Infection

ichroma™ Rota/Adeno

INTENDED USE

ichroma™ Rota/Adeno is a fluorescence immunoassay (FIA) for the qualitative determination of both rotavirus and adenovirus in human feces. It is useful as an aid in management and monitoring of viral gastroenteritis.

For *in vitro* diagnostic use only.

INTRODUCTION

The major symptoms of viral enteritis are diarrhea and vomiting. Viral enteritis is infectious disease caused by many viruses such as Rotavirus, Adenovirus.

Rotaviruses (RVs) are the main etiologic agents of serious diarrheal disease in infants and young children under while 2 years of age throughout the world. Group A RVs are the major cause of human infections. Outbreaks with a strict seasonal winter pattern occur in tropic climates. Infections are spread more evenly throughout the year. After a short incubation period of 24–48 h, the onset of illness is sudden, with watery diarrhea, vomiting, and rapid dehydration. Untreated RV infection is a major cause of infantile death in developing countries. [1)]

Adenoviruses may cause epidemics, endemics and sporadic infections in all geographical regions of the world. They do not show seasonal outbreak and can be seen throughout entire year. Adenovirus infection cause variety symptom in several region. Especially, the adenovirus type 40 and 41 cause acute gastroenteritis primarily in children like as Rotavirus. [2)]

Also, it is known that Rotavirus-Adenovirus co-infection rate is above 5 % in gastroenteritis patients. Diagnosing of the viral disease is important in reducing unnecessary use of antibiotics. [2,3)]

ichroma™ Rota/Adeno is an immunoassay for the detection of both Rotavirus and Adenovirus in human stool sample.

PRINCIPLE

The test uses a sandwich immunodetection method.

The detector antibodies in the buffer bind to antigens in the sample, forming antigen-antibody complexes, and migrate onto nitrocellulose matrix to be captured by the other immobilized-antibodies on a test strip.

More antigens in the sample will form more antigen-antibody complexes which lead to stronger fluorescence signal by detector antibodies, which is processed by the instrument for ichroma™ tests to show 'rotavirus positive' and 'adenovirus positive'.

COMPONENTS

ichroma™ Rota/Adeno consists of 'cartridges', 'detector tubes', 'detector diluent' and 'sample collection tubes'.

- The cartridge contains the membrane called a test strip, which has anti-human adenovirus at the test line 1, anti-human rotavirus at the test line 2, and chicken IgY at the control line. All cartridges are individually sealed in an aluminum foil pouch containing a desiccant, and they are further packaged in a box.
- The detector tube has a granule containing anti-human adenovirus-fluorescence conjugate, anti-human rotavirus -fluorescence conjugate, anti-chicken IgY fluorescence conjugate, bromophenol blue, sucrose, tween 20, bovine serum albumin (BSA) as a stabilizer and sodium azide as a preservative in phosphate buffered saline (PBS). All detector tubes are packed in a pouch.
- The detector diluent contains tween 20 and bovine serum albumin (BSA) as a stabilizer and sodium azide as a preservative in phosphate buffered saline (PBS), and it is pre-dispensed in a vial. The detector diluent is packed in a box.
- The sample collection tube contains bovine serum albumin (BSA), tween 20, sodium azide as a preservative in Tris-HCl and DDW. All sample collection tubes are packaged in a box.

