REF C89243 Σ 2×50 Tests CE

INTENDED USE

The iFlash-Troponin-I assay is a paramagnetic particle chemiluminescent immunoassay (CLIA) for the quantitative determination of Troponin-I in human serum and plasma using the iFlash Immunoassay Analyzer.

SUMMARY AND EXPLANATION

Troponin is a complex of three regulatory proteins (troponin C, Troponin-I, and troponin T) that is integral to muscle contraction in skeletal muscle and cardiac muscle, but not smooth muscle. Troponin-I, often denoted as cTnI, is a part of the troponin complex. It is presented in cardiac muscle tissue by a single isoform with molecular. Multiple reaction monitoring of human cTnI has revealed that there are 14 phosphorylation sites and the pattern of phosphorylation observed these sites is changed in response to disease.

A significant part of cTnI released into the patient's blood stream is phosphorylated. For more than 15 years cTnI has been known as a reliable marker of cardiac muscle tissue injury. It is considered to be more sensitive and significantly more specific in diagnosis of the myocardial infarction.

ASSAY PRINCIPLE

The iFlash-Troponin-I assay is a sandwich immunoassay.

- Incubation: Troponin-I in the sample, anti-Troponin-I coated paramagnetic microparticles and anti-Troponin-I acridinium-ester-labeled conjugate react to form a sandwich complex.
- Wash: The unbound materials are washed away from the solid phase in a magnetic field.
- Trigger of signal: The Pre-Trigger and Trigger Solutions are added to the reaction mixture. The resulting chemiluminescent reaction is measured as relative light units (RLUs).
- A direct relationship exists between the amount of Troponin-I in the sample and the RLUs detected by the iFlash optical system.
- Results are determined via a calibration curve, which is instrument-specifically generated by 3-point calibration and a master curve provided via the reagent QR code.

REAGENTS

Reagent kit, 100 tests, 2 packs, 50 tests/pack

R1	Anti- Troponin-I coated microparticles, 3.5 mL/pack, 0.05% ProClin 300.
R2	Anti- Troponin-I acridinium-ester-labeled conjugate; 4.0 mL/pack; 0.05% ProClin 300.
CAL1	Calibrator 1, 1 bottle, 1.0 mL, phosphate buffer with protein stabilizers, 0.05% ProClin 300, lyophilized.
CAL2	Calibrator 2, 1 bottle, 1.0 mL, Troponin-I in phosphate buffer with protein stabilizers, 0.05% ProClin 300, lyophilized.
CAL3	Calibrator 3, 1 bottle, 1.0 mL, Troponin-I in phosphate buffer with protein stabilizers, 0.05% ProClin 300, lyophilized.

MATERIALS REQUIRED (BUT NOT PROVIDED)

REF C89999/ C89959/ C89949, iFlash Pre-Trigger Solution: hydrogen peroxide solution.

REF C89998/ C89958/ C89948, iFlash Trigger Solution: sodium hydroxide solution.

REF C89997, iFlash Wash Buffer: phosphate buffered saline solution with 0.05% ProClin 300.

REF C80001, iFlash Wash Buffer (10×): phosphate buffered saline solution with 0.05% ProClin 300.

REF C89996, reaction vessels.

WARNINGS AND PRECAUTIONS

IVD For in vitro diagnostic use

- No known test method can offer the complete assurance that products derived from human sources will not transmit infection. Therefore, all humanized materials should be considered potentially infectious.
- Exercise the normal precautions required for handling all laboratory reagents.
- Disposal of all waste material should be in accordance with local guidelines.
- Wear gloves when handling specimens or reagents.
- Clean and disinfect all spills of specimens or reagents using a suitable disinfectant.
- iFlash Trigger solution contains sodium hydroxide (NaOH) and should be avoided contact with eyes.
- For further information on warnings and precautions, see Annex B.

REAGENT HANDLING

- The reagents may not be used after the stated expiration date.
- Avoid the formation of foam with all reagents.
- The reagents in the pack are ready for use.
- Each tube of lyophilized powder, as a calibrator, needs adding 1.0 mL deionized water, dissolving for 15 minutes and gently blending for the first time.
- Calibrators should be measured within 30 minutes after dissolving, otherwise preserved as frozen.
- Do not pool reagents within a reagent kit or between reagent kits.
- Prior to loading the iFlash-Troponin-I reagent pack on the system for the first time, resuspend the microparticles by inverting the reagent pack slightly.
- For further information on reagent handling precautions during system operation, refer to the iFlash system operating instruction.

