

INTEX®-Troponin I Test

Instructions for use

Rapid test for the detection of cardiac Troponin I

1 INTENDED USE

The INTEX® 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).

Cardiac Troponin I (cTnI) is a protein found in cardiac muscle with a molecular weight of 22.5 kDa¹. 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 INTEX® 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.

2 TEST PRINCIPLE

The INTEX® 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 colour 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 sample 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.

3 MATERIALS PROVIDED

- INTEX® Troponin I Test
- Disposable pipettes (inside pouch)
- Dropper vial with buffer

- Instruction sheet

4 MATERIALS REQUIRED BUT NOT PROVIDED

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

5 STORAGE AND STABILITY

Store as packaged in the sealed pouch at room temperature (2-30°C / 36-86 °F). The INTEX[®] 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!

6 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 specimens 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.

7 SPECIMEN COLLECTION AND PREPARATION

The INTEX[®] 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.

8 TEST PROCEDURE AND RESULTS

PROCEDURE

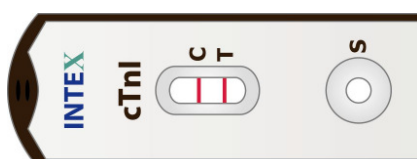
- 1 Allow the INTEX[®] Troponin I Test device, specimen, and/or controls to equilibrate to room temperature (15-30°C / 59-86 °F) prior to testing.
- 2 Remove the test device from the sealed pouch and use it as soon as possible. Best results will be obtained if the assay is performed within one hour. Place the INTEX[®] Troponin I Test device on a clean and level surface.
- 3 **A. 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 INTEX[®] Troponin I Test. Start the timer.
- B. 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 INTEX[®] Troponin I Test. Add **1 drop buffer** and start the timer.
- C. 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 INTEX[®] Troponin I Test. Add **1 drop buffer** and start the timer.
- 4 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.

RESULTS

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).

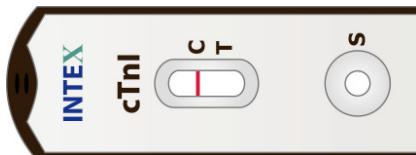


NOTE: The intensity of the red colour in the test line region (T) will vary depending on the concentration of cTnI present in the specimen. Therefore, also faint reddish test result lines (T) should be considered positive.

Negative:

One red line appears in the control line region (C).

No apparent red line appears in the test line region (T). Negative results should be confirmed after 20 minutes



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 your local distributor.



9 QUALITY CONTROL

As internal procedural control the INTEX[®] 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.

10 LIMITATIONS

- 1 The INTEX[®] Troponin I Test is for professional in vitro diagnostic use, and should only be used for the qualitative detection of cardiac Troponin I. No meaning should be inferred from the colour intensity or width of any apparent lines.
- 2 The INTEX[®] 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 If the test result is negative and clinical symptoms persist, additional testing using other clinical methods is recommended. The minimum detection limit of the assay is 1.0 ng/ml of cTnI in specimens. Thus, a negative result does not at anytime rule out the existence of Troponin I in blood, because the protein concentration may be below the minimum detection level of the test. Please keep in mind that the rise of Troponin I takes place several hours after the onset of pain. If the testing takes place too early, cTnI concentrations might still be too low to be detected by the assay. A negative test result does not exclude the possibility of myocardial infarction at any time.
- 4 Like with all diagnostic tests, a confirmed diagnosis should only be made by a physician after all clinical and laboratory findings have been evaluated.

- 5 Some specimens containing unusually high titers of heterophile antibodies or rheumatoid factor (RF) may affect expected results. Even if the test results are positive, further clinical evaluation should be considered with other clinical information available to the physician.
- 6 In rare cases it is possible that the antigen-antibody reaction of the test is inhibited by the presence of auto-antibodies in the patient's blood, which block the binding sites. False negative test results might be the consequence. Please note that this is a general problem with all detection methods based on an antigen-antibody reaction for the detection of proteins.

11 TEST PERFORMANCE

Clinical performance characteristics

The performance of the INTEX[®] Troponin I Test was calculated against a CE approved commercially available CMIA (chemiluminescence microparticle immunoassay). In order to take into account the different cut-offs of the two assays the performance characteristics were calculated in two different ways. For the first calculation the cut-off of 0.3 ng/ml was used. At this concentration results are rated as positive with the CMIA. In the second calculation a cut-off of 1.0 ng/ml was used. This is the declared sensitivity of the INTEX[®] Troponin I test. For the intermediate range (>0.3 but < 1.0 ng/ml) the obtained results (positive or negative) were counted as right results for the Troponin I assay.

	Calculated for a cut-off of ≥ 0.3 ng/ml *	Calculated for a cut-off of ≥ 1.0 ng/ml*
Diagnostic sensitivity	65.2 %	91.5 %
Diagnostic specificity	98.6 %	98.7 %
Positive PDV	95.6 %	95.6 %
Negative PDV	86.2 %	97.5 %
Reproducibility	87.8 %	97.1 %

Analytical performance characteristics

Analytical sensitivity

The minimum detection limit of the assay is 1.0 ng cTnI/ml. For the regular control of the minimum detection limits, standard materials of Hytest are used: (free cTnI). Please note that due to the heterogeneity of commercially available standard materials the sensitivity of the assay might vary slightly with different standard preparations.

Analytical specificity

No cross-reactivity was observed with 10'000 ng/ml Skeletal Troponin , 2'000 ng/ml Troponin T, and 20'000 ng/ml Cardiac Myosin, indicating that INTEX[®] Troponin I Test has a high degree of specificity for cardiac Troponin I (cTnI).

Interference Testing

The INTEX[®] Troponin I Test has been tested and no interference was observed in specimens containing 110 μ g/ml human albumin, 6 mg/ml bilirubin, 1 mg/ml haemoglobin, 100 μ g/ml cholesterol, and 10 mg/ml triglycerides.









Inter-batch reproducibility



Three independent LOT# were tested with various spiked samples of various concentrations and with negative fresh samples. No deviations were observed between the results of the different LOT#. All obtained results matched the expectations.

12 LITERATURE

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), „Myocardial Infarction Redefined,“ 2000.

13 SYMBOLS

	Article number		For single use only
	Lot number		Expiry date
	Storage		Content
	Only for in vitro diagnostics		Instructions for use

INTEX[®]-Troponin I Test		
	5 Test cassettes	TRPI9005
	20 Test cassettes	TRPI9020



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