

## IMMUNOQUICK® NoRotAdeno

Rapid test for the detection Norovirus, Rotavirus and Adenovirus in feces



### INTENDED USE

IMMUNOQUICK® NoRotAdeno is a rapid chromatographic immunoassay for the qualitative detection of Norovirus, Rotavirus and Adenovirus antigens in stool.

### INTRODUCTION

Acute gastroenteritis continues to be a major cause of morbidity and mortality throughout the world with around 700 million annual cases in children under five years and a mortality rate estimated to be 3-5 million cases per year. Although all bacteria, viral agents and parasites could be responsible for gastroenteritis, many studies have illustrated that viral agents are the most important factors.

Rotavirus, Norovirus, and Adenovirus have been reported as the common viral pathogens of acute gastroenteritis in children. The main symptoms of viral gastroenteritis are watery diarrhea and vomiting. The affected person may also have headache, fever, and abdominal cramps ("stomach ache").

Rotavirus is transmitted via the feco-oral route and, following establishment in the small intestine, results in nutrient malabsorption. The clinical spectrum of acute Rotavirus gastroenteritis ranges from a self-limited watery diarrhea illness accompanied with nausea, anorexia and mild vomiting or fever, to severe dehydration resulting in hospitalization or even death. Although Rotavirus primarily infects children, it could cause a mild disease in adults.

In some reports, Adenovirus is considered as another etiological agent of viral gastroenteritis after Norovirus and Rotavirus. Noroviruses are a major cause of gastrointestinal illness in closed and crowded environments, such as hospitals, nursing homes and cruise ships. Noroviruses commonly spread through food or water contaminated by fecal matter during preparation. Adenoviruses infection is mostly seen in pediatrics up to 2 years old.

Since most viruses causing gastroenteritis cannot be isolated in cell culture, direct visualization in stool specimens by electron microscopy is still the mainstay of diagnosis although it is limited to reference laboratories. The introduction of more sensitive techniques for antigen detection in stool based on immunoassay has improved the diagnosis of newly recognized viruses as Norovirus.

The symptoms of Norovirus illness usually include nausea, vomiting, diarrhea, and some stomach cramping. Sometimes people additionally have a low-grade fever, chills, headache, muscle aches, and a general sense of tiredness. The illness often begins suddenly, and the infected person may feel very sick. In most people the illness is self-limiting with symptoms lasting for about 1 or 2 days. In general, children experience more vomiting than adults.

### TEST PRINCIPLE

IMMUNOQUICK® NoRotAdeno is qualitative, lateral flow immunoassay for the detection of Norovirus, Rotavirus and Adenovirus antigens in stool. The assay uses specific monoclonal antibodies coated on the test membrane. During testing, the stool specimen reacts with the conjugate antibodies. The mixture migrates upward on the membrane chromatographically by capillary action to react with antibodies on the membrane and generates a colored line at the level of the N, R and A zone respectively. The presence of a colored line in N region indicates a positive result for Norovirus, in R region for Rotavirus, and in A region for Adenovirus respectively, while its absence indicates a negative result.

To serve as a procedural control, a colored line will always appear in the control reaction zone (C) indicating that proper volume of specimen has been added and membrane wicking has occurred.

### MATERIAL PROVIDED

- Tests in individual Pouch with desiccant.
- Buffer Bottle(s) filled with buffer Solution.
- Extraction Tubes.
- Swabs.
- Tube holder.
- Instructions for use.

### MATERIAL REQUIRED BUT NOT PROVIDED

- Micropipette.
- Clock or timer.
- Vortex.

### STORAGE AND STABILITY

IMMUNOQUICK® NoRotAdeno should be stored between 2- 30°C. Test is

sensitive to humidity as well as to heat. It is stable until expiration date printed on the bag. Cassettes must be kept in their bags until use. DO NOT FREEZE. Do not use beyond expiration date, indicated on the kit and the aluminium bags.

