STREPTOCOCCUS A CASSETTE
014D300
Flocked swabs

Rapid test for the qualitative detection of Strep A antigen in throat swab specimens
For professional in vitro diagnostic use only

INTENDED USE
The ulti med Streptococcus A Cassette is a rapid chromatographic immunoassay for the qualitative, presumptive detection of Strep A antigen from throat swab specimens to aid in the diagnosis of Group A Streptococcal infection.

SUMMARY
Streptococcus pyogenes is non-motile gram-positive cocci, which contains the Lancefield group A antigen that can cause serious infections such as pharyngitis, respiratory infection, impetigo, endocarditis, meningitis, puerperal sepsis, and arthritis.[1] Left untreated, these infections can lead to serious complications, including rheumatic fever and peritonsillar abscess.[2] 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.[3]
Rapid diagnosis and early antibiotic therapy of Group A Streptococcal infection appear to be the best means of preventing medical complications and reducing the spread of the disease.[4]
The ulti med Streptococcus A Cassette is a rapid test to qualitatively detect the presence of Strep A antigen in throat swab specimens, providing results within 5 minutes. The test utilizes antibodies specific for whole cell Lancefield Group A Streptococcus to selectively detect Strep A antigen in a throat swab specimen.

PRINCIPLE
The ulti med Streptococcus A Cassette is a qualitative, lateral flow immunoassay for the detection of Group A Streptococcus antigens in a throat swab. In this test, Anti-Strep A antibodies are immobilized on the test region of the membrane. During the test, the specimen reacts with polyclonal anti-Strep A antibodies conjugated to coloured particles and precoated onto the sample pad of the test. The mixture then migrates through the membrane by capillary action and interacts with reagents on the membrane. If there is sufficient Strep A antigen in the specimen, a coloured line will form at the test region of the membrane. The presence of this coloured line indicates a positive result, while its absence indicates a negative result. The appearance of a coloured line at the control region serves as a procedural control, indicating that proper volume of specimen has been added and membrane wicking has occurred.

REAGENTS
The test utilizes polyclonal antibodies to specifically identify Strep A antigen in throat swab specimens.

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PRECAUTIONS

- For professional in vitro diagnostic use only.
- Do not use beyond the expiration date indicated on the package.
- Do not use when protective foil is damaged.
- The test cassette should remain in the sealed pouch until use.
- Do not eat, drink or smoke in the area where the specimens and kits are handled.
- Wear protective clothing such as laboratory coats, disposable gloves and eye protection when specimens are assayed.
- Do not interchange or mix reagents from different lots.
- Read the entire procedure carefully prior to testing.
- This kit contains products of animal origin. Certified knowledge of the origin and/or sanitary state of the animals does not completely guarantee the absence of transmissible pathogenic agents. It is therefore recommended that these products be treated as potentially infectious, and handled by observing usual safety precautions (e.g., do not ingest or inhale).
- Humidity and temperature can adversely affect results.
- The used testing materials should be discarded according to federal state and local regulations.
- The positive control contains sodium azide (NaN_3) as a preservative
- Avoid cross-contamination of specimens by using a new extraction tube for each specimen obtained.
- Do not moisten nitrocellulose membrane with urine samples.
- Reagents B contains an acidic solution. If the solutions contact the eye or mucous membranes, flush with large volumes of water.
- Do not reuse tests.
- Do not interchange reagent bottle caps.
- Handle all specimens as if they contain infectious agents. Observe established precautions against microbiological hazards throughout the procedure and follow the standard procedures for proper disposal of specimens.

STORAGE AND STABILITY
The ulti med Streptococcus A Cassette can be stored at room temperature or refrigerated (2-30°C). The test cassette must remain in the sealed pouch until use. The test cassette and the reagents are stable through the expiration date printed on the box. Care should be taken to protect components in this kit from contamination. Do not use if there is evidence of microbial contamination or precipitation. Biological contamination of dispensing equipment, containers or reagents can lead to false results.

- Do not freeze.
- Do not use beyond the expiration date.

