

T3 QUANTI CARD

Fluorescence immunoassay (antigen-antibody) for
Quantitative measurement of Triiodothyronin (T3) in Human Serum/ Plasma

INTRODUCTION

Triiodothyronin (T3) is a thyroid hormone with molecular weight of 651 dalton. T3 affects almost every physiological function of body including growth & development, metabolism, body temperature & heart rate. T3 circulate in blood as an equilibrium mixture of free & protein bound hormone in blood. T3 is a useful marker for the diagnosis of hypothyroidism and hyperthyroidism.

INTENDED USE

T3 Quanti Card is a sensitive immunoassay for the quantitative determination of Triiodothyronin (T3) in human serum/ plasma with iQuant Analyzer.

PRINCIPLE (ANTIGEN-ANTIBODY REACTION)

T3 Quanti Card is based on principle of competitive immunoassay. The T3 from the sample is first released/extracted. Extracted T3 from test sample and T3 biotin conjugate competes for the limited number of binding sites of specific antibodies fluorochrome conjugate. As a result higher concentration of T3 produces a lower fluorescence signal and vice versa. The signal is interpreted and displayed on iQuant Analyzer in form of results.

KIT PRESENTATION & MATERIALS PROVIDED

T3 Quanti Card Test kit contains following components to perform the assay:

S. No.	Component	50 Test Pack
1.	T3 Quanti Card	50 Nos.
2.	Releasing Buffer	2 Vials
3.	Conjugate-A	2 Vials
4.	Microtube	50 Nos.
5.	Instruction Manual	1 No.

MATERIAL REQUIRED, BUT NOT PROVIDED















iQuant Analyzer	Micropipette & Microtips
Stop Watch	iHeating Block

STORAGE AND STABILITY


T3 Quanti Card should be stored at 2-8°C in the coolest & driest area available. Expiry date on the kit indicates the date beyond which kit should not be used. The kit should not be frozen & must be protected from exposure to humidity and direct sunlight.

DESCRIPTION OF SYMBOLS USED

The following are graphical symbols used in or found on J. Mitra diagnostic products and packing. These symbols are the most common ones appearing on medical cartridges and their packing. They are explained in more detail in the European Standard EN ISO 15223-1:2016.

	Manufactured By		In vitro diagnostic medical device
	No. of tests		See Instruction for use
	Lot Number Batch Number		Temperature Limitation
	Manufacturing Date		Caution, see instruction for use
	Expiry Date		Catalogue Number
	Single use only		Keep away from sunlight
	Do not use if package is damaged		Keep Dry

WARNING FOR USERS

 **CAUTION:** ALL THE SAMPLES TO BE TESTED SHOULD BE HANDLED AS THOUGH CAPABLE OF TRANSMITTING INFECTION. NO TEST METHOD CAN OFFER COMPLETE ASSURANCE THAT HUMAN BLOOD PRODUCTS WILL NOT TRANSMIT INFECTION.

1. The use of disposable gloves and proper biohazardous clothing is STRONGLY RECOMMENDED while running the test.
2. In case there is a cut or wound in hand, DO NOT PERFORM THE TEST.
3. Do not smoke, drink or eat in areas where specimens or kit reagents are being handled.
4. Tests are for in vitro diagnostic use only and should be run by competent person only.
5. Do not pipette by mouth.
6. All materials used in the assay and samples should be decontaminated in suitable disinfectant solution for 30-60 min. before disposal or by autoclaving at 121°C at 15psi for 60 min. Do not autoclave materials or solution containing sodium hypochlorite. They should be disposed off in accordance with established safety procedures and guidelines.

7. Wash hands thoroughly with soap or any suitable detergent, after the use of the kit. Consult a physician immediately in case of accident or contact with eyes, in the event that contaminated material are ingested or come in contact with skin puncture or wounds.
8. Conjugate-A contains Sodium Azide as a preservative. If these material are to be disposed off through a sink or other common plumbing systems, flush with generous amounts of water to prevent accumulation of potentially explosive compounds. In addition, consult the manual guideline "Safety Management No. CDC-22", Decontamination of Laboratory Sink Drains to remove Azide salts" (Centre for Disease Control, Atlanta, Georgia, April 30, 1976).

PRECAUTIONS

In order to obtain reproducible results, the following instructions must be followed:

1. Use disposable gloves while handling potentially infectious samples and performing the assay. Wash hands thoroughly afterwards.
2. Do not use the kit beyond the expiry date.
3. Do not mix reagents from different batches.
4. Do not open the foil pouch until it attains room temperature.
5. Do not re-use the test device.
6. Use separate pipette tips for each sample in order to avoid cross-contamination of samples which could cause erroneous results.
7. Do not interchange caps of reagents.
8. Follow the given test procedure and storage instructions strictly to get accurate result.