### PRECAUTIONS

- For in vitro diagnostic use only.
- Samples, reagents and tests should be brought to room temperature before running the test.
- Do not use the test beyond the expiry date. Use of the test beyond the expiry date may lead to incorrect results.
- Do not use other samples than feces.
- Do not freeze the kit. As the quality may change in a frozen kit, do not use it.
- Follow the product insert instructions carefully.
- Remove the test from its sealed pouch just before running the test, due to humidity sensitivity of the test.
- Do not eat, drink or smoke when operating samples or reagents.
- Avoid splashes and aerosol formation.
- Humidity and temperature can adversely affect results.
- Test is intended for single use only, do not reuse. Avoid cross-contamination of specimens by using a new extraction tube and specimen pipette for each specimen.
- If the extract solution accidentally enters the eyes or mouth or contacts the skin, wash the areas sufficiently with water and obtain treatment from a doctor is necessary because the sample extract solution contains sodium azide.

### SAMPLE COLLECTION AND HANDLING

- Handle all specimens as if they contained infectious agents. Wear protective clothing such as laboratory coats, disposable gloves and eye protection. Observe established precautions against microbiological hazards throughout all procedures and follow the standard procedures for proper disposal of specimens and contaminated devices.
- Immediately dilute the fecal sample after it has been collected and use it. The sample extract solution cannot be maintained as a suspension.
- Testing should be performed immediately after the specimens have been collected. For prolonged periods of storage, the specimens should be kept below -20°C. Specimens should not be frozen and thawed repeatedly.
- Bring specimens to room temperature prior to testing. Frozen specimens must be completely thawed and mixed well prior to testing
- In case of shipment of samples, regulatory rules related to shipment of samples of human origin should be followed.

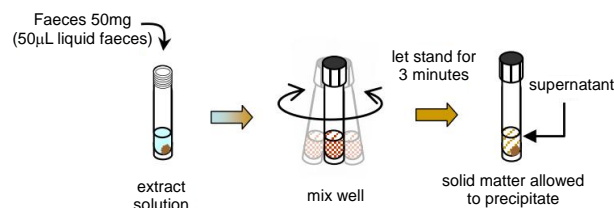
### TEST PROCEDURE

The specimens and tests should be brought to room temperature for at least 15-30 min before testing.

#### 1. Specimen extraction

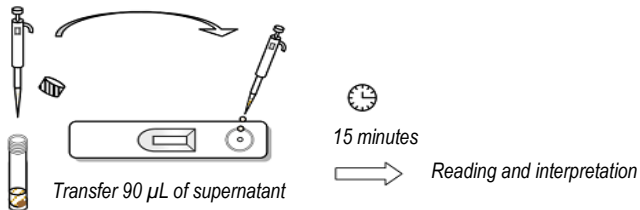
- Add 0.8 mL of diluent into the tube.
- Using the swab, add about 50 mg of stool specimen (about 50µL in the case of liquid feces) into the tube.
- Thoroughly mix with a vortex.
- Let stand for 3 minutes. The resultant supernatant is used as a sample.

**Note:** In the case of sample with high amount of feces or high viscosity, dilute it 2-fold with buffer solution or centrifuge it for 5 minutes at 3000xg and then carry out the test.



## 2. Reaction

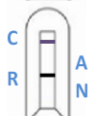
- Remove the test device from the foil pouch, and place it on a clean and flat surface.
- Avoiding solid matter whenever possible, Transfer 90µL of supernatant of the extracted specimen into the sample well.
- Read the test result at 15 minutes. Do not interpret test result after 15 minutes.



## RESULTS INTERPRETATION



**Adenovirus POSITIVE: Presence of 2 distinct lines:**  
A control line appears at the control zone (C) level and one line (even of weak intensity) appear at the Adenovirus zone (A).



**Rotavirus POSITIVE: Presence of 2 distinct lines:**  
A control line appears at the C level and a line (even of weak intensity) appears at the Rotavirus zone (R).



**Norovirus POSITIVE: Presence of 2 distinct lines:**  
A control line appears at the C level and a line (even of weak intensity) appears at the Norovirus zone (N).

**Note:** The intensity of test line vary according to the antigen concentration in the sample. Even if the colour is weak, make a positive judgment if the line can be visually seen.



**Negative:** Only one line appears at the C level.  
No line appears at the A, R and N level.



**INVALID:** No visible line at the C level (independent of the fact whether a line at the A, R or N level appears).  
The test is invalid and should be repeated with a new device.



## Notes:

- If extremely dark lines appear at test lines level (A, R and N) whereas no line appears at the control line (C) level, try to dilute the sample 5-10 fold in buffer solution and repeat the test with a new device.
- Negative results do not necessarily rule out viral infection. If negative results are obtained, make a comprehensive judgment in conjunction with clinical symptoms and other methods.