WARNINGS AND PRECAUTIONS

- For *in vitro* diagnostic use only.
- Follow the instructions and procedures described in this 'Instructions for use'.
- Use only fresh samples and avoid direct sunlight.
- Lot numbers of all the test components (cartridge, detector tube, detector diluent, sample collection tube and ID chip) must match each other.
- Do not interchange test components between different lots or use test components after the expiration date, either of which might yield incorrect test result(s).
- Do not reuse cartridges, detector tubes or sample collection tubes. A cartridge should be used for testing one sample only. A detector tube or a sample collection tube should be used for processing of one sample only.
- The cartridge should remain sealed in its original pouch until just before use. Do not use the cartridge, if pouch is damaged or has already been opened.
- Frozen sample should be thawed only once. For shipping, samples must be packed in accordance with local regulations.
- If test components and/or sample are stored in refrigerator, then allow cartridge, detector tube, detector diluent, sample collection tubes and sample to be at room temperature for approximately 30 minutes before use.
- The instrument for ichroma™ tests may generate slight vibration during use.
- Used cartridges, detector tubes, detector diluent, sample collection tubes, and pipette tips should be handled carefully and discarded by an appropriate method in accordance with relevant local regulations.
- The detector tube, the detector diluent and the sample collector tube contain sodium azide (NaN_3), and they may cause certain health issues like convulsions, low blood pressure and heart rate, loss of consciousness, lung injury

and respiratory failure. Avoid contact with skin, eyes, and clothing. In case of contact, rinse immediately with running water.

- **ichroma™ Rota/Adeno** will provide accurate and reliable results subject to the below conditions.
  - **ichroma™ Rota/Adeno** should be used only in conjunction with the instrument for ichroma™ tests.

LIMITATION OF THE TEST SYSTEM

- The test may yield false positive result(s) due to the cross-reactions and/or non-specific adhesion of certain sample components to the capture/detector antibodies.
- The test may yield false negative result(s) due to the non-responsiveness of the antigen to the antibodies which is the most common if the epitope is masked by some unknown components, so therefore not being able to be detected or captured by the antibodies. The instability or degradation of the antigen with time and/or temperature may also cause false negative result as it makes the antigen unrecognizable by the antibodies.
- Other factors may interfere with the test and cause erroneous results, such as technical/procedural errors, degradation of the test components/reagents or presence of interfering substances in the test samples.
- Any clinical diagnosis based on the test result must be supported by a comprehensive judgment of the concerned physician in conjunction with clinical symptoms and other relevant test results.

STORAGE AND STABILITY

Storage condition			
Component	Storage Temperature	Shelf life	Note
Cartridge	2 – 30 °C	20 months	Disposable
Detector tube	2 – 30 °C	20 months	Disposable
Detector diluent	2 – 30 °C	20 months	Unopened
		20 months	Opened
Sample collection tube	2 – 30 °C	20 months	Disposable

- After the cartridge pouch is opened, the test should be performed immediately.

MATERIALS SUPPLIED

REF CFPC-79

Components of **ichroma™ Rota/Adeno**

- Cartridge box:
  - Cartridge 25
  - Detector tube 25
  - Detector diluent 1
  - Sample collection tube 25
  - ID chip 1
  - Instructions for use 1

MATERIALS REQUIRED BUT SUPPLIED ON DEMAND

Following items can be purchased separately from **ichroma™ Rota/Adeno**.

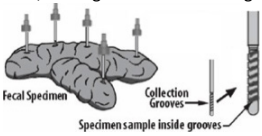
Please contact our sales division for more information.

- Instrument for ichroma™ tests
  - **ichroma™ II** REF FPRR021
  - **ichroma™ III** REF FPRR037
  - **ichroma™ M2** REF FPRR031
- **Boditech Rota/Adeno Control** REF CFPO-164

SAMPLE COLLECTION AND PROCESSING

The sample type for **ichroma™ Rota/Adeno** is human feces.

- Fecal samples must be taken as soon as the symptoms (diarrhea) appear.
- Collect random samples of feces in a clean, dry container or a receptacle, making sure to exclude large solid lumps.



※ Fill up the groove of a sampling stick with fecal samples and please check whether the quantity is too much or not.

- It is recommended to test the sample after collection for immediately.
- The sample (feces) may be stored for 3 days at 2-8 °C prior to being tested. If testing will be delayed more than 3 days, samples should be frozen at -20 °C or below.
- The sample (feces) stored frozen at -20 °C or below for 2 months showed no performance difference.
- As a repeated freeze-thaw cycle may affect the test result, do not refreeze previously frozen samples.

