INTENDED USE
SAS™ Adeno Test is a membrane-based immunogold assay for the detection of adenovirus and adenovirus antigens. The test is a rapid visual test for the qualitative detection of adenovirus serotypes present in eye swabs, nasal and pharyngeal secretions, fecal samples, and cell culture supernatant. This test is for professional use only.

SUMMARY AND EXPLANATION OF THE TEST
Adenovirus has six subgenera and 49 serotype based on DNA sequencing and biological and biochemical properties (1, 3, 4). Morphologically, the adenoviruses are nonenveloped icosahedral structures about 80 nm in diameter (4). Adenovirus has been implicated in diseases affecting the respiratory, the ocular and the gastrointestinal systems (1-3). The monoclonal antibody in the SAS™ Adeno Test is against the group-reactive antigen of human adenovirus.

Conjunctivitis caused by adenovirus is frequently seen in ocular infections. Several studies have confirmed that severe conjunctivitis such as keratoconjunctivitis (KCK), pharyngoconjunctival fever (PCF) and nonspecific follicular conjunctivitis (NFC) are caused predominantly by serotypes 3, 4, 6, 11, 19, and 37 in Japan (4).

Adenovirus is also a common cause of upper respiratory tract infections (URT). These infections are manifested in the form of common colds, pharyngitis, or tonsillitis and occur mostly in infants and young children (3). A notable feature of these infections serotypes is the persistence of virus in a latent state in the adenoidal and tonsillar tissues in about 50% of infected children. Another important feature of the infection of this virus is the excretion of virus in the stool for several months without recurrence of symptoms (3).

The gold standard for identifying Adenovirus in conjunctival specimens is culture or electron microscopy (7, 8). Other tests are also available such as immunofluorescence, enzyme immunoassay, and PCR (4, 7, 8). To perform any of these tests, it takes between several hours and a week, in addition, there is a need for sophisticated instruments to obtain the results.

URT and ocular infections frequently manifest similar symptoms of a bacterial infections (6), thus, rapid confirmation of viral infections in patients, often saves on unnecessary antibiotics prescriptions. The SAS™ Adeno Test can be used for the direct testing of eye swab samples, nasal and pharyngeal secretions, fecal samples, and cell culture supernatant, reducing the times required by traditional cell culture isolation.

PRINCIPLE OF THE TEST
The immunochromatographic test utilizes a pair of Adenovirus-specific monoclonal antibodies. An extract is first prepared by suspension of the specimen in the extraction buffer solution. The buffer solution contains sodium azide, which may react with lead and copper plumbing to form explosive metal azides. Azide build-up may be avoided by flushing drains with large volumes of water after disposal.

Materials and reagents provided:
1. Test devices contain a test strip with a monoclonal anti-adenovirus, colored conjugate, and polyclonal affinity purified and is specific to the hexon group of the virus.
2. Tubes containing extraction buffer – Buffer contains 0.1% sodium azide
3. Adenovirus specific sample transfer pipettes
4. Package insert

Materials NOT provided:
1. Sterile specimen collections swabs
2. Precision micropipette and micropipette tips to deliver 500 l (optional)
3. Vortex or centrifuge
4. Timer
5. Adenovirus positive control
6. Adenovirus negative control

PRECAUTIONS:
1. For in-vitro diagnostic use only.
2. The test device should remain in the sealed pouch until ready for use.
3. Do not mouth pipette samples.
4. Do not smoke, eat or drink in areas where specimens or kit components are handled.
5. All specimens, reagents, and controls should be considered potentially hazardous and handled in the same manner as an infectious agent.
6. Wear disposable gloves while handling samples and wash hands after the assay is complete. Warning: The user should refer to the relevant section of the CDC-NIH manual “Biosafety in Microbiology and Biomedical Laboratories,” 3rd Edition, 1984.
7. Avoid contact with the eyes, broken skin, or mucous membranes.
8. Avoid splashing or the generation of aerosols.
9. The test device and all materials should be discarded in a proper biohazard container after testing.

For In-Vitro Diagnostic Use
Store at 15° to 30°C

For Technical Assistance Call 800-272-2710
Outside the USA Call 210-699-8800

For Technical Assistance Call 800-272-2710
Outside the USA Call 210-699-8800
Eye Swabs:

1. Using a sterile swab, wipe the lower palpebral conjunctiva. Swabs must be extracted using SAS™ extraction buffer provided. Swirl the swab well in the extraction buffer-containing tube provided. Rub the swab thoroughly against the wall of the tube.

