INTENDED USE
Wampole ColorCard® Streptozyme® is a color-enhanced slide test for the rapid qualitative and semi-quantitative detection of antibodies to streptococcal extracellular antigens in serum, plasma, and peripheral blood.

SUMMARY AND EXPLANATION
Wampole’s ColorCard® Streptozyme® is a simple, rapid screen with titration method for detection of antibodies to the extracellular antigens of Group A Streptococcus. Such extracellular antigens may develop in rheumatic fever, streptococcal pharyngitis, pyoderma, glomerulonephritis, and other related conditions.

The serological procedure most widely used to detect Group A Streptococcus in symptomatic patients has been the anti-streptolysin O (ASO) test. Since streptolysin is only one of several Group A Streptococcus exoenzymes, the anti-streptolysin test will not detect the antibodies to other exoenzymes of Group A Streptococcus.

Wampole ColorCard® Streptozyme® will detect anti-streptolysin O, anti-streptokinase, anti-hyaluronidase, and antibodies to other streptococcal antigens in sera having an ASO titer of 166 Todd units or above, and in an additional 20% of patients with negative ASO titers.

Wampole ColorCard® Streptozyme® is therefore superior to the conventional ASO test for the detection of antibodies to extracellular antigens of Group A, and will detect more positive streptococcal exoenzyme antibody specimens than will any test for single antibodies.

PRINCIPLE
The Wampole ColorCard® Streptozyme® Reagent consists of a color enhanced standardized suspension of aldehyde-fixed sheep cells, sensitized with Group A Streptococcus extracellular antigens such as streptolysin, streptokinase, hyaluronidase, DNase, and NADase, which will react with antibodies to these antigens to give a positive agglutination reaction. The added coloration facilitates the recognition of positive and negative reactions.

REAGENTS
For in vitro diagnostic use, refrigerate between 2° and 8°C. Do NOT FREEZE.

1. Wampole ColorCard® Streptozyme® Reagent (Standardized sheep cells sensitized with Group A Streptococcus Extracellular Antigens): contains buffer and sodium azide 0.1% as preservative. Shake well before using.
2. Positive Control Serum (Human): In a buffer containing sodium azide 0.1% as preservative.
3. Negative Control Serum (Human): In a buffer containing sodium azide 0.1% as preservative.

BIBLIOGRAPHY
MATERIALS PROVIDED
1. Wampole ColorCard® Streptozyme® Reagent, containing buffer and sodium azide 0.1%.
2. POSITIVE CONTROL SERUM (HUMAN), containing buffer, stabilizer and sodium azide 0.1%.
3. NEGATIVE CONTROL SERUM (HUMAN), containing buffer, stabilizer and sodium azide 0.1%.
4. Pipettes/Stirrers.
5. Disposable Cards.

MATERIALS NOT PROVIDED
1. Test tubes for dilution.
2. Stainer.
3. Isotonic saline (0.95% sodium chloride).
4. Pipettes or strings with large hypodermic needles for dilution.
5. High-intensity light source.

TEST PROCEDURE (QUALITATIVE)
In order to obtain accurate and reproducible results, the test procedure must be carefully followed.

1. Dilute the sample 1:100 with isotonic saline.
2. Place one drop of diluted patient sample (50µL) into the center of a circle. Retain stainer for mixing (step 4).
3. Use one drop of Wampole ColorCard® Streptozyme® reagent.
4. Use the flat end of appropriate pipette/stirrer, spread the mixture uniformly over the entire test area.
5. Rock slide gently with rotary motion for two minutes and immediately observe for agglutination.

BLOOD (PERIPHERAL)
1. Draw blood from suitable area (fingertip, earlobe, etc.).
2. Using a clean pipette/stirrer, place one drop of diluted patient sample (50µL) into a tube containing 2 mL isotonic saline.
   On the basis of a 90% hematocrit, this 1:50 blood dilution is equivalent to a 1:100 serum dilution.
3. Proceed using steps 2, 3, 4 and 5 under Serum or Plasma Testing.

TEST PROCEDURE (SEMI-QUANTITATIVE)
In order to obtain accurate and reproducible results, the test procedure must be carefully followed.

1. Wampole ColorCard® Streptozyme® antibody, prepare the dilutions illustrated in the following table:

   PREPARATION OF DILUTIONS for Wampole ColorCard® Streptozyme® TITER
   | 0.1mL serum or plasma sample | + | 9.9mL saline | = | 1:100
   | 1.0mL primary dilution | + | 1.0mL saline | = | 1:200
   | 0.5mL primary dilution | + | 1.5mL saline | = | 1:400
   | 0.5mL primary dilution | + | 2.5mL saline | = | 1:600
   | 0.5mL primary dilution | + | 5.0mL saline | = | 1:800

   NOTE: When blood (peripheral) is used, follow the above dilution procedure, making sure the blood is not allowed to clot, if no anticoagulant is used.

QUALITY CONTROL
It is recommended that a positive and a negative control be tested for each new test kit and as often as the needs of the lab dictate.

INTERPRETATION OF RESULTS
Qualitative Test Results
POSITIVE A positive result is a visible agglutination of brown particles against a greenish background.

Quantitative Test Results
Utilizing the titration procedure, the last dilution that shows a positive agglutination on the slide is considered the Wampole ColorCard® Streptozyme titer, and should be reported as the Wampole ColorCard® Streptozyme titer.

When using blood, assuming a 50% hematocrit, the final blood dilution represents half the serum dilution in the above table, e.g., the 1:100 dilution of blood is equivalent to a 1:200 dilution of serum.

LIMITATIONS
It is important to note that serial titrations, performed weekly or biweekly for up to six weeks following the initial streptococcal infection, is much more significant than a single determination. The titer of positive sera should be determined using the Semi-Quantitative procedure. The progress of the disease and treatment can only be determined by sequential determination of the patient’s antibody titer.

EXPECTED VALUES
Todd units are commonly used to express ASO titer. Todd units refer to the reciprocal value of the highest dilution which shows no hemolysis. It is important to note that Todd units refer to ASO and only ASO, whereas Wampole ColorCard® Streptozyme® detects antibodies to multiple streptococcal extracellular antigens, including Streptolysin O. For this reason, it is technically incorrect to express the titer (dilutions) in Todd units.

It is possible for a patient to have a significant Wampole ColorCard® Streptozyme® titer and still have a negative response to ASO. Approximately 20% of patients that tested positive with Wampole ColorCard® Streptozyme® titers of 1:100 or higher will have negative ASO titers (less than 160 Todd units).

PERFORMANCE CHARACTERISTICS
Comparison Studies
QUALITATIVE TEST
One hundred ninety eight (198) serum and plasma specimens were examined employing the Wampole ColorCard® Streptozyme® test and a commercially available slide test for detection of antibodies to extracellular antigens of Group A Streptococci. The specimens tested were a mixture of 400 positives, 400 negatives, and unknown specimens, diluted 1:100 with isotonic saline. The tests were performed according to their respective manufacturer's directions insert. One hundred and forty six (146) of the same samples were positive by both assays, and sixty two (52) of the samples were negative by both assays.

Semi-Quantitative Test
When forty (40) positive specimens were compared using the semi-quantitative titration test procedure, the Wampole ColorCard® Streptozyme® test exhibited a slightly enhanced sensitivity and readability compared with the commercial test.