TEST SETUP

- Check the contents of **ichroma™ Rota/Adeno**: Sealed cartridges, detector tubes, a detector diluent, sample collection tubes, an ID chip and an instructions for use.
- Ensure that the lot number of the cartridge matches that of the detector tube, the detector diluent and the sample collection tube as well as an ID chip.
- If the sealed cartridge, the detector tube and the detector diluent have been stored in a refrigerator, place them on a clean and flat surface at room temperature for at least 30 minutes before testing.
- Turn on the instrument for ichroma™ tests.
- Insert the ID chip into the 'ID chip port'.

※ **Please refer to the instrument for ichroma™ tests operation manual for complete information and operating instructions.**

TEST PROCEDURE

▶ **ichroma™ II, ichroma™ M2**

Multi test mode / Read now mode

1) Collect random samples of feces in a clean, dry container or a receptacle, making sure to exclude large solid lumps.
2) Invert a sample collection tube and loosen the cap which is attached a sampling stick (yellow color).
3) Introduce the sampling stick into the fecal sample about 5 - 6 times at different sites. In order to get sampling even in the spirals of the stick and to ensure appropriate specimen to buffer ratio, try to avoid obtaining clumps of fecal matter.
4) Return the sampling stick to the sample collection tube and tighten the cap thoroughly.
5) Shake the tube vigorously to disperse the specimen

throughout the buffer in the tube.
6) Break off the black tip on the outside of the black cap.
7) Open the detector diluent and take 150 µL of detector diluent using a pipette and dispense it to the detector tube containing a granule. When the granule form is completely dissolved in the tube, it becomes detection buffer.
(The detection buffer must be used immediately. Do not exceed 30 seconds.)
8) Apply only 2 drops (about 30 µL) of feces sample using a sample collection tube to the detector tube.
9) Mix well by pipetting or inverting 10 - 20 times.
(The sample mixture must be used immediately.)
10) Take 100 µL of the sample mixture and dispense it into the sample well of the cartridge.
11) Leave the sample-loaded cartridge at room temperature for 12 minutes.
⚠ *Scan the sample-loaded cartridge immediately when the incubation time is over. If not, it will cause inaccurate test result.*
12) To scan the sample-loaded cartridge, insert it into the cartridge holder of the instrument for ichroma™ tests. Ensure proper orientation of the cartridge before pushing it all the way inside the cartridge holder. An arrow is marked on the cartridge especially for this purpose.
13) Tap the 'Start' button on the instrument for ichroma™ tests to start the scanning process.
(ichroma™ M2 is tested automatically after inserting.)
14) The instrument for ichroma™ tests will start scanning the sample-loaded cartridge immediately.
15) Read the test result on the display screen of the instrument for ichroma™ tests.

Single test mode / Walk away mode

1) The test procedure is same with the 'Multi test mode 1) – 11)'.
2) Insert the sample-loaded cartridge into the holder of the instrument for ichroma™ tests. Ensure proper orientation of the cartridge before pushing it all the way inside the cartridge holder. An arrow is marked on the cartridge especially for this purpose.
3) Tap the 'Start' button on the instrument for ichroma™ tests.
(ichroma™ M2 is tested automatically after inserting.)
4) The cartridge goes inside the instrument for ichroma™ tests and will automatically start scanning the sample-loaded cartridge after 12 minutes.
5) Read the test result on the display screen of the instrument for ichroma™ tests.

▶ **ichroma™ III**
1) The test procedure is same with the 'Single test mode'.

INTERPRETATION OF TEST RESULT

- The instrument for ichroma™ tests calculates the test result automatically and displays 'Positive' / 'Negative' / 'Indeterminate'.
- Ancillary value is served in the form of a cut-off index (COI) value.