STORAGE AND STABILITY

Storage:

- Store at 2-8°C in an upright position.
- The kit may be used immediately after removal from 2-8°C storage.

Stability:

- Unopened at 2-8°C: up to the stated expiration date.
- Opened at 2-8°C: 28 days.
- Store on-board: 28 days.

SPECIMEN COLLECTION AND PREPARATION

- Serum or plasma (lithium heparin, sodium heparin potassium EDTA, and sodium citrate) are the recommended samples. Other anticoagulants have not been validated for use with the iFlash-Troponin-I assay.
- Ensure that complete clot formation in serum specimens has taken place prior to centrifugation. For patients undergoing heparin (anticoagulant) treatment, prolong the time for clot formation in serum specimens.
- Centrifuge the specimens.
- Store specimens at room temperature (20 to 25°C) for no longer than 8 hours.
- If the testing will not be completed within 8 hours, refrigerate the samples at 2 to 8°C.
- If the testing will not be completed within 3 days, or for shipment of samples, freeze at -20°C or colder.
- Frozen specimens must be mixed thoroughly after thawing.
- The samples may be frozen for maximum 1 time.
- Centrifuge specimens with a lipid layer on the top, and transfer only the clarified specimen without the lipemic material.
- Centrifuge the specimens prior to test if clotted fibrin and cellular material exist, or after freeze-thawing.
- Ensure that residual fibrin and cellular matter have been removed prior to analysis.
- Use with caution in handling patient specimens to prevent cross-contamination.
- Do not use heat-inactivated samples.
- Ensure that the patient samples, calibrators and controls are at ambient temperature (20-25°C) before measurement.
- Due to the possible evaporation, specimens on the analyzers should be measured within 2 hours.

ASSAY PROCEDURE

- Refer to the system operating instruction or the online help system for detailed information on preparing the system.
- The test-specific parameters stored in barcode on the reagent pack are read in. In case the barcode cannot be read, enter the sequence numbers.
- Carry out calibration, if necessary.
- Place the calibrators CAL1, CAL2 and CAL3 in the calibrator rack in the sample zone. Only keep calibrators open during calibration.
- Test application.
- Load samples (80 µL of sample is needed for each determination in addition to the sample container and system dead volumes).
- Click RUN, the iFlash System performs all the functions automatically and calculates the results.

CALIBRATION

- Traceability: This assay is traceable to a commercial available kit.
- Every iFlash-Troponin-I reagent kit has a QR code label containing the specific information for calibration of the particular reagent lot.
- To perform an iFlash-Troponin-I calibration, test CAL1, CAL2 and CAL3 in duplicate, and the predefined master curve is adapted to the analyzer.
- Once an iFlash-Troponin-I calibration is accepted and stored, all subsequent samples may be tested without further calibration unless:
  - After 28 days when using the same reagent lot.
  - A reagent kit with a new lot number is used.
  - Controls are out of range.
  - Required by pertinent regulations.

MEASURING RANGE

- 0.01-100 ng/mL

QUALITY CONTROL

Quality control materials should be run as single determinations at least once every 24 hours when the test is in use, once per reagent kit and after every calibration. Include commercially available quality control materials that cover at least two levels of analyte. Follow manufacturer's instructions for reconstitution and storage. Each laboratory should establish mean values and acceptable ranges to assure proper performance. Quality control results that do not fall within acceptable ranges may indicate invalid test results.

RESULT

Calculation:

The iFlash system automatically calculates the analyte concentration of each sample. The results are given in ng/mL.

Expected Values:

A study of with iFlash-Troponin-I assay on samples from 289 apparently healthy people of various age groups yielded the following result:

<0.03 ng/mL (99th percentile)

It is recommended that each laboratory establish its own expected reference range for the specific population.

LIMITATIONS

- The iFlash-Troponin-I assay is limited to the determination of Troponin-I in human serum or plasma (lithium heparin, sodium heparin, potassium EDTA, and sodium citrate). It has not been validated for use with other types of plasma.
- The use of serum separator (gel) blood collection tubes has been validated for use with this assay. However,
- It is not possible to survey all manufacturers or tube types.
- If the results are inconsistent with clinical evidence, additional testing is suggested to confirm the result.
- For diagnostic purposes, the results should be interpreted in light of the total clinical presentation of the patient, including symptoms, clinical history results.
- Samples containing an apparent Troponin-I level as high as 200,000 ng/mL did not exhibit a hook effect in the iFlash-Troponin-I assay.
- The results from an alternative assays (i.e. EIA or RIA) may not be equivalent and cannot be used interchangeably.
- The assay is unaffected by icterus (bilirubin < 10 mg/dL), hemolysis (Hb < 500 mg/dL), lipemia

(Intralipid < 1,800 mg/dL) and total serum protein (< 10 g/dL).

