FREND™ Free T4

Free Thyroxine

Intended use

The FREND™ Free T4 Test System, is a rapid indirect competitive fluorescence immunoassay for the quantitative determination of free thyroxine (FT4) in human serum and lithium heparinized plasma specimens using the FREND™ System. Measurements of FT4 are used in the diagnosis of thyroid disorders. The FREND™ Free T4 Test System is intended for use in clinical laboratories.
For *in vitro* diagnostic use only. The test is not intended for use in point-of-care settings.

Summary and explanation of test

Human Thyroid Stimulating Hormone (hTSH) or thyrotropin stimulates the secretion of thyroxine (T4) and triiodothyronine (T3) by the thyroid gland.[1] From the moment it is secreted into the blood stream, thyroxine or tetraiodothyronine (T4), produced by the thyroid gland, is predominantly (>99%) bound to the carrier proteins TBG (Thyroxine Binding Globulin), TBPA (Thyroxine Binding PreAlbumin) and albumin. The fraction that remains free (FT4) is considered the active part of the hormone.[2] The mechanisms regulating thyroid function have a direct effect on the concentration of this free fraction, which explains why it is relatively independent of the concentration of carrier proteins.[3,4] In patients with hyperthyroidism, the FT4 concentration increases, whereas in patients with hypothyroidism it generally decreases. Patients on levothyroxine hormone replacement therapy (LT4) may have an elevation of FT4, although clinically they are euthyroid.

T3 and T4 are known to have diverse functions in regulating basal metabolic

rate, bone growth, neuronal development, and sex maturation.[5]
Underproduction of T3 and/or T4 can result in hypothyroidism, while overproduction of these hormones can result in hyperthyroidism.[6] Because of a negative feedback mechanism, elevation of T3 and T4 suppress the production of TSH.[7] TSH itself is stimulated by thyrotropin releasing hormone (TRH), a tripeptide produced in the hypothalamus.[8]

Primary hypothyroidism occurs when TSH levels are elevated while T3 and/or T4 are underproduced.[9,10] Secondary or tertiary hypothyroidism can occur due to abnormal response of TSH to TRH, while central hypothyroidism occurs from pituitary dysfunction.[11,12] Primary hyperthyroidism is marked by low levels of TSH and high levels of T3 and/or T4.[9] Anomalies to these types of classification exist, but TSH testing can (with the aid of other thyroid tests) help a clinician determine the presence of thyroid dysfunction.[13-15]

Principle of the assay

A 70 µL specimen is added to a single-use tube containing gold-T4 antibodies and incubated in FREND™ Heat Block for five minutes, while free thyroxines react with gold particles bound to antibodies against free thyroxine. After five minutes, 35 µL of sample in the tube is transferred to the sample inlet of a single use FREND™ Free T4 test cartridge and allowed to equilibrate for 30 seconds on the Heating Block warming plate. The cartridge is then placed into the FREND™ System, which is programmed to begin analysis once the sample has reacted with the reagents. The reaction and analysis time is approximately 4 minutes. The FT4 quantification is based on the ratio of fluorescence detected by the FREND™ System at the FREND™ Free T4 Test and Reference zones. The magnitude of the fluorescent ratio is inversely proportional to the amount of FT4 in the sample, so a lower ratio of fluorescence correlates with a higher FT4 concentration.

The FREND™ System is a bench-top fluorescence reader containing a touchscreen user interface. The System has a slot that accepts the FREND™ Free T4 test cartridge (which contains the reagents and sample), and is programmed to analyze the test when the sample has fully reacted with the on-board cartridge reagents. Results of the test are displayed on the screen and can be printed on an optional printer.

Material provided

Q'ty	Contents	Catalogue number
20	FREND™ Free T4 Cartridges	FRT4 020
20	FREND™ Free T4 Pretreatment tubes	
30	Disposable pipette tips	
01	FREND™ Free T4 Code chip	
01	FREND™ Free T4 Package insert	

One cartridge contains:
T3-BSA
Anti-T3 antibody
Fluorescent particles

One pretreatment tube contains
Anti-T4 antibody
Gold nano-particle

Materials required but not provided

- The FREND™ System
- Micro-pipette, or any pipettor, capable of delivering 35 and 70 µL
- FREND™ Companion Heating Block (with tube holes for tube incubation and warming plate for cartridge incubation at 37 ℃)
- Personal protective equipment and biohazard waste disposal containers

Warning and precautions

⚠ ***Caution:*** *In the United States, Federal law restricts this device to sale by or on the* order of a physician

- The FREND™ Free T4 cartridges are intended for *in vitro* diagnostic use only.
- The FREND™ Free T4 cartridges are only to be used on the NanoEntek FREND™ System.
- The FREND™ Free T4 cartridges are disposable, single use devices. Do not reuse them under any circumstances.