- **Rotavirus line**

Cut-off index (COI)	Result	Note
≤ 0.9	Negative for rotavirus	No need to additional test
> 0.9, < 1.0	Indeterminate	Need to retest
≥ 1.0	Positive for rotavirus	Need to confirmation test

- **Adenovirus line**

Cut-off index (COI)	Result	Note
≤ 0.9	Negative for adenovirus	No need to additional test
> 0.9, < 1.0	Indeterminate	Need to retest
≥ 1.0	Positive for adenovirus	Need to confirmation test

QUALITY CONTROL

- Quality control tests are a part of the good testing practice to confirm the expected results and validity of the assay and should be performed at regular intervals.
- Quality control tests should also be performed whenever there is any question concerning the validity of the test results.
- Control materials are provided on demand with **ichroma™ Rota/Adeno**. For more information regarding obtaining the control materials, contact Boditech Med Inc.'s Sales Division for assistance.
(Please refer to the instructions for use of control material.)

PERFORMANCE CHARACTERISTICS

- **Analytical Sensitivity**
Two Rotaivurs genotyped samples (genotype WA, DS-1 strain) were tested with the **ichroma™ Rota/Adeno**. All of genotyped samples were shown positive.
Two Adenovirus type samples (40 & 41) were tested with the **ichroma™ Rota/Adeno**. The Enteric Adenovirus (40&41) samples were shown positive.

- **Analytical Specificity**
- **Cross-reactivity**
There was no false positive result from 9 species virus samples and 24 species bacteria samples containing potentially cross-reactive substances with the **ichroma™ Rota/Adeno** test.

	Virus
#1	Norovirus VLP (GI)
#2	Norovirus VLP (GII)
#3	Enterovirus type 71
#4	Cytomegalovirus
#5	Poliovirus type 1
#6	Coxsackie virus B type 5
#7	Coxsackie virus B type 6
#8	Herpes simplex virus type 1
#9	Herpes simplex virus type 2
	Bacteria
#1	Staphylococcus aureus (ATCC 29213)
#2	Enterococcus faecalis (ATCC 29212)
#3	Escherichia coli (ATCC 25922)
#4	Kleb-siella oxytoca (ATCC 700432)
#5	Pseudomonas aeruginosa (ATCC 27853)

#6	Neisseria gonorrhoeae (ATCC 27853)
#7	Aeromonas hydrophila (KCCM 32586)
#8	Enterobacter cloacae (KCCM 32586)
#9	Vibrio parahaemolyticus (KCCM11965)
#10	Salmonella group B (Clinical isolate from patient)
#11	Salmonella group C (Clinical isolate from patient)
#12	Salmonella group D (Clinical isolate from patient)
#13	Salmonella group E (Clinical isolate from patient)
#14	Shigella group D (Clinical isolate from patient)
#15	Staphylococcus epidermidis (Clinical isolate from patient)
#16	Serratia marcescens (Clinical isolate from patient)
#17	Yersinia enterocolitica (Clinical isolate from patient)
#18	Salmonella typhi (Clinical isolate from patient)
#19	Clostridium difficile (Clinical isolate from patient)
#20	Candida albicans (Clinical isolate from patient)
#21	Candida parapsilosis (Clinical isolate from patient)
#22	Campylobacter spp.
#23	Proteus vulgaris
#24	Proteus mirabilis

- **Interference**

Interferents such as below the ones in the table were added to the test samples at concentrations much higher than their normal physiological levels in human feces. **ichroma™ Rota/Adeno** test results did not show any significant interference with these biomolecules and chemical drugs.

