OnSite® Troponin I Combo Rapid Test

REF R3002C CE

Instructions for Use

INTENDED USE

The *OnSite* Troponin I Combo Rapid Test is a lateral flow chromatographic immunoassay for the qualitative detection of cardiac Tropnin I (cTnI) and its complex in human serum, plasma or whole blood at a level equal to or higher than 1 ng/mL. It is intended to be used by healthcare professionals as an aid in the diagnosis of acute myocardial infarction (AMI).

Any use or interpretation of this preliminary test result must also rely on other clinical findings and the professional judgment of health care providers. Alternative test method(s) should be considered to confirm the test result obtained by this device.

SUMMARY AND EXPLANATION OF THE TEST

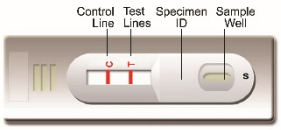
Cardiac Troponin I (cTnI) is a cardiac muscle protein with a molecular weight of 22.5 kilodalton. Together with troponin T (TnT) and troponin C (TnC), TnI forms a troponin complex in the heart to play a fundamental role in the transmission of intracellular calcium signals actin-myosin interaction[1]. The human cTnI has additional amino acid residues in its N-terminal that does not exist in the skeletal forms thus making cTnI a specific cardiac marker[1-3].

Normally the level of cTnI in the blood is very low. cTnI is released into the blood stream in forms of free cTnI and cTnI-C-T complex at 4-6 hours after myocardial cell damage[2,4,5]. The elevated level of cTnI could be as high as 50 ng/mL during 60-80 hours after AMI and remains detectable for up to 10-14 days post AMI[2,4,5]. Therefore, circulating cTnI is a specific and sensitive marker for AMI.

The *OnSite* Troponin I Combo Rapid Test is intended to detect elevated troponin I in human serum, plasma or whole blood. The test can be performed within 15 minutes by minimally skilled personnel without the need for sophisticated laboratory equipment.

TEST PRINCIPLE

The *OnSite* Troponin I Combo Rapid Test is a lateral flow chromatographic immunoassay. The test cassette consists of: 1) a colored conjugate pad containing monoclonal anti-cTnI antibody conjugated with colloidal gold (antibody conjugates), 2) a nitrocellulose membrane strip containing a test line (T line) and a control line (C line). The T line is pre-coated with another anti-cTnI antibody, and the C line is pre-coated with a control line antibody.



When an adequate volume of test specimen is dispensed into the sample well of the cassette, the specimen migrates by capillary action across the cassette. Elevated cTnI, if present in the specimen, will bind to the antibody conjugates. The immunocomplex is then captured on the membrane by the pre-coated anti-cTnI antibodies forming a colored T line, indicating a cTnI positive test result.

Absence of the T line suggests a negative result. The test contains an internal control (C line) which should exhibit a colored line of the immunocomplex of the control line antibodies conjugate regardless of the presence of cTnI in the specimen. If the C line does not develop, the test result is invalid and the specimen must be retested with another device.

REAGENTS AND MATERIALS PROVIDED

1. Individually sealed foil pouches containing:
   a. One cassette device
   b. One desiccant
2. Plastic droppers
3. Sample diluent (REF SB-R3002, 1 bottle, 5 mL)
4. Instructions for Use

MATERIALS MAY BE REQUIRED BUT NOT PROVIDED

1. Positive Control
2. Negative Control

MATERIALS REQUIRED BUT NOT PROVIDED

1. Clock or timer

WARNINGS AND PRECAUTIONS

For *in Vitro* Diagnostic Use

1. Read these Instructions for Use completely before performing the test. Failure to follow the instructions could lead to 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 components from any other type of test kit as a substitute for the components in this kit.
6. Do not use hemolized blood specimens 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 controls in the same manner as patient specimens.
12. The test results should be read within 15 minutes after a specimen is applied to the sample well or sample pad of the device. Reading the result after 15 minutes may give erroneous results.
13. Do not perform the test in a room with strong air flow, i.e. an electric fan or strong air-conditioning.

REAGENT PREPARATION AND STORAGE INSTRUCTIONS

All reagents are ready to use as supplied. Store unused test devices unopened at 2-30°C. If stored at 2-8°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 or expose the kit to temperatures above 30°C.

SPECIMEN COLLECTION AND HANDLING

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

Plasma/Serum

Step 1: Collect blood specimen into collection tube containing EDTA, citrate or heparin for plasma or collection tube containing no anticoagulants for serum by venipuncture.

Step 2: To make plasma specimen, centrifuge collected specimens and carefully withdraw the plasma into a new pre-labeled tube.

Step 3: To make serum specimen, allow blood to clot, then centrifuge collected specimens and carefully withdraw the serum into a new pre-labeled tube.

Test specimens as soon as possible after collecting. Store specimens at 2°C-8°C if not tested immediately. Specimens can be stored at 2-8°C for up to 5 days, and should be frozen at -20°C for longer storage.