Important Note: T3 Quanti Card is only operational in conjunction with iQuant Analyzer.

SAMPLE / SPECIMEN COLLECTION AND STORAGE

1. Collect the whole blood in a clean container (containing EDTA, sodium fluoride or heparin) by using standard venipuncture technique and the serum should be separated from the red cells. Fresh serum samples are preferred for testing.
2. If specimens cannot be tested immediately, they should be refrigerated at 2-8°C. For storage for more than 3 days, freeze the specimen at -20°C or below.
3. Frozen samples shall be thawed only once and repeated freezing and thawing of samples is not recommended.
4. Specimens containing precipitate or particulate matter may yield inconsistent test results. Such specimens must be centrifuged and the clear supernatant should only be used for testing.
5. The use of hemolytic, lipaemic, icteric or bacterially contaminated specimens should be avoided as it may lead to erroneous results.

BEFORE YOU START

1. Switch on the i Heating block before starting the test procedure and make sure it should attain the factory preset (50°C) temperature required for testing. As the iHeating block is switched ON, its "RED" LED indicator will glow and upon attaining the desired temperature its "GREEN" LED will glow. It may take approximately 15-20 min to achieve the factory preset temperature. The "RED" LED indicator may go ON/OFF as per heating requirement. However "GREEN" LED will glow continuously. Once its "GREEN" LED is ON, the instrument is ready for use.
2. Plug in the i Quant analyzer. Press the Power button of the i Quant analyzer, it will take approximately 1 minute for its self-checking and when the test screen will come, one can start the test procedure.
3. Plug in the iVortexer.
4. Bring the complete test kit and samples to be tested to room temperature (RT) prior to testing.

RT
20-30°C

TEST PROCEDURE

Reliable and reproducible results will be obtained when the assay procedure given in this Instructions for use is carried out with a complete understanding of the instructions and with adherence to good laboratory practice.

Step A: T3 Releasing Step:

- 1) Take 200µl of serum sample in a fresh micro tube provided in the kit and add 100µl of Releasing Buffer followed by addition of 50µl of Conjugate-A. Mix well by

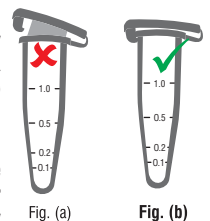


Fig. (a)

Fig. (b)

vortexing for 15 sec using iVortexer and incubate for 30 minutes by placing it in the pre-heated Dual iHeating block. **Use separate tips** for each pipetting step.

Note: Close the lid of the microtube properly by pressing it twice to ensure proper locking as shown in fig. (b) and it should be followed for each step.

Step B: Assay Procedure for running the test:

- i) Remove the test cartridge from foil pouch prior to use and place it on a flat and dry surface. The test should be performed immediately after removing the test cartridge from foil pouch.
- ii) Label the test cartridge with patient's name or identification number. **DO NOT write on QR Code.**
- iii) After incubation is complete, load **75µl** of the mixed solution to the sample well of the cartridge and insert the test cartridge into the cartridge slot of Dual iHeating Block. **Care should be taken to avoid any spillage on the QR-Code pasted on the cartridge & on the result reading window.**
- iv) Allow the reaction to occur for **20 minutes** in Dual iHeating Block. In the meantime enter the patient's details in the iQuant analyzer testing window and select the T3 test from the pop down menu in the testing window of the iQuant analyzer.
- v) Insert the test cartridge after 20 minutes into the iQuant analyzer with arrow (←) marked side on the top of the cartridge facing towards the iQuant analyzer.
- vi) Press the "RUN" icon on the iQuant analyzer test window.
- vii) Note down the T3 value displayed on the result window of the iQuant analyzer.
- viii) **Discard the T3 Quanti Card immediately after reading the results at 20 minutes considering it to be potentially infectious.**



MEASURING RANGE

The measuring range of T3 Quanti Card is 0.3 - 7.5 ng/ml. (0.46 - 2.76 nmol/L)
 Detection Limit: 0.3 ng/ml
 Each laboratory should establish its own range of normal value. The values given below are only indicative.
 Distribution of normal values ranges from 0.5 to 1.8 ng/ml.

