todylaboratories®

Rapid Tests

CE IVD REF C61RT1005

Rotavirus Rapid Test Cassette (Feces)
Package Insert

A rapid, one step test for the qualitative detection of rotavirus in human feces.
For professional in vitro diagnostic use only.

INTENDED USE
The Rotavirus Rapid Test Cassette (Feces) is a rapid chromatographic immunoassay for the qualitative detection of rotavirus in human feces specimens to aid in the diagnosis of rotavirus infection.

SUMMARY
Rotavirus is the most common agent responsible for acute gastroenteritis, mainly in young children.[1] Its discovery in 1973 and its association with infantile gastro-enteritis represented a very important advancement in the study of gastro-enteritis not caused by acute bacterial infection. Rotavirus is transmitted by oro-faecal route with an incubation period of 1-3 days. Although specimen collections taken within the second and fifth day of the illness are ideal for antigen detection, the rotavirus may still be found while diarrhoea continues. Rotaviral gastroenteritis may result in mortality for populations at risk such as infants, the elderly, and immunocompromised patients.[2] In temperate climates, rotavirus infections occur mainly in the winter months. Endemics as well as epidemics affecting some thousand people have been reported.[3] With hospitalised children suffering from acute entric disease up to 50% of the analysed specimen were positive for rotavirus.[4] The viruses replicate in the cell nucleus and tend to be host species specific producing a characteristic cytopathic effect (CPE). Because rotavirus is extremely difficult to culture, it is unusual to use isolation of the virus in diagnosing an infection. Instead, a variety of techniques have been developed to detect rotavirus in feces.
The Rotavirus Rapid Test Cassette (Feces) is a rapid chromatographic immunoassay for the qualitative detection of rotavirus in human feces specimen, providing results in 10 minutes. The test utilizes antibody specific for rotavirus to selectively detect rotavirus from human feces specimens.

PRINCIPLE
The Rotavirus Rapid Test Cassette (Feces) is a qualitative, lateral flow immunoassay for the detection of rotavirus in human feces specimens. In this test, the membrane is pre-coated with anti-rotavirus antibody on the test line region of the test. During testing, the specimen reacts with the particle coated with anti-rotavirus antibody. The mixture migrates upward on the membrane chromatographically by capillary action to react with anti-rotavirus antibody on the membrane and generate a colored line in the test line region. The presence of this colored line in the test region indicates a positive result, while its absence indicates a negative result. To serve as a procedural control, a colored line will always appear in the control line region indicating that proper volume of specimen has been added and membrane wicking has occurred.

REAGENTS
The test contains anti-rotavirus antibody coated particles and anti-rotavirus antibody coated on the membrane.

PRECAUTIONS
- For professional *in vitro* diagnostic use only. Do not use after expiration date.
- The test cassette should remain in the sealed pouch until use.
- Do not eat, drink or smoke in the area where the specimens or kits are handled.
- Do not use test if pouch is damaged.
- Handle all specimens as if they contain infectious agents. Observe established precautions against microbiological hazards throughout testing and follow standard procedures for proper disposal of specimens.
- Wear protective clothing such as laboratory coats, disposable gloves and eye protection when specimens are being tested.
- The used test should be discarded according to local regulations.
- Humidity and temperature can adversely affect results.

