CE
IVD

Cholera Ag Rapid Test Kit (Colloidal Gold)

Instruction for Use

Read this instruction carefully before use

A rapid test for the qualitative detection of Cholera antigen in Fecal Specimen.For professional medical institutions use only,Not for self testing.

PRODUCT NAME

Cholera Ag Rapid Test Kit (Colloidal Gold)

SPECIFICATION

25 tests/kit, 5 tests/kit, 1 test/kit

INTENDED USE

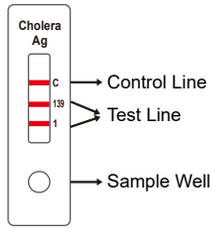
The Cholera Ag Rapid Test is a lateral flow chromatographic immunoassay for the qualitative detection and differentiation of Vibrio Cholerae O139 antigen and O1 antigen in human fecal specimen. It is intended to be used by professionals as a screening test and as an aid in the diagnosis of infection with V. Cholerae. Any reactive specimen with the Cholera Ag Rapid Test must be confirmed with alternative testing method(s) and clinical findings.

SUMMARY AND EXPLANATION THE TEST

Cholera is an acute infectious disease that is characterized by massive loss of body fluids and electrolytes through severe diarrhea. The etiological agent of cholera has been identified as Vibrio cholerea (V. Cholerae), a gram negative bacterium, which is generally transmitted
to humans via contaminated water and food.
The species V. Cholerae is divided into several serogroups on the basis of O antigens. The subgroups O1 and O139 are of special interest because both can cause epidemic and pandemic cholera. It is critical to determine as quickly as possible the presence of V. cholerae O1 and O139 in clinical specimens, water, and food so that appropriate monitoring and effective preventive measures can be undertaken by public health authorities.
The Cholera Ag Rapid Test can be used directly in the field by untrained or minimally skilled personnel and the result is available in less than 10 min, without cumbersome laboratory equipment.

PRINCIPLE

The Cholera Ag Rapid Test is a lateral flow chromatographic immunoassay. The test cassette consists of: 1) a burgundy colored conjugate pad containing monoclonal anti-V. Cholera O1 and O139 antibodies conjugated with colloid gold (O1/O139-antibody conjugates) and rabbit IgG-gold conjugates, 2) a nitrocellulose membrane strip containing two test band (1 and 139 bands) and a control band (C band). The 1 band is pre-coated with monoclonal anti-V. Cholera O1 antibody. The 139 band is pre-coated with monoclonal anti-V. Cholera O139 antibody. The C band is pre-coated with goat anti-mouse IgG antibody.



When an adequate volume of test specimen is applied into the sample well of the test cassette, the specimen migrates by capillary action across the cassette. V. Cholera O1/O139 antigen if present in the specimen will bind to the corresponding O1/O139-antibody gold conjugate. This immunocomplex is then captured on the membrane by the pre-coated anti-V. Cholera O1/O139 antibody, forming a burgundy colored test band, indicating a Cholera O1/O139 positive test result. Absence of the test band suggests a negative result.

The test contains an internal control (C band) which should exhibit a burgundy colored band of the immunocomplex of goat anti-mouse IgG/ mouse IgG-gold conjugate regardless of the color development on the test band. Otherwise, the test result is invalid and the specimen must be retested with another device.

REAGENTS AND MATERIALS PROVIDED

Components	25 tests/kit	5 tests/kit	1 test/kit
Cassettes	25 cassettes with dependent sealed foil pouch	5 cassettes with dependent sealed foil pouch	1 cassette with dependent sealed foil pouch
Specimen vial with buffer	1mL/bottle, 25pcs	1mL/bottle, 5pcs	1mL/bottle, 1pcs
Transfer tube	25 pcs	5 pcs	1 pcs
Package insert	1 pcs	1 pcs	1 pcs

MATERIALS REQUIRED BUT NOT PROVIDED

Timer for timing use

WARNINGS AND PRECAUTIONS

For *in Vitro* Diagnostic Use

- 1.This package insert must be read completely before performing the test. Failure to follow the insert gives inaccurate test results.
- 2.Do not open the sealed pouch, unless ready to conduct the assay.
- 3.Do not use expired devices.
- 4.Bring all reagents to room temperature (15°C-30°C) before use.
- 5.Do not use the components in any other type of test kit as a substitute for the components in this kit.
- 6.Do not use hemolized blood specimen for testing.
- 7.Wear protective clothing and disposable gloves while handling the kit reagents and clinical specimens. Wash hands thoroughly after performing the test.
- 8.Users of this test should follow the US CDC Universal Precautions for prevention of transmission of HIV, HBV and other blood-borne pathogens.
- 9.Do not smoke, drink, or eat in areas where specimens or kit reagents are being handled.
- 10.Dispose of all specimens and materials used to perform the test as biohazardous waste.
- 11.Handle the Negative and Positive Control in the same manner as patient specimens.
- 12.The testing results should be read within 25 minutes after a specimen is applied to the sample well or sample pad of the device. Read result after 25 minutes may give erroneous results.
- 13.Do not perform the test in a room with strong air flow, ie. electric fan or strong air-conditioning.