- Allow sealed cartridges to come to room temperature for 15-30 minutes prior to use.
- Cartridges should not be frozen.
- Assure the humidity in the laboratory is in the 10-80% range when tests run.
- Use the Heat Block as instructed when performing the assay.
- Avoid cross-contamination between samples by using a new pipette tip for each new specimen.
- Avoid high humidity, direct sunlight or heat in the area used for cartridge storage.
- Inaccurate results are possible if the sample used is contaminated in any way.
- Using specimens containing clotted fibrin could result in erroneous results.
- Over or under loading the cartridge with sample may result in inaccurate results.
- Do not use the cartridges beyond the expiration date on the pouch.
- Do not use the cartridge if the pouch is damaged or the seal is broken.
- Perform testing as specified in the Package Insert Sheet and User Manual.
- Keep the cartridge sealed in the pouch until ready for use.
- Use the cartridge immediately after opening the pouch.
- Handle specimens in accordance with the OSHA Standard on Bloodborne Pathogens.
- Human specimens are not used in the preparation of this product, however, since human specimens will be used for samples and other quality control products in the laboratory may be derived from human materials, Use Universal Precautions when handling all specimens and controls.
  Wear disposable gloves when handling the cartridges and the samples.
- Wash hands thoroughly and often after handling reagent cartridges or samples.
- Do not ingest the silica gel packet found in the cartridge pouch.

Storage and Stability

All unopened materials are stable until the expiration date on the label when stored at the specified temperature. Reagent stability has been demonstrated for twelve months from the date of manufacture.

The expiration date is clearly indicated on the product outer box and the cartridges.

Materials	Catalogue number
Refrigerator temperature storage (2~8 ℃)	
FREND™ Free T4 Cartridges	FRT4 020
FREND™ Free T4 Pretreatment tubes	None
Room temperature storage (18~25 ℃)	
Pipette tips	None

Specimen collection and handling

Serum or lithium heparinized plasma is required for the assay. No special patient preparation is necessary. Collect the appropriate venous blood sample aseptically. For serum, allow the sample to clot for 30 minutes at room temperature. For lithium heparin, centrifuge after collection. Centrifuge the sample for 10 minutes at 3,000 rpm within 2 hours of collection and immediately separate the serum or plasma from the packed cells.

Separated Samples may be stored at 2-8 °C for up to 1 week prior to analysis. If the analysis is scheduled to be done more than 1 week after collection, the sample should be stored frozen at -20 °C or below for future use.

Repeated freeze-thaw cycles should be avoided. Turbid serum samples or samples containing particulate matter such as fibrin clots or visible strands should be re-centrifuged before being tested. Prior to assay, slowly bring frozen samples to room temperature and mix gently but thoroughly before testing.

The sample required for the incubation step is 70 µL. The sample required for running the test on the FREND™ Free T4 cartridge is 35 µL.

For optimal results, avoid grossly hemolytic, lipemic, or turbid specimens. Specimens should be free of aggregated fibrin, red blood cells, or other particulate matter. When pipetting into the FREND™ Free T4 cartridge sample inlet, ensure that bubbles in the sample are avoided. Bubbles may restrict flow and result in an incomplete or erroneous test result.

Procedure

Calibration

There is no need for calibration to be performed by the end user. All calibration statistics and information have been electronically stored on the FREND™ Free T4 Code chip included in each box of FREND™ Free T4 cartridges. The FREND™ Free T4 Code chip is specific for each manufactured lot of FREND™ Free T4 cartridges.

Always run external quality control samples to verify that the FT4 results obtained on the FREND™ System meet the laboratory criteria for acceptability for each lot of FREND™ Free T4 cartridges.