Avoid multiple freeze-thaw cycles. Prior to testing, bring frozen specimens to room temperature slowly and mix gently. Specimens containing visible particulate matter should be clarified by centrifugation before testing.

Do not use samples demonstrating gross lipemia, gross hemolysis or turbidity in order to avoid interference on result interpretation.

Whole Blood

Collect whole blood by either finger tip puncture or by veinpuncture into collection tube containing EDTA, citrate or heparin for plasma. Do not use any hemolized blood for testing.

Whole blood specimens should be stored in refrigeration (2°C-8°C) if not tested immediately. The specimens must be tested within 24 hours of collection.

ASSAY PROCEDURE

Step 1: Bring the specimen and test components to room temperature if refrigerated or frozen. Once thawed, mix the specimen well prior to performing the assay.

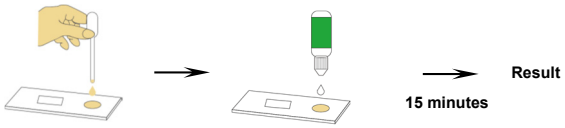
Step 2: When ready to test, open the pouch at the notch and remove device. Place the test device on a clean, flat surface.

Step 3: Be sure to label the device with specimen's ID number.

Step 4: **For serum/plasma specimens:**

Fill the pipette dropper with the specimen. Holding the dropper vertically, dispense 1 drop of serum/plasma (about 30-45 µL) into the sample well making sure that there are no air bubbles.

Then add 1 drop of sample diluent (about 35-50 µL) immediately.

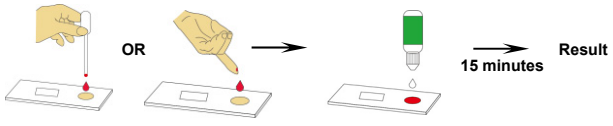


1 drop of serum/plasma **1 drop of sample diluent**

For whole blood specimens:

Fill the pipette dropper with the specimen. Holding the dropper vertically, dispense 1 drop of whole blood (about 40-50 µL) into the sample well making sure that there are no air bubbles.

Then add 1 drop of sample diluent (about 35-50 µL) immediately.



1 drop of whole blood **1 drop of sample diluent**

Step 5: Set up timer.

Step 6: Read results at 15 minutes. Positive results may be visible in as short as 1 minute. Negative results must be confirmed at the end of the 15 minutes only. ***Any results interpreted outside of the 15 minute window should be considered invalid and must be repeated. Discard used devices after interpreting the result following local requirements governing the disposal of devices.***

QUALITY CONTROL

1. **Internal Control**: This test contains a built-in control feature, the C line. The C line develops after adding the specimen and the sample diluent. If the C line does not develop, review the whole procedure and repeat the test with a new device.
2. **External Control**: Good Laboratory Practice recommends using external controls, positive and negative, to assure the proper performance of the assay, particularly under the following circumstances:

a. A new operator uses the kit, prior to performing the testing of the specimens.
b. A new lot of test kits is used.
c. A new shipment of kits is used.
d. The temperature used during storage of the kits fall outside of 2°C-30°C.
e. The temperature of the test area falls outside of 15°C-30°C.
f. To verify a higher than expected frequency of positive or negative results.
g. To investigate the cause of repeated invalid results.

INTERPRETATION OF ASSAY RESULT

1. **NEGATIVE RESULT:** If only the C line is present, the absence of any color in the test line indicates that no detectable cTnI is present in the specimen. The result is negative or non-reactive.

2. **POSITIVE RESULT:** If both C and T lines develop, the test indicates that the level of cTnI is equal to or greater than 1 ng/mL. The result is positive or reactive.

Specimens with positive results should be confirmed with alternative testing method(s) and clinical findings before a diagnosis is made.

3. **INVALID:** If no C line develops, the assay is invalid regardless of color development on the T line as indicated below. Repeat the assay with a new device.

PERFORMANCE CHARACTERISTICS

1. **Clinical performance**

A total of 770 specimens were collected from susceptible subjects and tested by *OnSite* Troponin I Combo Rapid Test and a commercial chemiluminescence test as reference. Comparison for all subjects is showed in the following table.

	OnSite Troponin I Combo Rapid Test		
CLIA Reference	**Positive**	**Negative**	**Total**
Positive	158	7	165
Negative	2	603	605
Total	160	610	770

Relative Sensitivity: 95.8% (95% CI: 91.5-97.9%)
Relative Specificity: 99.7% (95% CI: 98.8-99.9%)
Overall Agreement: 98.8% (95% CI: 97.8-99.4%)

2. **Analytical sensitivity**

The *OnSite* Troponin I Combo Rapid Test can detect cTnI in serum, plasma or whole blood with concentration of 1.0 ng/mL or greater.

3. **Hook effect**

No hook effect was found with cTnI concentration up to 3,521 µg/mL.

4. **Cross-Reactivity**

4.1 To evaluate potential cross-reactivity due to structurally related molecules, the following potentially cross-reacting compounds were spiked into cTnI negative serum samples. The results demonstrate that, at the concentrations tested, the compounds studied do not affect the performance of the *OnSite* Troponin I Combo Rapid Test.