Biomolecule			
#1	Bilirubin	#4	Cholesterol
#2	Hemoglobin	#5	BSA
#3	Triglycerides		
Chemical drug			
#1	Cephradine	#7	amoxicillin
#2	cefuroxime axetil	#8	loperamide oxide
#3	cefpodoxime proxetil	#9	metronidazole
#4	cefixime	#10	ibuprofen
#5	tetracycline HCl	#11	acetaminophen
#6	levofloxacin	#12	barium sulfate

■ **Precision**

- Between Lot

One person tested three different lots of **ichroma™ Rota/Adeno**, ten times at each concentration of the control standard.

- Between person

Three different persons tested one lot of **ichroma™ Rota/Adeno**, five times at each concentration of the control standard.

- Between day

One person tested one lot of **ichroma™ Rota/Adeno** for three days, five times at each concentration of the control standard.

- Between site

One person tested one lot of **ichroma™ Rota/Adeno** at three different sites, five times at each concentration of the control standard.

Sample		Between lot		Between person	
		Positive /Test no.	Positive rate	Positive /Test no.	Positive rate
Rota virus	Negative	0/10	0%	0/5	0%
	Low	10/10	100%	5/5	100%
	Mid	10/10	100%	5/5	100%
	High	10/10	100%	5/5	100%
Adeno virus	Negative	0/10	0%	0/5	0%
	Low	10/10	100%	5/5	100%
	Mid	10/10	100%	5/5	100%
	High	10/10	100%	5/5	100%

Sample		Between day		Between site	
		Positive /Test no.	Positive rate	Positive /Test no.	Positive rate
Rota virus	Negative	0/5	0%	0/5	0%
	Low	5/5	100%	5/5	100%
	Mid	5/5	100%	5/5	100%
	High	5/5	100%	5/5	100%
Adeno virus	Negative	0/5	0%	0/5	0%
	Low	5/5	100%	5/5	100%
	Mid	5/5	100%	5/5	100%
	High	5/5	100%	5/5	100%

■ **Clinical performance**

Rotavirus		Real-time PCR		
		Positive	Negative	Total
ichroma™ Rota/Adeno	Positive	56	3	59
	Negative	1	66	67
	Total	57	69	126

Adenovirus		Real-time PCR		
		Positive	Negative	Total
ichroma™ Rota/Adeno	Positive	19	2	21
	Negative	1	51	52
	Total	20	53	73

- Rotavirus
  - Clinical sensitivity: 98.2 %
  - Clinical specificity: 95.7 %
- Adenovirus
  - Clinical sensitivity: 95 %
  - Clinical specificity: 96.2 %

REFERENCES

1. Rotavirus Methods and Protocols. James Gray et al., Methods in Molecular Medicine., 2000, 6-7 pp.
2. Diarrheagenic pathogens in polymicrobial infections, Brianna Lindsay et al., Emerging infectious disease, 2011, 17:4 606-611
3. Rotavirus and adenovirus frequency among patients with acute gastroenteritis and their relationship to clinical parameters: a retrospective study in Turkey, Asia Pacific Family Medicine, 2009, 8:8 1-8.

Note: Please refer to the table below to identify various symbols.

	Sufficient for <n> tests
	Read instruction for use
	Use by Date
	Batch code
	Catalog number
	Caution
	Manufacturer
	Authorized representative of the European Community
	In vitro diagnostic medical device
	Temperature limit
	Do not reuse
	This product fulfills the requirements of the Directive 98/79/EC on in vitro diagnostic medical devices

For technical assistance, please contact:
Boditech Med Inc.'s Technical Services
Tel: +(82) -33-243-1400
E-mail: sales@boditech.co.kr

 **Boditech Med Inc.**
43, Geodudanji 1-gil, Dongnae-myeon, Chuncheon-si,
Gang-won-do, 24398, Republic of Korea
Tel: +(82) -33-243-1400
Fax: +(82) -33-243-9373
www.boditech.co.kr

 **Obelis s.a**
Bd. Général Wahis 53, 1030 Brussels, Belgium
Tel: +(32) -2-732-59-54
Fax: +(32) -2-732-60-03
E-Mail: mail@obelis.net

CE IVD