SPECIMEN COLLECTION AND PREPARATION
Only use reagents and sterile swabs provided in the ulti med Streptococcus A Cassette. Collect the throat swab specimen with the sterile swab that is provided in the kit. Collect throat swab specimens by standard clinical methods. Swab the posterior pharynx, tonsils and other inflamed areas. Avoid touching the tongue, cheeks and teeth with the swab.[5] The test should be performed immediately after the specimen have been collected. Swab specimens may be stored in a sterile, dry plastic tube for up to 4 hours at room temperature or 24 hours at 2-8°C.

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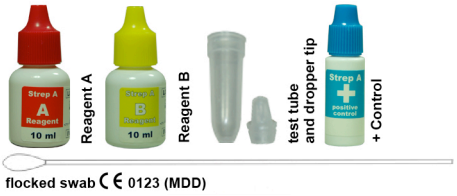
MATERIALS PROVIDED

- Test Cassettes
- Test tubes and dropper tips
- Package insert
- Reagent A (1 M Sodium Nitrite)
- Reagent B (0.4 M Acetic Acid)
- Positive control (Non-viable Strep A; <0.1% NaN_3)
- Workstation
- flocked swabs
  (includes nylon fibers, swab is sterile packaged)
  According to Directive 2007/47/EC



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MATERIALS REQUIRED BUT NOT PROVIDED:

- Timer
- negative control

SAFETY INFORMATION REAGENTS

Reagent A (1 M Sodium Nitrite)		**H272:** May intensify fire; oxidizer **H301:** Toxic if swallowed **H400:** Very Toxic to aquatic organisms **P273:** Avoid release to the environment **P309:** IF exposed or you feel unwell: **P310:** Immediately call a POISON CENTER or doctor/physician For more detailed information read the MSDS (Material Safety Data Sheet) carefully!
Reagent B (0.4 M Acetic Acid)		For more detailed information read the MSDS carefully!

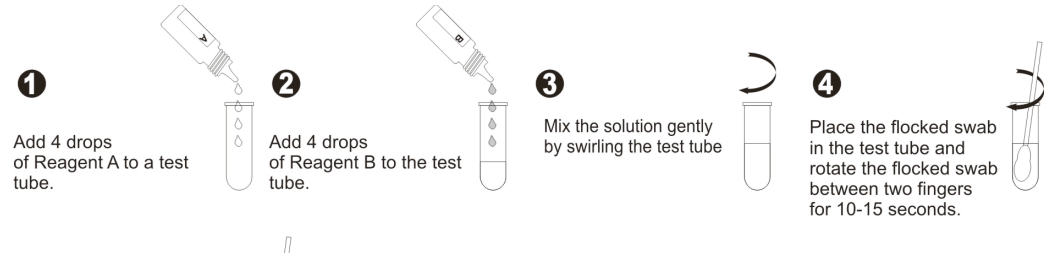
DIRECTIONS FOR USE

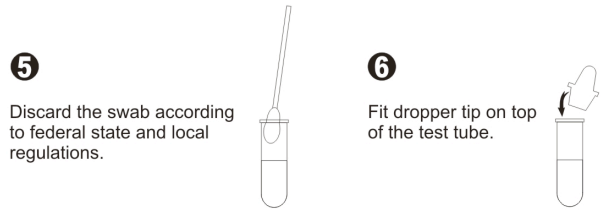
Allow the test cassette, reagents, and/or controls to reach room temperature (15-30°C) prior to testing. Remove the test cassette from the sealed foil pouch and use it as soon as possible. Best results will be obtained if the test is performed immediately after opening the foil pouch.