PERFORMANCE CHARACTERISTICS OF T3 Quanti Card

1. Precision
 Intra-Assay: Within-run and between-run precision have been determined by testing 10 replicates of 3 different samples with T3 concentration (0.5ng/ml, 2.0 ng/ml & 4.0 ng/ml respectively) on the same lot on same day. The C.V (%) is ≤ 10%.
 Inter-Assay: The inter-assays were performed with 10 replicates of 3 different samples T3 concentration (0.5ng/ml, 2.0 ng/ml & 4.0 ng/ml respectively) of three different lots on 10 sequential days. The C.V (%) is ≤10%.
2. Accuracy
 The T3 concentration of 576 clinical specimen were quantified independently with T3 Quanti Card and commercially available certified kit. The following results were obtained:
 Slope : 0.99300
 Y-Intercept : 0.0025
 R² : 0.9923
3. Sensitivity
 The analytical sensitivity of T3 Quanti Card is 0.5 ng/ml.
4. Specificity
 The cross reactivity of the T3 antibody to selected substances was evaluated by adding the interfering substance to a serum matrix at various concentration.

Substances	Cross Reactivity	Concentration
I-Triiodothyronine	1.000	
I-Thyroxine	<.0002	10 µg/ml
Iodothyrosine	<.0001	10 µg/ml
Diiodothyrosine	<.0001	10 µg/ml
Diiodothyronine	<.0001	10 µg/ml

LIMITATIONS AND INTERFERENCES

1. The test procedure, precautions and interpretation of results for this test must be strictly followed.
2. As with all diagnostic tests, the test result must always be correlated with clinical finding and laboratory data available.
3. Any modification to the above procedure and/ or use of other reagents will invalidate the test procedure.
4. Technical / procedural errors as well as the presence of additional substances in blood samples may interfere with product performance and may cause erroneous results.
5. The test has been developed for testing Human serum/ plasma samples only.

LIMITED EXPRESSED WARRANTY DISCLAIMER

The manufacturer limits the warranty to the test kit, as much as that the test kit will function as an in vitro diagnostic assay within the limitations and specifications as described in the product instruction-manual, when used strictly in accordance with the instructions contained therein. The manufacturer disclaims any warranty expressed or implied including such expressed or implied warranty with respect to merchantability, fitness for use or implied utility for any purpose. The manufacturer's liability is limited to either replacement of the product or refund of the purchase price of the product and in no case liable to claim of any kind for an amount greater than the purchase price of the goods in respect of which damages are likely to be claimed. The manufacturer shall not be liable to the purchaser or third parties for any injury, damage or economic loss, howsoever caused by the product in the use or in the application there of.

REFERENCES

1. Chopra I.J., HoR. s., & Lam R. "An improved radio immunoassay of triiodothyronine in human serum" J. Lab Clinical Med 80, 729 (1971).
2. Sterling L., "Diagnosis & Treatment of thyroid disease" cleveland CRC press, p-9-51 (1975).
3. Emlinger MW, Kuhnel W, Lambrecht HG, Ranke MB. Reference intervals from birth to adulthood for serum thyroxine (T4), triiodothyronine (T3), free T3, free T4, thyroxine binding globulin (TBG) and thyrotropin (TSH). Clin Chem Lab Med 2001;39:973-9.

TROUBLE SHOOTING

PROBLEM	POSSIBLE CAUSE	SOLUTION
1. Unexpected low test result	a) Incubation period of Releasing step less than 30 minutes.	Repeat the test with incubation period of 30 minutes.
	b) High volume of Conjugate-A used.	Use accurate volume of Conjugate-A
	c) Reagents used were too cold & were not brought to room temperature (R.T.)	Bring the whole test kit to R.T. before testing.
	d) Use of uncalibrated pipette, improper pipetting	Use only calibrated pipettes with well fitted tips & pipette carefully without bubbling.
2. Unexpected high test result	a) Low volume of reaction mixture applied to sample well of cartridge.	Repeat test with appropriate loading Volume.
	b) Expired test kit used.	Repeat test using a new test kit that has not passed the expiration date.
	c) Low volume of Conjugate-A used.	Use accurate volume of Conjugate-A.
	d) Conjugate-A is continuously exposed to light for more than 8 hrs.	Always recap the Conjugate-A vial after use. Store at 2-8°C.
3. Test Result do not match with other Elisa or CLIA method	e) Use of uncalibrated pipettes, improper pipetting.	Use only calibrated pipettes with well fitted tips & pipette
	a) Result on Elisa or CLIA kit can be more or less than actual value due to improper calibration of instrument.	Recheck the sample after proper calibration of instrument.
	b) Procedural error.	Check the procedure and perform test again as per kit product insert. Retest the sample on standard ELISA or CLIA kit to confirm the correct value of the sample.

in vitro diagnostic reagent, not for medicinal use

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