List of potentially cross-reacting compounds and concentrations tested:

1. Skeletal muscle TnI	10 µg/mL	3. Cardiac muscle Myosin	20 µg/mL
2. Cardiac muscle TnT	2,000 ng/mL		

4.2 No false positive results were observed from the following special immunological conditions, respectively:

1. CMV IgM (n=3)
2. HAMA (n=10)
3. HBsAg (n=10)
4. HBsAb (n=10)
5. HBeAg (n=10)
6. HBeAb (n=10)
7. HBcAb (n=10)
8. HCV (n=10)
9. HIV (n=10)
10. H. pylori (n=3)
11. Mono (n=3)
12. Rubella IgM (n=3)
13. Syphilis (n=10)

5. **Interference**

Common substances (such as pain and fever medication and blood components) may affect the performance of the *OnSite* Troponin I Combo Rapid Test. This was studied by spiking these substances into two levels of controls (negative and positive). The results demonstrate, at the concentrations tested, the substances studied do not affect the performance of the *OnSite* Troponin I Combo Rapid Test.

List of potentially interfering substances and concentrations tested:

1. Acetaminophen	20 mg/dl	7. Cholesterol	800 mg/dl
2. Acetylsalicylic acid	20 mg/dl	8. Creatinine	200 mg/dl
3. Albumin	10.5 g/dl	9. Gentistic acid	20 mg/dl
4. Ascorbic acid	20 mg/dl	11. Hemoglobin	1000 mg/dl
5. Bilirubin	1,000 mg/dl	12. Oxilic acid	600 mg/dl
6. Caffeine	20 mg/dl	13. Triglycerides	1,600 mg/dl

STANDARDIZATION

The *OnSite* Troponin I Combo Rapid Test has been calibrated against United States of America, National Institute of Standards & Technology, Standard Reference Material 2921.

LIMITATIONS OF TEST

1. The Assay Procedure and the Interpretation of Assay Result sections must be followed closely when testing for the presence of elevated Troponin I in serum, plasma or whole blood from individual subjects. Failure to follow the procedure may give inaccurate results.
2. The *OnSite* Troponin I Combo Rapid Test is limited to the qualitative detection of Troponin I by professionals in human serum, plasma or whole blood. The intensity of the test line does not have a linear correlation with the cTnI level in the specimen.
3. A negative result for an individual subject indicates the level of cTnI is not detectable. However, a negative test result does not preclude the possibility of AMI.
4. A negative result can occur if the level of cTnI present in the specimen is below the detection limits of the assay or cTnI that is detected is not present during the stage of AMI in which a sample is collected.
5. AMI progresses rapidly. If symptoms are suspicious or persist while the result from the *OnSite* Troponin I Combo Rapid Test is negative or non-reactive, it is recommended to test with an alternative test method, such as ECG.
6. Unusually high titers of heterophile antibodies or rheumatoid factor present in specimens may affect the expected results.
7. The results obtained with this test should only be interpreted in conjunction with other diagnostic procedures and clinical findings.

REFERENCES

1. Adams JE, Bodor GS, Dávila-Román VG, *et al.* Cardiac troponin I. A marker with high specificity for cardiac injury. Circulation. 1993;88,101-6.
2. Adams JE, Sicard GA, Allen BT, *et al.* Diagnosis of perioperative myocardial infarction with measurement of cardiac troponin I. N. Eng. J. Med. 1994;330, 670-4.
3. Bodor GS, Porterfield D, Voss EM, *et al.* Cardiac troponin-I is not expressed in fetal and healthy or diseased adult human skeletal muscle tissue. Clin. Chem. 1995;41,1710-5.
4. Brogan GX Jr, Hollander JE, McCuskey CF, *et al.* Evaluation of a new assay for cardiac troponin I vs creatine kinase-MB for the diagnosis of acute myocardial infarction. Biochemical Markers for Acute Myocardial Ischemia (BAMI) Study Group. Academic Emerg. Med. 1997;4, 6-12.
5. Tucker JF, Collins RA, Anderson AJ, *et al.* Early diagnostic efficiency of cardiac troponin I and Troponin T for acute myocardial infarction. Acad Emerg Med. 1997;4, 13-21.

Index of CE Symbols

	Consult instructions for use	IVD	For *in vitro* diagnostic use only		Use by
REF	Catalog #	LOT	Lot Number	N	Tests per kit
2°C 30°C	Store between 2-30°C	EC REP	Authorized Representative		Do not reuse
	Manufacturer		Date of manufacture		

CTK Biotech, Inc.
13855 Stowe Drive
Poway, CA 92064, USA
Tel: 858-457-8698
Fax: 858-535-1739
E-mail: info@ctkbiotech.com

EC REP **MDSS GmbH**
Schiffgraben 41
30175 Hannover, Germany

PI-R3002C Rev. E2.1
Date released: 2020-07-17
English version

For Export Only, Not For Re-sale in the USA