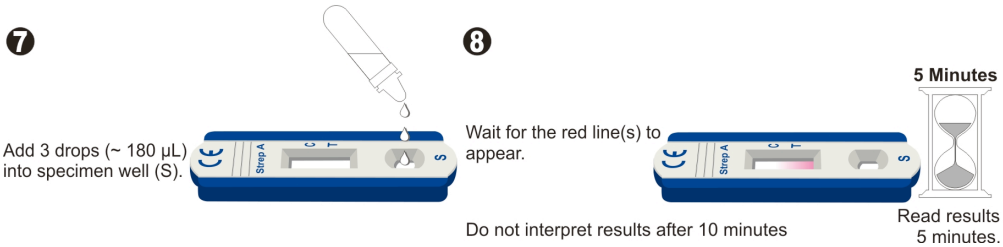
Note:

1. Different volume for the same number of drops bases on the different physical properties (surface tension)
2. Avoid trapping air bubbles in the specimen well (S)
3. Do not add any solutions to the test region
4. Very mucosal specimens can delay the start of chromatography and possibly reduce the speed.

TEST PROZEDURE







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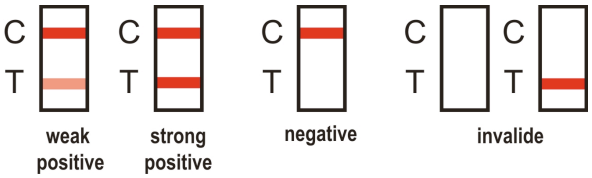
Interpretation of Results



POSITIVE:* Two distinct red lines appear. One line should be in the control region (C) and another line should be in the test region (T). A positive result indicates that Strep A was detected in the sample.
* **NOTE**: The intensity of the red colour in the test line region (T) will vary depending on the concentration of Strep A present in the sample. Therefore, any shade of red in the test region (T) should be considered positive.

NEGATIVE: One red line appears in the control region (C). No apparent red or pink line appears in the test region (T). A negative result indicates that Strep A antigen is not present in the sample, or is present below the detectable level of the test. The patient's sample should be cultured to confirm the absence of Strep A infection. If clinical symptoms are not consistent with results, obtain another sample for culture.

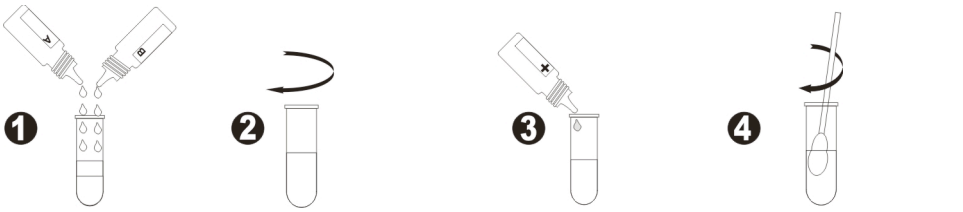
I**NVALID**: Control line fails to appear. Insufficient sample volume, incorrect procedural techniques are the most likely reasons for control line failure. Review the procedure and repeat the test with a new test cassette. If the problem persists, discontinue using the test kit immediately and contact distributor / manufacturer.

QUALITY CONTROL

Internal Quality Control
During quality control, the procedures are performed exactly in compliance with the above-mentioned details in "Sample preparation", "Testing" as well as in the "Test evaluation" section, however, with negative sample material. A red line that appears in the control area (C) confirms that sufficient amounts of sample have been used and that the test has been carried out correctly.

External quality control
In addition to your laboratory's standard quality control procedures, it is recommended that a positive external control be tested at least once within each kit and by each operator performing testing within a kit. This will verify that the reagents and test cassettes are working properly and the operator is able to correctly perform the test procedure. An external positive control containing heat-killed Group A Streptococcus is supplied in the kit.

Procedure for External Quality Control Testing



1	2	3	4
Add 4 drops of Reagent A and B to a test tube.	Mix the solution gently by swirling the test tube.	Add 1 drop of positive control to the test tube.	Place a clean sterile swab in the test tube and proceed as described in TEST PROCEDURE.

If controls do not yield expected results, do not use the test. Repeat the test or contact your distributor.