- No interference was observed from rheumatoid factors up to a concentration of 2,000 IU/mL.
- No interference was observed from HAMA up to a concentration of 600 ng/mL.
- No interference was observed from anti-nuclear antibodies up to a concentration of 500 AU/mL.

PERFORMANCE CHARACTERISTICS

Below are the representative performance data, and the results obtained in individual laboratories may differ.

Precision

The iFlash-Troponin-I is designed to have a precision of ≤10% total CV.

The precision of iFlash-Troponin-I was determined using Troponin-I reagents, samples and controls. Three serum samples, consisting low, median, and high concentration of Troponin-I were assayed.

The within run precision was determined by testing each sample in replicates of 10 (n = 10), and calculating percent coefficient of variation (%CV). The results of the study are shown below:

Sample	Mean (ng/mL)	SD	%CV
1	0.021	0.002	9.52
2	0.039	0.003	7.69
3	10.77	0.60	5.57

The between run precision was determined by testing each sample in duplicate, two separate runs daily for 20 days (n = 80), and calculating percent coefficient of variation (%CV). The results of the study are shown below:

Sample	Mean (ng/mL)	SD	%CV
1	1.18	0.05	4.24
2	10.80	0.50	4.63

Analytical Sensitivity

The detection limit representing the lowest measurable analyte level is 0.01ng/mL, which can be distinguished from zero. It is calculated as the value lying two standard deviations above that of the lowest standard of the master curve (standard 1 + 2 SD, n = 20).

Method comparison

A comparison of the iFlash-Troponin-I assay (y) with a commercially available Troponin-I assay (x) using clinical samples was performed, and the curve is fitted with Linear regression)

y = 1.009x + 3.522

r = 0.994

Sample concentration: 0.01-100 ng/mL

Number of samples measured: 88

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SHENZHEN YHLO BIOTECH CO., LTD.
Building 1, YHLO Biopark, Baolong 2nd Road, Baolong Subdistrict, Longgang District, 518116 Shenzhen, PEOPLE'S REPUBLIC OF CHINA

EC REP Well Kang Limited (www.CE-marking.eu)
The Black Church, St. Mary's Place, Dublin 7, D07 P4AX, Ireland

ANNEX A:

Explanation of abbreviation

Abbreviation	Explanation
REF	Catalogue number
CAL	Calibrator
R	Reagent
Σ	Contains sufficient for <n> tests
	Manufacturer
EC REP	Authorized representative in the European Community
CE	CE Conformity Marking
	Caution
	Consult instructions for use
IVD	In vitro diagnostic medical device
LOT	Batch code
	Date of manufacture
	Use-by date
2℃ 8℃	Temperature limit (2-8°C)
	Biological risks
GHS07	Pictograms for Caution
GHS09	Pictograms for Hazardous to the aquatic environment
	This way up

ANNEX B:

WARNINGS AND PRECAUTIONS (Proclin 300)

- Hazardous Component: 0.05% Proclin 300

  (Reaction mass of: 5-chloro-2-methyl-4-isothiazolin-3-one [EC no. 247-500-7] and 2-methyl-4-isothiazolin-3-one [EC no. 220-239-6] (3:1))
- Hazard Statement:

  H317: May cause an allergic skin reaction.

  H410: Very toxic to aquatic life with long-lasting effects.
- Precautionary Statement:

  P261: Avoid breathing dust/ fume/ gas/ mist/ vapours/ spray.

  P272: Contaminated work clothing should not be allowed out of the workplace.

  P273: Avoid release to the environment.

  P280: Wear protective gloves/protective clothing/eye protection/face protection.

  P302+P352: IF ON SKIN: Wash with plenty of soap and water.

  P333+P313: If skin irritation or rash occurs: Get medical advice/attention.

  P321: Seek immediate care from a doctor.

  P363: Wash contaminated clothing before reuse.

  P391: Collect spillage.

  P501: Dispose of contents/container in a safe way.