REAGENT PREPARATION AND STORAGE INSTRUCTIONS

All reagents are ready to use as supplied. Store unused test device unopened at 2°C-30°C. The positive and negative controls should be kept at 2°C-8°C. If stored at 2°C-8°C, ensure that the test device is brought to room temperature before opening. The test device is stable through the expiration date printed on the sealed pouch. Do not freeze the kit or expose the kit over 30°C

SPECIMEN COLLECTION AND HANDLING

Consider any materials of human origin as infectious and handle them using standard biosafety procedures.

Procedure A: Solid stool samples

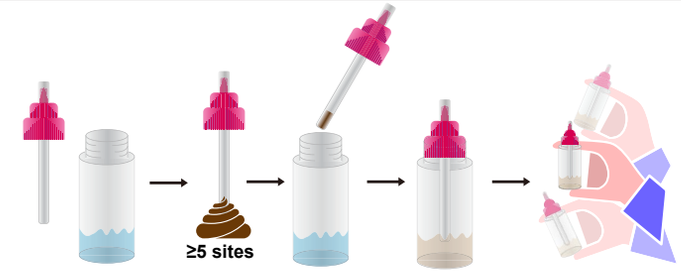
Step 1: Collect a random stool sample in a clean, dry receptacle.

Step 2: Open the stool collection device by unscrewing the top and use the collection stick to randomly pierce the stool sample in at least five different sites. Do not scoop stool sample as this may lead to an invalid test result.

Step 3: Ensure stool sample is only in the grooves of the collection stick. **Excess stool sample may lead to an invalid test result.**

Step 4: Replace the collection stick and tighten securely to close the stool collection device.

Step 5: Shake the stool collection device vigorously.



Note: Specimens extracted may be stored at 2-8°C for up to 3 days. If longer storage is required, freezing at ≤-20°C is recommended.

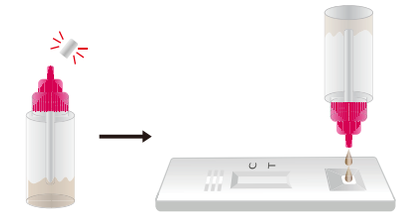
ASSAY PROCEDURE

Step 1: Bring the specimen and test components to room temperature if refrigerated or frozen.

Step 2: When ready to test, open the pouch at the notch and remove the test device. Place the test device on a clean, flat surface.

Step 3: Shake the stool collection device vigorously to ensure an effective liquid suspension.

Step 4: Position the stool collection device upright and twist off the dispenser cap. Holding the stool collection device vertically, dispense 2 drops of the solution (85-95uL) into the sample well of the test device. Do not overload sample.



Step 5: Set up timer.

Step 6: Results can be read after 15 minutes. Positive results can be visible in as short as 1 minute.

Don't read results after 30 minutes.To avoid confusion, discard the test device after interpreting the result.

QUALITY CONTROL

Internal Control: This test contains a built-in control feature, the C band. The C line develops after adding specimen and sample diluent. Otherwise, review the whole procedure and repeat test with a new device.

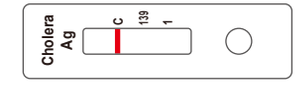
External Control: Good Laboratory Practice recommends using the external controls, positive and negative, to assure the proper performing of the assay, in particularly, under the following circumstances:

a. New operator uses the kit, prior to performing testing of specimens.
b.A new lot of test kit is used.
c.A new shipment of kits is used.
d.The temperature used during storage of the kit fall outside of 2 -30℃ .
e.The temperature of the test area falls outside of 15℃ -30℃ .

INTERPRETATION OF ASSAY RESULT

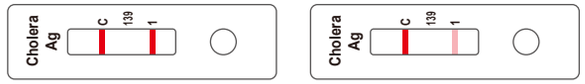
Negative Control

If only the C band is developed, the test indicates that no detectable V. Cholera antigen is present in the specimen. The result is non-reactive.



Positive Control:

O1 Positive: If both C and 1 bands are developed, the test indicates for the presence of V. CholeraO1 antigen in the specimen. The result is V. Cholera O1 reactive.



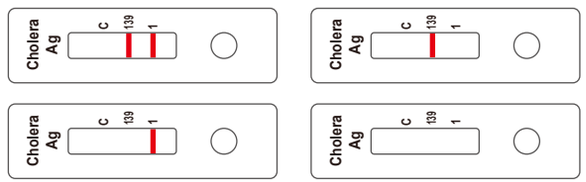
O139 Positive: If both C and 139 bands are developed, the test indicates for the presence of V. Cholera O139 antigen in the specimen. The result is V. Cholera O139 reactive.

O1/O139 Positive: In addition to the presence of C band, both 1 and 139 bands are developed, the test indicates for the presence of V. Cholera O1 antigen and V. Cholera O139 antigen. The result is both V. Cholera O1 and O139 reactive.

Samples with positive or reactive results should be confirmed with alternative testing method(s) such as ELISA or PCR and clinical findings before a diagnostic decision is made.