## QUALITY CONTROL

An internal control of procedure is integrated into the test (control line (C)). That makes it possible to control that the volume of supernatant is sufficient and that the procedure was followed correctly.

## LIMITATIONS OF PROCEDURE

- This test should be used for the detection of Norovirus, Rotavirus, and Adenovirus antigens in human stool only.
- IMMUNOQUICK® NoRotAdeno will only indicate the presence of Norovirus, Rotavirus, Adenovirus antigens in the specimen and should not be used as the sole criteria for the diagnosis of infection.
- As with all diagnostic tests, result must be considered together with other clinical information available to the physician.
- If the test result is negative and clinical symptoms persist, additional follow-up testing using other clinical methods is suggested. A negative result at any time does not preclude the possibility of infection.

## PERFORMANCES

1. **Cross reactivity:** The cross-reactivity between bacteria present in faecal samples when presenting various symptoms of acute gastroenteritis, bacteria which are predicted to be present in the gut and bacteria known to usually be present in the gut was investigated. No cross-reactivity was shown with following bacteria:

- |                              |                             |
|------------------------------|-----------------------------|
| - <i>S. enteritidis</i>      | - <i>E. coli</i> O-111 : NM |
| - <i>S. typhimurium</i>      | - <i>C. coli</i>            |
| - <i>L. monocytogenes</i>    | - <i>C. jejuni</i>          |
| - <i>S. flexneri</i>         | - <i>C. perfringens</i>     |
| - <i>S. sonnei</i>           | - <i>C. difficile</i>       |
| - <i>V. parahaemolyticus</i> | - <i>B. cereus</i>          |
| - <i>V. cholerae</i> non-O1  | - <i>S. aureus</i>          |
| - <i>V. cholerae</i> O-139   | - <i>P. aeruginosa</i>      |
| - <i>E. coli</i> O-157 : H7  | - <i>C. freundii</i>        |
| - <i>E. coli</i> O-157       | - <i>P. mirabilis</i>       |
| - <i>E. coli</i> O-111       | - <i>E. faecium</i>         |

2. **Detection limit:** Concentrations up to 6.25ng/mL for norovirus VLP GII/4, 9.6×10<sup>4</sup>TCID<sub>50</sub>/mL for Rotavirus SA11 and 7.5 × 10<sup>4</sup>TCID<sub>50</sub>/mL for Adenovirus type 3 were judged to be positive. Consequently, the above concentrations were taken as the minimum detection sensitivity.

3. **Sensitivity- specificity:** A comparison study with the RT- PCR method and competitor rapid test was performed.

| Norovirus                  |          | RT-PCR               |                     |       |
|----------------------------|----------|----------------------|---------------------|-------|
|                            |          | positive             | negative            | total |
| IMMUNOQUICK®<br>NoRotAdeno | Positive | 50                   | 0                   | 50    |
|                            | Negative | 2                    | 48                  | 50    |
|                            | Total    | 52                   | 48                  | 100   |
|                            |          | Sensitivity<br>96,2% | Specificity<br>100% |       |

| Rotavirus                  |          | Competitor Rapid Test |                      |       |
|----------------------------|----------|-----------------------|----------------------|-------|
|                            |          | positive              | negative             | total |
| IMMUNOQUICK®<br>NoRotAdeno | Positive | 49                    | 1                    | 50    |
|                            | Negative | 0                     | 62                   | 62    |
|                            | Total    | 49                    | 63                   | 112   |
|                            |          | Sensitivity<br>100%   | Specificity<br>98,4% |       |

| Adenovirus                 |          | Competitor Rapid Test |                     |       |
|----------------------------|----------|-----------------------|---------------------|-------|
|                            |          | positive              | negative            | total |
| IMMUNOQUICK®<br>NoRotAdeno | Positive | 6                     | 0                   | 6     |
|                            | Negative | 0                     | 106                 | 106   |
|                            | Total    | 6                     | 106                 | 112   |
|                            |          | Sensitivity<br>100%   | Specificity<br>100% |       |

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## SYMBOLS



Attention, see instructions for use



Lot number



For *in vitro* diagnostic use only



Manufacturer



Store between 2-30°C



Do not reuse



Tests per kit



Catalog number



Expiry

Version 03 BR 09/2014



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