2. Nasopharyngeal or Tonsilophaqaryngeal swabs: a sterile swab is inserted into one or both nostrils to the nasopharyngeal area. The swab is allowed to remain in the nostril for a few seconds to absorb secretions, rotated gently, and then withdrawn. A separate swab used for each nostril may increase the specimen volume. Alternatively, rub the tonsils and the posterior pharynx thoroughly with a sterile swab. Swabs must be extracted in SAS™ extraction buffer. Swirl the swab well in the extraction buffer-containing tube provided. Rub the swab thoroughly against the wall of the tube.

3. Cell Culture Specimens:
   - Grow the cell culture according to the manufacturer’s guidelines. Aspirate 500 ul of the supernatant fluid for testing. Add sample to the tube containing SAS™ extraction buffer. Shake the mixture well or vortex the tube. Some culture media may contain stabilizers, detergents and animal sera that may adversely affect test results. To qualify cell culture media, seed the media with known positive and negative organisms and test.

4. Fecal Samples:
   - It is recommended that the specimen be collected during the acute phase of gastroenteritis, because a large number of viral particles and viral antigens are excreted during this period. A sample can be collected from a soiled diaper of young children and neonates or an adult stool sample. Alternately, rectal swabs may be used. When using rectal swabs, care should be taken to ensure that a sufficient sample (40-50 mg) is obtained. Both loose and solid stools may be used. Approximately 40-50 mg of raw stool should be collected and added to the extraction buffer. Rub the swab meticulously against the inner wall of the tube containing the extraction buffer. For best results, vortex the sample, then allow the coarse particles to settle before applying the sample to the test.

TEST PROCEDURE

Allow the pouch (test device), specimen and/or controls to reach room temperature (15° - 30°C) prior to testing. Prepare swabs or samples containing the provided SAS™ extraction buffer. Rub the swab carefully against the tube containing extraction buffer.

1. Remove the test device from the protective pouch and place it on a flat surface. Label the device with control or test identifications.
2. Using the sample transfer pipettes provided, dispense 4 drops (approximately 150µl) of the specimen into the round sample well (see illustration below). Wait for colored lines to appear.
3. Read results within 15 minutes. Some positive results may be observed in as short as 30 seconds depending on the concentration of antigen. Do not interpret results after 15 minutes.

INTERPRETATION OF RESULTS

Negatives
   - The test is negative if only one colored line appears in the (control) area.

Positive results
   - The test is positive if two colored lines appear. One colored line will appear in the S (specimen) area and one in the C (control) area. Any colored line in the S area should be considered positive. Colored lines may be lighter or darker than each other.

Invalid Results
   - The test is invalid if no colored line appears in the C (control) area even if a colored line appears in the S (specimen) area. The test should be repeated (see the “Invalid Results” section). If the line in the C area still does not appear, contact our Technical Support Department at (210) 699-8800. It is recommended that when a new shipment of product is received, negative and positive controls for adenovirus should be tested and the appropriate results obtained. (See NCCLS C24-A for guidance on appropriate quality control practices.)

LIMITATIONS OF THE PROCEDURE

1. SAS™ Adeno Test is highly sensitive and specific for adenovirus antigen. The monoclonal antibody in this test reacts with the group specific hexon antigen. It will detect all known serotypes, but cannot be used to differentiate types.
2. The test is highly dependent on the collection and transportation of clinical specimen. Care should be taken to adhere to proper procedures.
3. A negative result does not exclude the possibility of adenovirus infection in the patient. False negative results may occur due to low concentration levels of the adenovirus antigen below the sensitivity level of the test, improper sampling or handling of the specimen, etc.
4. Test results depend on the level of antigen in clinical specimens and may not correlate with cell cultures.
5. Adenovirus may be found in both solid and loose stools. Our data was obtained using both stool types. Adenovirus may be found in the stools of asymptomatic children. Asymptomatic shedding may occur up to 18 months after infection. Enteric adenovirus may be found in the stools of asymptomatic children.

EXPECTED VALUES

The prevalence of adenovirus infection will vary based on many factors such as infection category, geographic location, method of sample collection, sample handling and transportation, and the general health environment of the patient population under study. Normal healthy individuals tested should be negative for adenovirus. Some infected individuals may show symptoms or only minor symptoms, and these patients may test negative.