Code chip installation

Please refer to the FREND™ System User manual for more detailed instructions relative to the Code chip installation. Abbreviated instructions are as follows:

1) Insert the FREND™ system electrical cord into an appropriate outlet.
2) Insert the Code chip into the Code chip slot at the rear of the system following the arrows.
3) Press the ‘Setup’ button on the ‘Main’ screen.
4) Press the ‘Code chip’ button on the ‘Setup’ screen
5) The information embedded on the FREND™ Free T4 Code chip is automatically saved on the FREND™ System
6) When the Code chip installation is completed, press the ‘OK’ button to go to the ‘Setup’ screen
7) Press the ‘Item’ button on the ‘Setup’ screen.
8) Check the FREND™ Free T4 cartridge lot number and the installation date of the Code chip.
9) Press the ‘Home’ button to go to the ‘Main’ screen to begin running external quality control and patient samples.

Quality control

• FREND™ System QC cartridges

The FREND™ QC cartridge contains multiple controls to check the optics of the system. By testing with the QC cartridge, the analytical components of the system; (1) laser power (2) alignment, and (3) mechanical integrity are confirmed.

For each day of patient testing, perform QC Cartridge testing. Refer to the Quality Control Procedures section in the User manual of FREND™ System. In brief, perform QC Cartridge testing for the following conditions:

(1) Upon initial setup of the system
(2) Each day of patient testing
(3) When the system has been transported or moved
(4) Whenever there is uncertainty about the performance of the system
(5) Whenever required by your laboratory's quality control requirements

• Internal procedural controls

The FREND™ Free T4 test cartridge contains built-in control features.
Fluorescence signal in the Reference Zone of each cartridge shows: (1) that enough sample volume is added, (2) that proper flow is obtained, and (3) that the antibody is reactive. If this Reference Zone signal is missing or lower than the threshold, the FREND™ System considers it as an incorrect or failed test, and produces an error message instead of a test result. In addition, with each cartridge run, the system monitors, in part, for (1) flow of sample, (2) speed of sample flow, (3) shelf-life of cartridge components, (4) function of internal barcode scanner, and (5) function of scanner's mechanical components.

• **External quality control testing**

Commercially available controls that contain FT4 as a measured analyte are available from a variety of manufacturers. It is recommended that a minimum of two (2) levels of controls be run at least once per month or once for each new lot, whichever comes earlier. However, Controls should be run with a minimum frequency, depending on number of tests run in the laboratory. Each laboratory should establish its own criteria based on the following parameters

(1) Each new lot
(2) Each new shipment (even if from the same lot previously received)
(3) Each new operator (an individual who has not run the tests for at least two weeks)
(4) Monthly, as a continued check on storage conditions

Individual laboratory policy will dictate exactly which control materials and lot numbers should be run, the frequency with which controls are to be tested, criteria for acceptance of the results and required corrective action to be taken if results do not meet laboratory criteria. Refer to your laboratory policies in how to determine acceptability of external control material results. Each laboratory operates under a different set of regulations. Every laboratory must follow the standardized procedures acceptable to the regulatory agencies to whom the laboratory is responsible.

Specimen processing

• Preparation

Remove sufficient FREND™ Free T4 cartridges and Gold-T4 antibody tubes from the refrigerator to test the number of patient samples and required external quality control materials. Allow the tubes and the sealed pouches containing the cartridges to come to room temperature for 15-30 minutes prior to the start of the testing sequence. The FREND™ Companion Heating Block provided with FREND™ system also needs 7-8 minutes to warm up.

If using refrigerated patient samples, remove those from the refrigerator and allow them to come to room temperature prior to testing. If frozen samples will be utilized, be sure these are removed from the freezer, thawed naturally and then mixed gently but thoroughly prior to testing. Testing should not begin on previously frozen samples until they have reached room temperature.

There are no other reagents or sample preparations necessary.

• Assay procedure

Note: *When processing samples, rinse the pipette several times with the sample and dispense 70 µL into the pretreatment tube.*

1) Prepare the FREND™ Free T4 cartridge, pretreatment tube and specimen at room temperature (18~25 °C). Open the pouch and place FREND™ Free T4 cartridge into the cartridge tray of the AP device. Press **"NEXT"** to close the cartridge tray and open the pretreatment tube tray.
2) Transfer a 70 µL of specimen to the pretreatment tube.