INVALID:

If no C band is developed, the assay is invalid regardless of color development on the test band as indicated below. Repeat the assay with a new device.



PERFORMANCE CHARACTERISTICS

1. Sensitivity and Specificity:

A clinical study was performed with 236 patient fecal samples using a commercial Cholera Rapid Test as a reference test. Comparison for all subjects is shown in the table below:

	Cholera Ag Rapid Test (V. cholera O1)		
Reference	Positive	Negative	Total
V. cholera O1 Positive	65	2	67
V. cholera O1 Negative	9	160	169
Total	74	162	236
Relative Sensitivity 97.0%, Relative Specificity 94.7%, Overall Agreement 95.3%.			

	Cholera Ag Rapid Test (V. cholera O139)		
Reference	Positive	Negative	Total
V. cholera O139 Positive	59	2	61
V. cholera O139 Negative	10	165	175
Total	69	167	236
Relative Sensitivity 96.7%, Relative Specificity 94.3%, Overall Agreement 94.9%			

2. Limit of Detection（LOD）

The limit of detection of the Cholera Ag Rapid test was determined using suspension of V. Cholera O1 and O139 cultures. Serial dilutions (in triplicate) were made and the number of colony forming units (cfu) was calculated by plating the bacteria on TCBS (thiosulfate citrate bile salts sucrose) agar plate. The limit of detection is defined as the number of bacteria in a specimen that gives 95% detection rate (detected 95% of the time). Cholera Ag Rapid test consistently detects suspensions containing at least 105 cfu/ml V. Cholera O1 and/or at least 105 cfu/ml V. Cholera O139.

3. Precision:

Three specimens composed of strong, weak and negative cholera antigen were tested against 10 devices at each condition. All of the devices identified the specimens correctly with the same band intensity at each given condition.

4. Cross-reactivity

The cross-reactivity of Cholera Ag Rapid test with other organisms was assessed using suspension of cultures of the following organisms at a concentration of 108 cfu/ml. None of the organisms show any cross-reactivity in the test:

Name	Name	Name
Escherihia coli	Salmonella typhi	Shigella dysenterae type 1
Pseudomonas aeruginosa	Vibrio damsela	Vibrio vulnificus
Serratia marcescens	Vibrio hollisae	Vibrio harveyi
Vibrio cincinnatiensis	Vibrio ordalii	

5. Interference

The effects of multiple elevated analytes on the test performance of Cholera Ag Rapid test were assessed:

Analytes	Concentration tested
Uric acid	600 to 940 µmol/L
Hemoglobin	18 to 20 gm/dl
Total bilirubin	34 to 65 µmol/L
Triglycerides	200 to 500 mg/dl

The results indicated that none of the above conditions interfered with Cholera Ag Rapid test.

TEST LIMITATIONS

1.The Assay Procedure and the Interpretation of Assay Results must be followed closely when testing the presence of V.Cholera antigen in human fecal specimen from individual subject. Failure to follow the procedure may give inaccurate results.
2.The OnSite Cholera Ag Rapid Test is limited to the qualitative detection of V. Cholera O1 and O139 antigen in human fecal specimen. The intensity of the test band does not have linear correlation with the antigen concentration of the specimen.
3.A nonreactive result for an individual subject indicates absence of detectable V. Cholera antigen. However, a nonreactive test result does not preclude the possibility of exposure to or infection with V. Cholera bacteria.
4.A nonreactive result can occur if the quantity of the V. Cholera antigen present in the specimen is below the detection limits of the assay or the antigen that are detected are not present in the fecal specimen picked by the sample extraction tube device.
5.If the symptom persists, while the result from OnSite Cholera Ag Rapid Test is non-reactive, it is recommended to re-sample the patient or test with an alternative test method.
6.Some specimens containing unusually high titer of heterophile antibodies or rheumatoid factor may affect expected results.
7.The results obtained with this test should only be interpreted in conjunction with otherdiagnostic procedures and clinical findings.

CAUTION

1.This product is used for in vitro diagnosis only.
2.Must strictly follow the instructions for operation and interpretation of the results.
3.The product is qualitatively tested, and the result cannot be used as a quantitative basis.should be tested using reagents within the validity period.
3.The cassetes, collectors,droppers,and tubes are for single person one-time use, cannot be reused.
4.Because the sample titer is different, the red lines of the test line will show different shades of color, all of which indicate positive results. The depth of the test line color cannot be used as the basis for determining the antibody titer in the sample.
5.The samples stored at low temperature should be balanced to room temperature and fully mixed before testing.
6.Samples and waste must be treated as a potential source of infection and the desiccant in the foil bag is not edible.

SYMBOLS

Symbol	Used For	Symbol	Used For
	Use-by date		Consult instructions for use
	Batch code		In vitro diagnostic medical device
	Temperature limit		Manufacturer
	Please don't reuse it		Keep away from sunlight
	Don’t use the product when the package is damaged		Keep dry
	Date of manufacture		Tests per kit
	CE Mark		Biological Risks
	Authorized representative in the European Community		

BASIC INFORMATION

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