LIMITATIONS

1. The ulti med Streptococcus A Cassette is for professional in vitro diagnostic use only. The test should be used for the detection of Strep A antigen in throat swab specimens only. Neither the quantitative value nor the rate of increase in Strep A antigen concentration can be determined by this qualitative test.
2. The accuracy of the test depends on the quality of the swab specimen. False negatives may result from improper specimen collection or storage. A negative result may also be obtained from patients at the onset of the disease due to low antigen concentration.
3. The test does not differentiate asymptomatic carriers of Group A Streptococcus from those with symptomatic infection. If clinical signs and symptoms are not consistent with laboratory test results, a follow-up throat culture is recommended.
4. 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, a follow-up culture and grouping procedure should be performed.
5. Respiratory infections, including pharyngitis, can be caused by streptococci from serogroups other than Group A, as well as other pathogens.
6. As with all diagnostic tests, a definitive clinical diagnosis should not be based on the results of a single test, but should only be made by the physician after all clinical and laboratory findings have been evaluated.
7. This test will only indicate the presence of Strep A antigen in the specimen from both viable and non-viable Group A Streptococcus bacteria.
8. A negative result obtained from this kit must be confirmed by culture. A negative result may be obtained if the concentration of the Strep A antigen present in the throat swab is not adequate or is below the detectable level of the test.
9. Excess blood or mucus on the swab specimen may interfere with test performance and may yield a false positive result. Avoid touching the tongue, cheeks, and teeth and any bleeding areas of the mouth with the swab when collecting specimens.

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EXPECTED VALUES
Approximately 15% of pharyngitis in children ages 3 months to 5 years is caused by Group A beta-hemolytic Streptococcus.[6] In school-aged children and adults, the incidence of Strep throat infection is about 40%.[7] This disease usually occurs in the winter and early spring in temperate climates.[3]

PERFORMANCE CHARACTERISTICS
The following performance characteristics were conducted with Streptococcus A test device with polyester swabs.
Measurements with the flocked swabs showed that they didn`t show any interference with the Streptococcus A test kit.
Furthermore measurements showed that usage of flocked swabs increases the limit of detection of the Rapid Streptococcus A testing kit when compared to the polyester swabs.

Sensitivity and Specificity
A total of 244 throat swabs were collected from patients exhibiting symptoms of pharyngitis. Each swab was rolled onto a sheep blood agar plate, and then tested by ulti med Strep A Rapid Test Device (Throat Swab). The plates were further streaked for isolation, and then incubated at 37 °C with 5 – 10 % CO_2 and a Bacitracin disk for 18 – 24 hours. The negative culture plates were incubated for an additional 18 – 24 hours. Possible GAS colonies were subcultured and confirmed with a commercially available latex agglutination grouping kit.

Of the 244 total specimens, 160 were found to be negative by culture and 84 were found to be positive by culture. These swabs were tested using the ulti med Strep A Rapid Test Device (Throat Swab).

		Culture	
		+	-
Strep A Rapid Test	+	82	4
	-	2	156

Sensitivity: 97,6% (82 + 2)
Specificity: 97,5% (4 + 156)

Minimum detection limit study
Eight (8) different strains of Strep A were evaluated with the ulti med Strep A Rapid Test device. The minimum detectable level differed slightly depending upon the strain being tested. The detection level of all of the strains was roughly within one magnitude in concentration of each other. Five (5) strains showed a minimum detectable level at roughly $1x10^4$ organisms per swab while three (3) strains showed a minimum detectable level at roughly $1x10^5$ organisms per swab.

Dose Hook
Swabs were spiked with Strep A cells yielding concentrations of 1.0×10^9, 1.0×10^8, and 1.0×10^7 organisms per swab to determine if a prozone effect, or a decrease in signal with increasing analyte, has occured in this assay. Three lots of cassettes were tested. The swabs were run according to the package insert in replicates of three. The results showed that the ulti med Streptococcus A Cassette is capable of detecting up to 10^9 organisms / swab without deleterious effect.