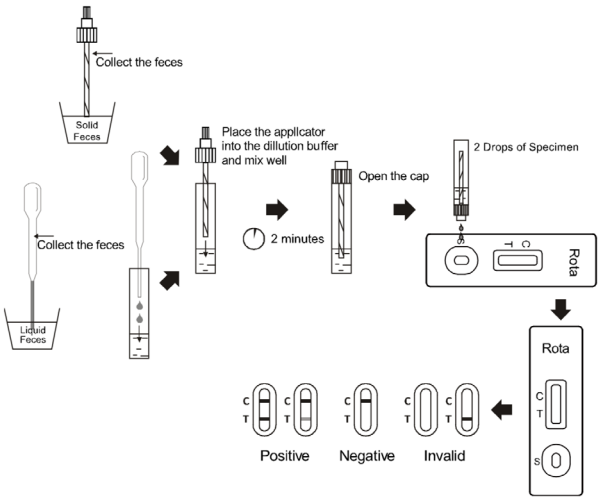
STORAGE AND STABILITY
Store as packaged in the sealed pouch either at room temperature or refrigerated (2-30°C). The test is stable through the expiration date printed on the sealed pouch. The test must remain in the sealed pouch containing desiccant until use. **DO NOT FREEZE.** Do not use beyond the expiration date.

SPECIMEN COLLECTION AND PREPARATION
1. Viral detection is improved by collecting the specimens at the onset of the symptoms. It has been reported that the maximum excretion of rotavirus in the feces of patients with gastroenteritis occurs 3-5 days after onset of symptoms. If the specimens are collected long after the onset of diarrheic symptoms, the quantity of antigen may not be sufficient to obtain a positive reaction or the antigens detected may not be linked to the diarrheic episode.
2. The feces specimen must be collected in a clean, dry, waterproof container containing no detergents, preservatives or transport media.
3. Bring the necessary reagents to room temperature before use.

MATERIALS
Materials provided
• Test Cassettes • Droppers • Specimen collection tubes with extraction buffer • Package Insert
Materials required but not provided
• Specimen Collection Containers • Centrifuge and pipette to dispense 80 µL if required • Timer

DIRECTIONS FOR USE
Allow the test, specimen, buffer, and/or controls to reach room temperature (15-30°C) prior to testing.
1. To collect fecal specimens:
Collect sufficient quantity of feces (1-2mL or 1-2g) in a clean, dry specimen collection container to obtain enough virus particles. Best results will be obtained if the assay is performed within 6 hours after collection. Specimen collected may be stored for 3 days at 2-8°C if not tested within 6 hours. For long term storage, specimens should be kept below - 20°C.
2. To process fecal specimens:
• For **Solid Specimens**:
Unscrew the cap of the specimen collection tube, then randomly **stab the specimen collection applicator into the fecal specimen in at least 3 different sites** to collect approximately **50 mg of feces** (equivalent to 1/4 of a pea). Do not scoop the fecal specimen.
• For **Liquid Specimens**:
Hold the dropper vertically, aspirate fecal specimens, and then transfer 2 drops of the liquid specimen (approximately 50 µL) into the specimen collection tube containing the extraction buffer.
Tighten the cap onto the specimen collection tube, then **shake the specimen collection tube vigorously** to mix the specimen and the extraction buffer. Leave the collection tube for reaction for 2 minutes.
3. Bring the pouch to room temperature before opening it. Remove the test cassette from the 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.
4. Hold the specimen collection tube upright and **unscrew the tip** of the specimen collection tube. Invert the specimen collection tube and **transfer 2 full drops of the extracted specimen** (approximately 80 µL) to the specimen well (S) of the test cassette, then start the timer. Avoid trapping air bubbles in the specimen well (S). See illustration below.
5. Read the results at **10 minutes** after dispensing the specimen. Do not read results after 20 minutes.
Note: If the specimen does not migrate (presence of particles), centrifuge the diluted sample contained in the extraction buffer vial. Collect 80 µL of supernatant, dispense into the specimen well (S). Start the timer and continue from step 5 onwards in the above instructions for use.

INTERPRETATION OF RESULTS
(Please refer to the illustration above)
POSITIVE:* Two distinct colored lines appear. One colored line should be in the control region (C) and another colored line should be in the test region (T).
***NOTE**: The intensity of the color in the test line region (T) will vary depending on the concentration of rotavirus antigen present in the specimen. Therefore, any shade of color in the test line region (T) should be considered positive.
NEGATIVE: One colored line appears in the control region (C). No apparent colored line appears in the test region (T).
INVALID: Control line fails to appear. Insufficient specimen volume or 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 your local distributor.



