

LABORATORY PROCEDURE

Qtest™ Strep

I. INTENDED USE

Qtest™ Strep test is a liposome immunoassay test for the rapid, qualitative detection of group A streptococcal antigen directly from throat swabs.

II. SUMMARY AND EXPLANATION

Group A streptococci are organisms that typically cause illness such as tonsillitis, pharyngitis, and scarlet fever. If untreated, these infections can lead to complications such as rheumatic fever. The Qtest Strep test is performed directly on throat-swab extracted antigens and the test is completed in approximately two minutes after collection of the specimen. Antibody-coated liposomes are used to detect the extracted antigen.

III. PRINCIPLES OF THE PROCEDURE

To perform the test, a throat swab specimen is collected, then streptococcal antigens are extracted from the specimen with Reagents A, B, and C. The resulting solution of extracted antigen is added to the Qtest™ device. The Qtest device contains a membrane that has antibodies to group A *Streptococcus* coated on its surface. As the specimen passes through the membrane, the extracted antigen attaches to the antibodies on the surface.

Reagents 1 and 2 are then added to the Qtest device. When Reagent 1 is added to the Qtest device, antibody-coated liposomes containing pink dye attach to the streptococcal antigen previously bound to the membrane. If group A streptococcal antigen is present in the specimen, a pink triangle will appear, indicating a positive result. In the absence of antigen, a pink negative sign (-) will appear, indicating a negative result.

The Qtest Strep test contains built-in quality control features that include both positive and negative internal procedural controls for the Qtest device and color-coded reagents for the extraction step of the assay. The appearance of a pink negative sign (-) when a specimen is negative is the internal positive procedural control and indicates that Reagents 1 and 2 were working properly and added in the correct sequence. The membrane background area is the internal negative procedural control for the Qtest device and should be white-to-light pink in color. If the test is not conducted correctly or the reagents are not performing properly, no distinct test result will appear.

IV. REAGENTS AND MATERIALS SUPPLIED

Reagent A: 2M sodium nitrite. (7.0 mL sufficient for 20 or 40 tests or 14.0 mL sufficient for 80 tests.)

Reagent B: 2N acetic acid. (7.0 mL sufficient for 20 or 40 tests or 14.0 mL sufficient for 80 tests.)

Reagent C: Buffered 0.66N sodium hydroxide. (8.0 mL sufficient for 20 tests or 12.0 mL sufficient for 40 tests or 24.0 mL sufficient for 80 tests.)

Reagent 1: Anti-group A *Streptococcus* (rabbit antibody) coated liposomes, with 0.2% sodium azide (preservative). (4.5 mL sufficient for 20 tests or 9.0 mL sufficient for 40 tests or 18.0 mL sufficient for 80 tests.)

Reagent 2: Buffered wash solution. (5.5 mL sufficient for 20 or 40 tests or 11.0 mL sufficient for 80 tests.)

Qtest Strep Devices: Membrane coated with Anti-group A *Streptococcus* (rabbit antibody) and group A streptococcal antigen, (one per test).

Sterile Throat Swabs: One per test.

DispensTube™ Specimen Holder: One per test.

DispensTube™ Tips: One per test.

Positive Control: Heat inactivated group A *Streptococcus* in solution with 0.2% sodium azide (preservative). (2.0 mL.)

Negative Control: Heat inactivated *Streptococcus*, not group A, in solution with 0.2% sodium azide (preservative). (2.0 mL.)

Precautions: For *in vitro* Diagnostic Use

Reagents: Do not use beyond the expiration date marked on the kit. **Do not mix reagents from different kit lot numbers.** Do not remove the Qtest device from the pouch until just prior to use.

To assure proper drop delivery, reagent bottles must be held vertically while dispensing free-falling drops.

Observe established precautions against microbiological hazards in specimen handling, disposal and throughout all procedures. Use good laboratory procedures when collecting and handling specimens and controls. Wearing appropriate laboratory personal protective equipment (lab coats, gloves, etc.) is recommended. Discard used materials into a receptacle approved for biohazardous waste.

Warning: Reagent A and Positive Control contain combustible and toxic materials. Contact with combustible material may cause fire. Toxic if swallowed. Keep away from combustible material. When using do not eat or drink. Wear suitable protective clothing, gloves, and eye/face protection. In case of accident or if you feel unwell, seek medical advice immediately.

Reagent B contains acid; Reagent C contains strong alkali. If these reagents contact the skin or eyes, flush with large volumes of water.