⚠ ***Caution:*** *Once the sample is added to the pretreatment tube, do not invert the tube.*

3) Insert the pretreatment tube into the tube hole in the FREND™ AP pretreatment tube tray. Refer to the FREND™ AP User manual for complete operating instructions.
4) Press the **"NEXT"** button. The pretreatment tray will close and the first incubation step (5 minutes) will begin.
5) After the first incubation is complete, 35 µL of mixed sample will be loaded onto the cartridge and the second incubation step (2 minutes) will begin.
6) When both incubation steps are completed, the cartridge tray will open and the cartridge will be ready to be inserted into the FREND™ System.
7) Press the **'Test'** button on the **'Main'** screen of the FREND™ System.

8) The system moves to the Patient ID screen automatically.
9) Type the Patient ID and press the **'Enter'** button to begin the test.
10) Insert the cartridge into the cartridge slot using the cartridge arrow as a guide.

⚠ ***Caution:*** *Check the direction of the cartridge before insertion and assure the insertion is complete.*

11) When the reaction in the cartridge is completed, the FREND™ System will automatically begin the reading process.
12) When the measurements are completed, the cartridge will automatically be expelled and the results displayed.

⚠ ***Caution:*** *Do not disconnect power cord or shut off power on the FREND™ System while a cartridge is in the reading chamber. This may cause a system error.*

13) If the FREND™ System is connected to the optional printer, press the **'Print'** button and the results will be output on the printer paper. For more detailed instructions, please refer to the FREND™ System User Manual.

Specimen Dilution Procedures

Samples cannot be diluted for FT4 determinations. Samples which read >6.00 ng/dL should be reported as such.

Calculation of results

The FREND™ System performs all sample and reagent handling operations automatically within the cartridge once the sample has been manually loaded to the sample inlet in the cartridge and the cartridge placed into the FREND™ System. The rate of fluorescence produced by the reaction is read at various intervals during the analysis process. Blank readings are subtracted after which the net rate is automatically converted to FT4 concentration in ng/dL based upon information stored on the Free T4 Code chip. This result is then displayed on the screen and can be sent to the optional printer. It is also stored in memory on the FREND™ System.

Screen displayed for various concentration scenarios

Displayed result	Description
	FT4 concentration Less than 0.40 ng/dL
	FT4 concentration Not less than 0.40 ng/dL and not higher than 6.00 ng/dL
	Free T4 concentration Higher than 6.00 ng/dL

Limitations of the procedure

1) When used for diagnostic purposes, the results obtained from this assay should be used in conjunction with other data (e.g., symptoms, results of other tests, clinical impressions, medical history, therapy, etc).

2) The FREND™ System paired with a FREND™ Free T4 cartridge, is programmed to report 6.00 ng/dL as the highest concentration of FT4 measurable without dilution. The lowest measurable concentration is 0.40 ng/dL – the assay limit of quantitation.
3) Specimens from patients with heterophilic antibodies, such as anti-mouse (HAMA), anti-goat (HAGA), or anti-rabbit (HARA) antibodies, may show falsely elevated or depressed values or may result in the error message "Incomplete Test". Patients routinely exposed to animals or animal serum products can be prone to these types of heterophilic interferences. If the FT4 level is inconsistent with clinical evidence, additional FT4 or other thyroid testing using a different method is suggested to confirm the results
4) Certain medications may interfere with assay performance. All results should be interpreted with respect to the clinical picture of the patient.
5) Although hemolysis has an insignificant effect on the assay, hemolyzed samples may indicate mistreatment of a specimen prior to assay and results should be interpreted with caution.
6) Lipemia has an insignificant effect on the assay except in the case of gross lipemia where interference with the lateral flow of the sample in the cartridge may occur.
7) The concentration of FT4 in a given sample determined using assays from different manufacturers can vary due to differences in assay methods, calibration, and antibody specificity.
8) Please refer to the Specimen Collection and Handling, Warnings and Precautions, Storage and Stability, and Procedural Notes sections in this insert sheet.
9) FREND™ Free T4 has not been validated in point-of-care settings.
10) Performance of this assay has not been established with neonatal specimens or specimens from pregnant patients.
11) FREND™ Free T4 is to be used in licensed clinical laboratories with trained technologists.

Expected values

As with every clinical diagnostic test, a reference interval corresponding to the characteristics of the population being tested should be determined by each laboratory. Historically, it has been shown that there are no race- or gender-specific differences in the reference interval for FT4, so a single adult reference interval is reasonable and justified.

Serum samples from a total of 196 normal, apparently healthy adult individuals were assayed on 3 lots of the FREND™ Free T4 assay using a single FREND™ System. The reference interval was determined according to CLSI guideline C28-A3, was found to be 0.83-1.60 ng/dL.

