamine Succinate	5 mg/mL
Brompheniramine Maleate	5 mg/mL	Guaifenesin (Guaiacol Glyceryl)	5 mg/mL
Chlorpheniramine Maleate	5 mg/mL	Ibuprofen (Advil)	5 mg/mL
Dextromethorphan HBr	5 mg/mL	Phenylephrine HCl	5 mg/mL
Diphenhydramine HCl	5 mg/mL		

Inclusivity

Inclusivity testing was performed with currently available commercial stock strains of Strep A to determine if the Rapid Strep A Antigen Test can detect target analytes across a variety of strains at or near the LoD. All data showed inclusivity for the five different strains tested (see table 5).

Table 5. Results of Inclusivity Testing

Source	Detection Concentration	No. Positive/Tested	Positive Agreement (%)
ATCC 12344	1.44×10^5CFU/mL	9/9	100%
ZeptoMetrix 0801512	2.95×10^5CFU/mL	21/21	100%
ATCC 700294	2.375×10^5 CFU/mL	21/21	100%
ATCC 14289	3.75×10^5CFU/mL	21/21	100%
ATCC 51339	3.305×10^5 CFU/mL	21/21	100%

Precision/Reproducibility

A precision study was performed to evaluate the precision of the Healgen® Rapid Strep A Antigen Test. The study was performed at three external, CLIA-waived testing sites consisting of three replicates each of positive (prepared at 2.5x LoD), low positive (prepared at 1 x LoD), low negative samples (prepared at 0.5x LoD), and true negative samples (Diluent only) tested by six (6) untrained operators, two (2) runs per day over 5 days, i.e., 1, replicates x 6 operators x 2 runs per day x 3 lots x 5 days = 180 replicates per concentration and a total of 720 data points collected. Three (3) test lots were used in this study, so lot-to-lot variability was also assessed. Fifty (50) µL of the prepared sample were applied to kit swabs, shipped and stored frozen at -20°C/-4°F until testing. The results were >90% agreement between expected and read result within run, by lot, by operator, by day, between sites and overall.

Table 6. Results of Multisite Precision Study (Reproducibility)

Site	True Negative		Low Negative		Weak Positive		Positive	
	Correct Reads/Total	PPA	Correct Reads/Total	PPA	Correct Reads/Total	PPA	Correct Reads/Total	PPA
1	60/60	100%	29/60	48.3%	59/60	98.3%	60/60	100%
2	60/60	100%	26/60	43.3%	56/60	93.3%	60/60	100%
3	60/60	100%	25/60	41.7%	57/60	95.0%	60/60	100%
Total	**180/180**	**100%**	**80/180**	44.4%	**172/180**	**95.5%**	**180/180**	**100%**

REFERENCES

1. Webb, KH. Does Culture Confirmation of High-sensitivity Rapid Streptococcal Tests Make Sense? A Medic Decision Analysis. Pediatrics (Feb 1998), 101:2, 2.
2. Bisno AL, Gerber MA, Gwaltney JM, Kaplan EL, Schwartz RH. Diagnosis and Management of Group A Streptococcal Pharyngitis. Clinical Infectious Diseases (1997), 25: 574-83.
3. Needham CA, McPherson KA, Webb KH. Streptococcal Pharyngitis: Impact of a High-sensitivity Antigen Test on Physician Outcome. Journal of Clinical Microbiology (Dec 1998), 36: 3468-3473.
4. Shea, Y.R., Specimen Collection and Transport, Clinical Microbiology Procedures Handbook, Isenberg, H.D., American Society of Microbiology, Washington D.C., 1.1.1-1.1.30, 1992.
5. Nussinovitch, M, Finkelstein Y, Amir J, Varsano, I. Group A beta-hemolytic streptococcal pharyngitis in preschool children aged 3 months to 5 years. Clinical Pediatrics (June 1999), 38: 357-360.
6. Woods WA, Carter CT, Stack M, Connors Jr AF, Schlager TA. Group A Streptococcal Pharyngitis in Adults 30 to 65 years of age. Southern Medical Journal (May 1999), 491-492.

INDEX OF SYMBOLS

	Do not reuse		See Instruction for Use		Expiration Date
Σ	Tests per Kit	86 °F 30 °C / 36 °F 2 °C	Store Between 2-30°C (36-86°F)		Keep Dry
LOT	Batch Number	REF	Catalog#		Keep Away from Sunlight
UDI	Unique Device Identifier	IVD	For *in vitro* diagnostic use only		Manufacturer
℞ Only	Prescription Use Only		Warning		

ASSISTANCE

If you have any questions regarding the use of this product, please call our Technical Support Number 1-866-982-3818 (8:30 a.m. to 5 p.m. CT).

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