Reagent 1 and the Negative Control contain sodium azide which is very toxic by inhalation, in contact with skin and if swallowed. After contact with skin, wash immediately with plenty of water. Sodium azide may react with lead and copper plumbing to form highly explosive metal azides. If disposed into a drain, flush with a large volume of water to prevent azide build-up.

Swabs: Use the swabs supplied in this kit to obtain throat specimens.

DispensTube Tip: Must contain white filter materials to ensure proper test performance.

Storage: Refrigerate reagents at 36–46°F (2–8°C). DO NOT FREEZE. Allow reagents to reach room temperature before use. See room temperature storage option on procedure card.

Extractions: Optimal results are obtained when the throat swab is squeezed during the extraction process.

Interpretation: Interpretation of test results is best made immediately after Reagent 2 is absorbed, as color intensity may change over time.

V. SPECIMEN HANDLING

Collect a throat specimen by standard recommended procedures using the swabs supplied in this kit. Consult standard reference procedures, such as the collection method described by Facklam.¹

Processing samples for Qtest Strep testing is recommended as soon as possible after collection. Swabs supplied with the kit provide optimal results. Rayon or Dacron™ fiber swabs on a plastic shaft, without transport media, may be used. Transport devices containing charcoal, agar, wooden shafts or calcium alginate material should not be used for rapid antigen tests. If culture is desired, streak the swab onto a culture plate using standard laboratory procedure before conducting the Qtest Strep test.

VI. TEST PROCEDURE

Materials Provided: All materials listed under “Reagents and Materials Supplied.”

Materials Required But Not Provided: Timer

Quality Control: Liquid Positive and Negative Controls are provided in order to monitor the integrity of the reagents and assure that the test procedure has been performed correctly. Prior to using a new kit or lot number of Qtest Strep, it is recommended that the Positive and Negative Controls be tested in accordance with the laboratory’s established Quality Control procedures. Upon observing the expected results, the kit is ready for use with patient samples.

Sample Extraction:

- A. Place a DispensTube sample tube and a Qtest device in the designated areas of the Workstation. Add 3 drops Reagent A into DispensTube sample tube.
- B. Add 3 drops Reagent B into the DispensTube sample tube. The solution should turn gold. Immediately place the throat swab specimen in the DispensTube sample tube. **Mix by rolling the swab against the side of the sample tube, squeezing the swab through the tube several times during the extraction process so that the liquid is expressed from the swab and reabsorbed.** Let stand for one minute but not longer than 3 minutes.

Caution: Assay performance depends upon thorough antigen extraction.

- C. Holding the swab shaft to the side, add 5 drops Reagent C directly into the DispensTube sample tube. Mix the solution by squeezing the tube and moving the swab. The color of the solution should change to red. If the solution remains purple continue squeezing the swab. Alternatively, an additional drop of Reagent B may be added. Solution should then turn red.

Remove the liquid from the swab by rolling the swab against the wall of the DispensTube sample tube and squeezing the flexible sides. Discard the swab.

Insert the DispensTube tip into the DispensTube sample tube. The red solution may be held for up to one hour at room temperature before testing without affecting the results.

Sample Testing:

Invert tube; allow the fluid to be absorbed into the filter material. Dispense entire contents of the DispensTube sample tube into the Qtest well. Allow the fluid to be absorbed. Proceed immediately to Step 1.

1. Place 3 drops of Reagent 1 into the Qtest well. Allow the fluid to be absorbed. Proceed immediately to Step 2.
2. Place 3 drops Reagent 2 into the Qtest well. Allow the fluid to be absorbed. Immediately interpret the test results.

VII. RESULTS

Interpretation of test results is best made immediately after Reagent 2 is absorbed, as color intensity may change over time.

Positive Test – A pink triangle appears in the Qtest well. The background area should be white-to-light pink.

Note: The negative sign may be visible; however, the presence of any shade of a pink triangle indicates a positive test.

Negative Test: - A pink negative sign (-) appears in the Qtest well. The background area should be white-to-light pink and no triangle should be visible.

Invalid Results: No distinct test result will appear if reagents are added incorrectly or are not performing properly. Retest the patient or contact ***BD Diagnostic Systems Technical Services, toll free (800)-638-8663, selection 2.***

VIII. QUALITY CONTROL

“Quality control requirements must be performed in accordance with applicable local, state and/or federal regulations or accreditation requirements and your laboratory’s standard Quality Control procedures. It is recommended that the user refer to pertinent NCCLS guidance and CLIA regulations for appropriate Quality Control practices.”