The frequency of adenovirus infections will vary with the clinical syndrome and age of the individual. Approximately 5% of acute respiratory disease in children under the age of 5 is due to adenovirus (9). Enteric adenoviruses (types 40 and 41) have been implicated in approximately 10% of pediatric patients suffering from gastroenteritis, and appears most frequently in children under 2 years old (10). Approximately 10% of childhood pneumonia may be of adenovirus etiology (14)."

“Adenovirus has at times been implicated in cervicitis (11) and in acute respiratory disease (12) in adults. Occul infections such as epidemic keratoconjunctivitis due to adenovirus can occur in any age group (13). In a Japanese study of 1105 patients of various ages with viral conjunctivitis, 536 (49%) were determined to be caused by adenovirus. Similarly, studies in three East Asian cities found that 70% of epidemic kerato-conjunctivitis cases were caused by adenovirus (15). Our clinical studies produced similar results. In 178 out of 292 patients tested (61.0%), was confirmed via PCR to be caused by adenovirus.”

PERFORMANCE CHARACTERISTICS

The SAS™ Adeno Test was tested in laboratories, clinics, and hospitals in the United States, Japan, and France for tissue culture confirmation, and for the direct testing of stool samples, eye swabs, and nasopharyngeal swabs. Tissue culture samples were CPE formation in culture and to Meridian Premier Adenoclone®; Stool samples were confirmed by EM and results compared to Orion Diarlex™ Rota-Adeno; eye swabs were confirmed by PCR.
Serial two-fold dilutions of each virus suspension were assayed and the following results were obtained:

<table>
<thead>
<tr>
<th>Adenovirus Serotype</th>
<th>Protein Concentration (ug/ml)</th>
<th>SAS™ Adeno Test Results</th>
</tr>
</thead>
<tbody>
<tr>
<td>+</td>
<td>1.260</td>
<td>+</td>
</tr>
<tr>
<td>+</td>
<td>0.600</td>
<td>+</td>
</tr>
<tr>
<td>+</td>
<td>0.300</td>
<td>+</td>
</tr>
<tr>
<td>+</td>
<td>0.150</td>
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<tr>
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<td>+</td>
<td>0.014</td>
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</tr>
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</table>

**CROSS REACTIVITY**

The following organisms and viruses have been tested and showed no cross-reactivity:

<table>
<thead>
<tr>
<th>Microorganisms</th>
<th>Viruses</th>
</tr>
</thead>
<tbody>
<tr>
<td>S. pneumoniae</td>
<td>Coxsackievirus B-4</td>
</tr>
<tr>
<td>N. gonorrhoea</td>
<td>Enterovirus</td>
</tr>
<tr>
<td>E. coli</td>
<td>Rhinovirus</td>
</tr>
<tr>
<td>S. epidermidis</td>
<td>Chlamydia pneumonia</td>
</tr>
<tr>
<td>P. aeruginosa</td>
<td></td>
</tr>
</tbody>
</table>

**ASSAY PRECISION**

Two samples, one positive and one negative, were tested twenty times by three technicians. In each test, the positive sample produced a positive result, and the negative sample produced a negative result.

**Inter-Assay**

Positive and negative samples were run using test devices from different lots of SAS™ Adeno Test. In each test, the positive sample produced a positive result and the negative sample produced a negative result.

**BIBLIOGRAPHY**


12. SAS #70-PI-ADE Rev 10-07

Authored Representative: MegCor Code Sr. 40 C/777 Lichtenfelder Eschbacher – Germany

**Site IV**

<table>
<thead>
<tr>
<th>Comparison</th>
<th>PCR</th>
<th>SAS™ Adeno Test</th>
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<tbody>
<tr>
<td>+</td>
<td>+</td>
<td>Ad +</td>
</tr>
<tr>
<td>+</td>
<td>+</td>
<td>Ad +</td>
</tr>
<tr>
<td>-</td>
<td>-</td>
<td>Ad -</td>
</tr>
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</table>

**Site V**

<table>
<thead>
<tr>
<th>Comparison</th>
<th>PCR</th>
<th>SAS™ Adeno Test</th>
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</thead>
<tbody>
<tr>
<td>+</td>
<td>+</td>
<td>Ad +</td>
</tr>
<tr>
<td>-</td>
<td>-</td>
<td>Ad -</td>
</tr>
</tbody>
</table>

Note: Please be advised that "relative" refers to the comparison of this assay's results to that of a similar assay. There was not an attempt to correlate the assay's results with disease presence or absence. No judgement can be made on the comparison assay's accuracy to predict disease.

**LIMITS OF DETECTION**

A study was performed at a University School of Medicine in Japan to measure the detection limits of the SAS™ Adeno Test.