CE

Troponin I

en

In-vitro diagnostic test for the qualitative detection of human cardiac Troponin I in whole blood, serum or plasma

RAPU04A097

IN VITRO DIAGNOSTIC

DIAsource ImmunoAssays SA - Rue de l'Industrie 8, B-1400 Nivelles, Belgium - Tel: +32 67 88 99 99 - Fax : +32 67 88 99 96

INTENDED USE

The **DIAsource Troponin I Test** is a rapid one-step chromatographic immunoassay for the qualitative detection of human cardiac Troponin I in whole blood, serum or plasma as an aid in the diagnosis of myocardial infarction (MI).

SUMMARY

Cardiac Troponin I (cTnI) is a protein found in cardiac muscle with a molecular weight of 22.5 kDa.[1] Troponin I is part of a three subunit complex comprising of Troponin T and Troponin C. Along with tropomyosin, this structural complex forms the main component that regulates the calcium sensitive ATPase activity of actomyosin in striated skeletal and cardiac muscle.[2] After cardiac injury occurs, cTnI is released into the blood 4-6 hours after the onset of pain. The release pattern of cTnI is similar to CK-MB, but while CK-MB levels return to normal after 72 hours, cTnI levels remain elevated for 6-10 days, thus providing a longer window of detection for cardiac injury. The high specificity of cTnI measurements for the identification of myocardial damage has been demonstrated in conditions such as the perioperative period, after marathon runs, and blunt chest trauma.[3] cTnI release has also been documented in cardiac conditions other than acute myocardial infarction /AMI) such as unstable angina, congestive heart failure, and ischemic damage due to coronary artery bypass surgery.[4] Because of its high specificity and sensitivity in the myocardial tissue, cTnI has recently become the most preferred biomarker for myocardial infarction.[5]

The **DIAsource Troponin I Test** is a simple test that utilizes a combination of particle conjugated anti-cTnI antibodies and capture reagent to selectively detect cTnI in whole blood, serum or plasma. The minimum detection level is 1.0 ng/mL.

PRINCIPLE

The **DIAsource Troponin I Test** is a qualitative, membrane based immunoassay for the detection of cTnI in whole blood, serum or plasma. The membrane is pre-coated with capture reagent on the test line region of the test. During testing the Troponin I in the whole blood, serum or plasma specimen reacts with two specific anti-cTnI antibodies. One of the antibodies mediates binding to the capture reagent, the other antibody is color labelled.

The mixture migrates upward on the membrane by capillary action. In the test line region the cTnI-antibody complex is captured by the immobilised capture reagent so that a red line is generated. The presence of a red line in the test line region indicates a positive result.

If the sampled does not contain cTnI no line will form in the test result line region indicating a negative result.

In addition a red line must form in the control line region (C) independent of the cTnI concentration in the sample. The control line serves as a procedural control and indicates that sufficient volume of specimen has been added and membrane wicking has occurred.

PRECAUTIONS

- For single professional in vitro diagnostic use only.
- Do not use after the expiration date.
- The test device should remain in the sealed pouch until ready to use.
- Do not eat, drink or smoke in the area where the specimens or kits are handled.
- Do not use test of pouch has been damaged.
- Dispose the used test device according to the local regulations.
- Humidity and high temperature can adversely affect results.
- All specimen might be potentially infectious. Proper handling and disposal methods should be established. Wear protective clothing such as laboratory coats, disposable gloves and eye protection when specimens are being tested.

STORAGE AND STABILITY

Store as packaged in the sealed pouch between 2-30°C / 36-86 °. The **DIAsource Troponin I Test** is stable until the expiration date printed on the sealed pouch. The test device must remain in the sealed pouch until use.

- Do not freeze
- Do not use beyond the expiration date

SPECIMEN COLLECTION AND PREPARATION

The **DIAsource Troponin I Test** can be performed using whole blood (from venipuncture or fingertip), serum or plasma.

Whole blood from fingertip:

- Wash the hand of the patient with soap and warm water or clean the puncture site thoroughly with alcohol.
- Massage the hand in direction of the fingertip of the middle finger or ring finger without touching the puncture.
- Prick the fingertip with a sterile lancet. Wipe the first drop of blood.
- Rub the hand from the wrist to the palm and to the finger to form a round drop at puncture.

Collection of whole blood from fingertip **using a capillary**:

- Take a blood sample filling the capillary with 120 µl of the sample. Avoid air pockets.
- Put the dispensary bulb at the top of the capillary and press it to dribble blood sample in the specimen well (S) of the test device.