Each Qtest Strep device contains built-in procedural controls that are reagent-activated. The appearance of a pink negative sign (-) and a white-to-light pink background area in the Qtest test well validate the immunological integrity of the test. It also provides assurance that Reagent 1 and Reagent 2 were added in the correct sequence. Note: A pink negative sign (-) may not be apparent with a strong positive sample reaction.

A procedural control is also provided for the specimen extraction step of the test. A sequence of color changes occurs with each step of the extraction process: red to gold, and then back to red. This assures extraction reagent integrity and proper extraction procedure.

Positive and Negative Liquid Controls are supplied with each kit. Liquid controls may be used in place of a patient sample in order to monitor the integrity of the reagents/device, assure that the test procedure has been performed correctly, and/or to demonstrate a positive or negative reaction. *After shaking or vortexing thoroughly*, add 2 drops of Control into a DispensTube sample tube containing 3 drops Reagent A and 3 drops Reagent B. Add dry swab. Mix. After one minute, but not longer than 3 minutes, add 5 drops Reagent C. Mix well, discard swab and insert DispensTube Tip. Dispense entire contents of DispensTube device into the Qtest device. Proceed with Step 1 “Sample Testing”.

If expected control results are not obtained, do not report patient results. Contact your local BD representative or Technical Service for assistance.

IX. LIMITATIONS OF THE PROCEDURE

Respiratory infections, including pharyngitis, can be caused by streptococci from serogroups other than group A as well as other pathogens. QTest Strep test will not differentiate asymptomatic carriers of group A *Streptococcus* from those exhibiting streptococcal infection. In rare cases, test specimens heavily colonized with *Staphylococcus aureus* can yield false positive results. If clinical signs and symptoms are not consistent with clinical test results, follow-up culture and grouping procedures should be performed.

Use of liquid transport systems may result in reduced intensity of test results relative to testing with the kit swab or a comparable dry swab.

X. PERFORMANCE CHARACTERISTICS

The ability of the Qtest Strep test to identify group A *Streptococcus* from throat swab specimens was determined by correlation with conventional culture techniques during internal trials. One hundred eighty eight (188) throat swab specimens were collected on swabs from patients with pharyngitis. Swabs in transport medium were shipped overnight from 15 clinical sites to an

internal laboratory for processing. Immediately after arrival, each swab was inoculated onto a sheep blood agar plate. Swabs were either refrigerated or frozen while inoculated plates were incubated at 37° C for 48 hours. Beta-hemolytic colonies were identified as group A streptococci by a serologic method. After culture results were determined, stored swabs were selected on the basis of culture classification² result and the Qtest Strep test was performed.

Swabs were selected to yield a population with a prevalence of infection between 25-30%, dominated by 3+ and 4+ culture classification (as is usually seen during strep season).

Culture versus Qtest Strep				Specimen Mix in Positive Sample Population	
		Culture		Culture Classification	% in Sample Pop.
		+	-		
QTest Strep	+	49	5	1+	2
	-	3	131	2+	11
				3+	56
				4+	31

Sensitivity 94.2%
Specificity 96.3%
Overall Accuracy 95.7%

XI. AVAILABILITY

Catalog No.	Description
494775	20 Test Kit
494776	40 Test Kit
494780	80 Test Kit

XII. REFERENCES

1. Facklam R.R.: U.S. Dept. of Health and Human Services, PHS CDC, Pub.No. CDC 77 – 13.
2. Data on File, BD Diagnostic Systems.

BIBLIOGRAPHY

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Youmans, G.P., Paterson, P.Y., Sommers, H.M.: *The Biological and Clinical Basis of Infectious Disease*, p. 172-181 (1975).

Moody, M.D., Siegle, A.C., Pittman, B., Wilnter, C.C.: *Am. J. Public Health*, 53:1083-1092 (1963).

Lue, Y.A. Howit, I.P., Ellmer, PD.: *J. Clin. Microbiol.*, 8:326-328 (1970).

Rosner, R.: *Clin. Microbiol.*, 6:23-26 (1977).

XIII. TECHNICAL INFORMATION:

In the United States, telephone Technical Services, toll free (800)-638-8663, selection 2.

Approved by:

Supervisor: _____

Date: _____

Director: _____

Date: _____

Effective Date: _____

Reviewed by: _____

Date: _____

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