QUALITY CONTROL
An internal procedural control is included in the test. A colored line appearing in the control line region (C) is an internal positive procedural control. It confirms sufficient specimen volume, adequate membrane wicking and correct procedural technique.
Control standards are not supplied with this kit; however, it is recommended that positive and negative controls be tested as a good laboratory practice to confirm the test procedure and to verify proper test performance.

LIMITATIONS
1. The Rotavirus Rapid Test Cassette (Feces) is for in vitro diagnostic use only. The test should be used for the detection of human rotavirus in feces specimens only. Neither the quantitative value nor the rate of increase in human rotavirus concentration can be determined by this qualitative test.
2. The Rotavirus Rapid Test Cassette (Feces) will only indicate the presence of rotavirus in the specimen and should not be used as the sole criteria for the conforming rotavirus to be etiological agent for diarrhea.
3. As with all diagnostic tests, all results must be interpreted together with other clinical information available to the physician.
4. If the test result is negative and clinical symptoms persist, additional testing using other clinical methods is recommended. A negative result does not at any time preclude the possibility of rotavirus infection with low concentration of virus particles.

EXPECTED VALUES
The Rotavirus Rapid Test Cassette (Feces) has been compared with latex agglutination method, demonstrating an overall accuracy of 97.2%.

PERFORMANCE CHARACTERISTICS
Clinical Sensitivity, Specificity and Accuracy
The performance of the Rotavirus Rapid Test Cassette has been evaluated with 501 clinical specimens collected from children and young adults in comparison with latex agglutination method. The results show that the relative sensitivity of the Rotavirus Rapid Test Cassette (Feces) is 97.3% and the relative specificity is 97.1%.

Rotavirus Rapid Test Cassette vs. Latex Agglutination

Method		Latex Agglutination		Total Results
	Results	Positive	Negative	
Rotavirus Rapid Test Cassette	Positive	251	7	258
	Negative	7	236	243
Total Results		258	243	501

Relative Sensitivity: 97.3% (95%CI:*94.5%-98.9%)
Relative Specificity: 97.1% (95%CI:*94.2%-98.8%)
Overall Accuracy: 97.2% (95%CI:*95.4%-98.5%) *Confidence Intervals

Precision
Intra-Assay
Within-run precision has been determined by using 10 replicates of four specimens: a negative, a low positive, a medium positive and a high positive. The specimens were correctly identified >99% of the time.
Inter-Assay
Between-run precision has been determined by 10 independent assays on the same four specimens: a negative, a low positive, a medium positive and a high positive. The specimens were correctly identified >99% of the time.
Cross-reactivity
Cross reactivity with following organisms has been studied at 1.0 x 10^8 organisms/ml. The following organisms were found negative when tested with the Rotavirus Rapid Test Cassette (Feces).

Staphylococcus aureus	Pseudomonas aeruginosa	Enterococcus faecalis	Group C Streptococcus
Klebsiella pneumoniae	Branhamella catarrhalis	Hemophilus influenzae	Neisseria gonorrhea
Group B Streptococcus	Proteus vulgaris	Enterococcus faecium	Proteus mirabilis
Candida albicans	Neisseria meningitides	Acinetobacter spp	Salmonella choleraesius
Gardnerella vaginalis	Acinetobacter calcoaceticus	E.coli	Chlamydia trachomatis

Interfering Substances
The following potentially Interfering Substances were added to Rotavirus negative and positive specimens. No interference observed.

Ascoribic acid: 20mg/dl	Oxalic acid: 60mg/dl	Bilirubin: 100mg/dl
Uric acid: 60mg/dl	Urea: 2000mg/dl	Caffeine: 40mg/dl
Glucose: 2000mg/dl	Aspirin: 20mg/dl	Albumin: 2000mg/dl

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