Dispensary of whole blood from fingertip by **hanging drops**:

- Position the finger of the patient exactly above the specimen well (S) of the test device.
- Drop 2-(3) hanging drops of whole blood from the puncture of the finger into the specimen well (S). The finger of the patient can be moved over the specimen well so that the drop has contact with the well. A direct contact of the finger and the specimen well should be avoided.

General comments

- Separate serum or plasma from blood as soon as possible to avoid hemolysis.
- Heparin, EDTA or citrate blood can be used for the plasma extraction.
- Use only clear, non-hemolyzed specimens.
- Testing should be performed immediately after specimen collection. Do not leave the specimens at room temperature for prolonged periods. Serum and plasma specimens may be stored at 2-8° C for up to 3 days. For long-term storage, specimens should be kept below -20°C. Whole blood collected by venipuncture should be stored at 2-8°C if the test is to be run within 2 days of collection. Do not freeze whole blood specimens. Whole blood collected by fingertip should be tested immediately.
- Bring specimens to room temperature prior to testing. Frozen specimens must be completely thawed and mixed well prior to testing. Specimens should not be frozen and thawed repeatedly.
- If specimens are to be shipped, they should be packed in compliance with local regulations covering the transportation of etiologic agents.

MATERIALS PROVIDED

- CARD
  20 DIAsource Troponin I Test devices
- PIPETTE
  20 Disposable pipettes (inside pouch)
- Package insert

MATERIALS REQUIRED BUT NOT PROVIDED

- Tubes for taking blood samples
- Centrifuge (for plasma / serum)
- Heparinised capillaries and dispensary bulb (only for whole blood from fingertip)
- Lancets (only for whole blood from fingertip)
- Timer

TESTING PROCEDURE

1 Allow the **DIAsource Troponin I Test** device, specimen, and/or controls to equilibrate to room temperature (15-30°C / 59-86 °F) prior to testing.

2 Remove the test device from the sealed pouch and use it as soon as possible. Best results will be obtained if the assay is performed within one

hour. Place the **DIAsource Troponin I Test** device on a clean and level surface.

3 **Serum or plasma or venipuncture blood**

Hold the provided pipette vertically and transfer **3 drops of serum or plasma** (ca. 120 µl) into the round specimen well (S) of the **DIAsource Troponin I Test**

Venipuncture Whole Blood

Hold the provided pipette **vertically** and transfer **3 drops (ca. 120 µl) of whole blood** into the round specimen well (S) of the **DIAsource Troponin I Test**.

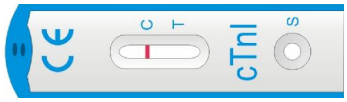
Fingertip Whole Blood

Transfer **2-(3) hanging drops of whole blood from the fingertip** puncture / or approximately **120 µl whole blood** from the heparinised capillary into the round specimen well (S) of **DIAsource Troponin I Test**.

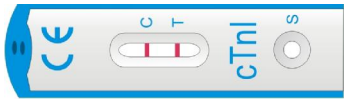
4 Start the timer. Wait for the red line(s) to appear. The result should be read **at 10 minutes**. Do not read results after more than 20 minutes.

INTERPRETATION OF RESULTS

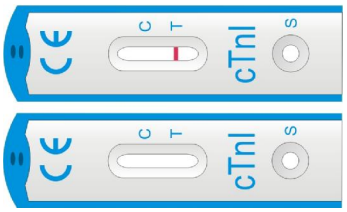
Negative:
One red line appears in the control line region (C).
No apparent red line appears in the test line region (T).



Positive*:
Two distinct red lines appear. One line forms in the control line region (C) and another line forms in the test line region (T).



Invalid:
The control line (C) is not formed. In this case the result is invalid even if the test result is visible**.** Insufficient specimen volume or incorrect procedural techniques are the most likely reasons for control line failure. Review the test procedure and repeat the assay with a new test device. If the problem persists, discontinue using the test kit immediately and contact you local distributor.



*Note: The intensity of the red color in the test line region (T) will vary depending on the concentration of cTnI present in the specimen. Therefore, also faint reddish test result lines (T) should be considered positive.

QUALITY CONTROL

As internal procedural control the **DIAsource Troponin I Test** includes the control line. It is only formed if sufficient specimen volume has been added and the chromatography has been finished successfully. Control standards are not supplied with this kit; yet, we recommend that positive and negative controls should be tested as a good laboratory practice to confirm the test procedure and to verify proper test performance.

LIMITATIONS

1 The **DIAsource Troponin I Test** is an in vitro diagnostic device for professional use only. The test should be used for the detection of cTnI in whole blood, serum of plasma specimens. The **DIAsource Troponin I Test** only allows a qualitative detection of cTnI; quantitative determinations cannot be made with this test.