Strep A Organism / swab	Test A	Test B	Test C
1.0×10^9	Pos	Pos	Pos
1.0×10^8	Pos	Pos	Pos
1.0×10^7	Pos	Pos	Pos

Interfering Substances
Swabs were spiked with the interfering substances (cough drops, cough syrup, aseptic spray, or mouthwash) at a starting concentration of 1 %. These swabs were then spiked with either low or medium Strep A specimen levels. The swabs were tested according to the package insert in replicates of three. The tests were rated as either positive or negative at the read time. The results demonstrated that these substances do not interfere with the expected results.

Interfering Substances	Level Tested		
	Neg	Low	Medium
Cherry Halls cough drops	Neg	Pos	Pos
Menthol halls cough drops	Neg	Pos	Pos
Robitussin cough drops	Neg	Pos	Pos
Dimetapp cough syrup	Neg	Pos	Pos
Vicks Chloraseptic spray	Neg	Pos	Pos
Cepacol Chloraseptic spray	Neg	Pos	Pos
Listerine mouthwash	Neg	Pos	Pos
Scope Mouthwash	Neg	Pos	Pos

Cross-Reactivity
The bacterial strains below were spiked on swabs at a final concentration of 1.0×10^7 org/swab. These swabs were tested according to the package insert in duplicates. The Devices were rated as either positive or negative at the read time. The results demonstrate that there is no cross reactivity at the prescribed read time.

Organisms	ATCC No.	Visual Call
Bordetella pertussis	8467	Neg
Branham ella catarrhalis	25238	Neg
Candida albicans	1106	Neg
Corynebacterium diphtheriae	13812	Neg
Enterococcus durans	19432	Neg
Enterococcus faecalis	19433	Neg
Hem ophilus influenzae	9006	Neg
Klebsiella pneumoniae	9987	Neg
Neisseria gonorrhea	27633	Neg
Neisseria meningitides	13077	Neg
Neisseria sicca	9913	Neg
Neisseria subflava	14799	Neg
Pseudomonas aeruginosa	9721	Neg
Serratia marcescens	8100	Neg
Staphylococcus aureus	12598	Neg
Staphylococcus epidermidis	1228	Neg

Organisms	ATCC No.	Visual Call
Strep B	12386	Neg
Strep C	12401	Neg
Strep F	12392	Neg
Strep G	12394	Neg
Streptococcus agalactiae	13813	Neg
Streptococcus canis	43496	Neg
Streptococcus equisim ilis	9528	Neg
Streptococcus equisim ilis	9542	Neg
Streptococcus equisim ilis	12388	Neg
Streptococcus mutants	25175	Neg
Streptococcus pneumonie	27338	Neg
Streptococcus sanguis	10556	Neg
Streptococcus oralis	9811	Neg
Streptococcus mitis	903	Neg
Streptococcus anginosus	33397	Neg
Streptococcus intermedius	27335	Neg

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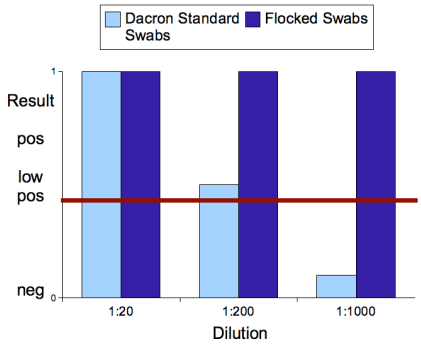
Field Study
Three physicians' offices were used to conduct an evaluation of ulti med Streptococcus A Cassette. Personnel with various educational backgrounds performed the testing. Each physician's office tested a randomly coded panel of samples consisting of negative (20), low positive (20), and medium positive (20) for three days. The results obtained had a 96% correlation with the expected results.

Detection of Streptococcus A Antigen*



* The comparison of the swabs was performed with Streptococcus A Test Cassette and different dilutions of Streptococcus A antigen

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	Manufacturer	Σn	Contents sufficient for <n> tests
IVD	For in vitro diagnostic use only	LOT	Lot. no.
	For single use only		Expiration date
	Read instructions for use		Store at
	Keep away from direct sunlight		

This operating manual conforms to the latest technology / revision. Subject to change without prior notice!

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