2 The **DIAsource Troponin I Test** will only indicate the presence of cTnI in the specimen and should not be used as the sole criteria for the diagnosis of myocardial infarction.

3 Due to the heterogeneity of commercially available standard materials the sensitivity of the assay might vary slightly with different standard preparations However, a sensitivity of 1 ng/ml is always reached. Concentrations below 0.5 ng cTnI/ml cannot be detected with the **DIAsource Troponin I Test**.

4 As with all diagnostic tests, all results must be interpreted together with other clinical information available to the physician. *A negative result does not exclude the possibility of myocardial infarction at any time*.

5 Some specimens containing unusually high titres of heterophile antibodies or rheumatic factor (RF) may affect the test result. Even if the test results are positive, further clinical evaluation should be considered with other clinical information available to the physician

6 It is possible that the test does not yield any results if whole blood specimens have a high viscosity or if the whole blood specimens have been stored for more than 2 days. In this case the test should be repeated with a new test card using a plasma or serum specimen of the same patient.

TEST PERFORMANCE

The performance of the **DIAsource Troponin I Test** has been compared with a commercially available test accredited for the sale in the EU of another manufacturer. The results of the study demonstrated an overall accuracy of >99 % for the tested specimens. Specimen without cTnI consistently yielded negative results with both test. Specimen with cTnI-concentrations above the detection limit showed positive results with both assays.

Intra- and inter-lot-variance

The intra- and inter-lot-variance has been determined for 3 independent lots. cTnI specimen at concentrations of 0 ng/ml, 5 ng/ml, 10 ng/ml, 20 ng/ml, and 40 ng/ml were tested in a 10fold determination with each lot. The specimens were correctly identified >99% of the time.

Reproducibility

The reproducibility has been determined by 3 independent assays on the same five specimens: 0 ng/ml, 5 ng/ml, 10 ng/ml, 20 ng/ml, and 40 ng/ml cTnI. Three different lots of the **DIAsource Troponin I Test** have been tested over a 3-month period using these specimens. The specimens were correctly identified >99% of the time.

Cross-Reactivity

Sera containing known amounts of cTnI antigen have been tested with 10,000 ng/ml Skeletal Troponin I, 2,000 ng/ml Troponin T, and 20,000 ng/ml Cardiac Myosin. No cross-reactivity was observed, indicating that **DIAsource Troponin I Test** has a high degree of specificity for cardiac Troponin I (cTnI).

The **DIAsource Troponin I Test** has been tested and no interference was observed in specimens containing 110 µg/ml human albumin, 6 mg/ml bilirubin, 1 mg/ml haemoglobin, 100 µg/ml cholesterol and 10 mg/ml triglycerides.

The following drugs have also been tested using **DIAsource Troponin I Test.** No interference was observed at a concentration of 50 µg/ml.

Acetaminophen	Captopril	Hydrochloride	Pentoxifyline
Acetylsalicylic acid	Chloramphanicol	Furosemide	Phenobarbital
Anisodamine	Chlordiazepoxide	Hydrochlorothiazide	Quinine
Ascorbin Acid	Cilazapril	Isosorbide Mononitrat	Ramipril
Atenolol	Diclofenac	Labetalol	DL-Tyrosine
Atorvastatin	Digoxin	Metoprolol Tartrate	Trimethoprim
Calcium	Erythromycin	Moracizine	Verapamil
Bisoprolol Fumerate	Felodipine	Nifedipine	
Caffeine	Flunarizine	Oxazepam	

BIBLIOGRAPHIE

1. Adams, et al. *Biochemical markers of myocardial injury*, Immunoassay Circulation 88: 750-763 (1993).
2. Mehegan JP, Tobacman LS, *Cooperative interaction between troponin molecules bound to the cardiac thin filament*. J. Biol. Chem. 266:966 (1991)
3. Adams, et al. *Diagnosis of Perioperative myocardial infarction with measurements of cardiac troponin* I. N.Eng.J. Med 330:670 (1994).
4. Hossein-Nia M, et al. *Cardiac troponin / release in heart transplantation*. Ann.Thorac. Surg. 61 : 227 (1996).
5. Joint European Society of Cardiology/American College of Cardiology: J.Am. Coll. Cardio., 36(3), „Myocardnial Infarction Redefined," 2000.

SYMBOLS

Consult instructions for use	Manufacturer
Storage temperature	Contains sufficient for n tests
Use by	IVD In vitro diagnostic medical device
LOT Batch code	CARD Card Test
REF Catalogue number	For single use only
PIPETTE Pipette	

Available packaging size: 20 test devices

Revision date : 2009-06-04