As in all *in vitro* diagnostic testing, a Free T4 result generated using the FREND™ Free T4 on the FREND™ System should be interpreted in the light other clinical findings and diagnostic procedures. Any FT4 results not correlating with the clinical condition should be repeated and other testing performed to clarify the situation.

Performance characteristics

Precision

A single lot imprecision study was performed at the NanoEntek laboratory as described in the CLSI guideline EP5-A3. Three serum pools were assayed for 20 days, 2 runs per day in duplicate using a single lot of Free T4 reagent cartridge. The results are summarized below:

FREND™ Free T4 Single Site Single Lot Precision

Sample ID	Mean Free T4 Conc. (ng/dL)	Repeatability		Between-run		Between-day		Within-laboratory	
		SD	CV%	SD	CV%	SD	CV%	SD	CV%
1	0.917	0.067	7.3	0.000	0.0	0.032	3.5	0.074	8.1
2	1.850	0.103	5.6	0.000	0.0	0.069	3.7	0.124	6.7
3	3.979	0.186	4.7	0.152	3.8	0.093	2.3	0.258	6.5

Linearity

To demonstrate the linearity of the assay, a serum base pool with an elevated FT4 (7.5 ng/dL) was prepared and diluted to a total of 11 levels according to the dilution protocol outlined in CLSI guideline EP6-A: Evaluation of the Linearity of Quantitative Measurement Procedures: A Statistical Approach. At each dilution level, the samples were tested in duplicate to determine the experimental value of FT4. Linearity was demonstrated from <0.4 ng/dL to >6.0 ng/dL. The measuring range for the FREND™ Free T4 is 0.4 ~ 6.0 ng/dL.

Method comparison

The comparison studies were performed in a CLIA-certified laboratory using de-identified fresh and frozen specimens. The reference method was the Abbott ARCHITECT Free T4 Assay run on the Abbott ARCHITECT i System. All samples (358) analyzed in the clinical testing were split and tested by both the ARCHITECT Free T4 and the FREND™ Free T4 devices.

Results generated using the FREND™ Free T4 on the FREND™ System (y) were compared to those obtained using a previously FDA cleared ARCHITECT Free T4 assay (x) by Ordinary least-square regression analysis. Results of this study are shown below.

Slope: 1.010 (95% CI: 0.992 to 1.028	y-Intercept: 0.057(95% CI: 0.021 to 0.094)
Number of Samples: 358	Range Tested: 0.43~ 5.99 ng/dL
r: 0.986	

Comparability using CLSI guideline EP9-A3 shows that the two methods compare favorably.

Matrix comparison

The matrix comparison study was performed according to CLSI EP14-A3. Free T4 concentrations in 48 paired serum and lithium heparin samples were measured using the FREND™ Free T4. Passing-Bablok regression analysis of serum results (x) compared to lithium heparin plasma results (y) yielded an acceptable regression (Slope = 1.017, Intercept = -0.0881, R^2 = 0.9948),

indicating that FREND™ Free T4 can be measured equally well in serum and lithium heparin plasma.

Sensitivity

The Limit of Detection (LoD) for the FREND™ Free T4 was established according to the CLSI guideline EP17-A2 and was determined to be 0.32 ng/dL. The Functional Sensitivity (LoQ) was determined to 0.36 ng/dL.

Specificity and Interferences

Interference was defined as recovery values outside of 10% of the known specimen mean concentration. Recovery within 90% to 110% of the expected Free T4 was considered as lack of interference. The interference studies were performed as recommended in the CLSI guideline EP07-A2 using two concentrations of free T4. Results are summarized in the table below.

	Interferent (Concentration tested)	%Recovery Free T4 Low	%Recovery Free T4 High
Endogenous substances	Hemoglobin (500 mg/dL)	109.5	106.3
	Bilirubin conj. (20 mg/dL)	90.5	101.5
	Bilirubin unconjugated (20 mg/dL)	103.0	101.0
	Triglyceride (3 g/dL)	108.1	105.5
	Total protein (12 g/dL)	107.7	103.6
	Biotin (2.5 µg/mL)	101.5	94.0
	IgG (2.5 mg/mL)	102.9	101.7
	IgA (60 µg/mL)	100.7	95.8
	IgM (45 µg/mL)	106.2	94.7
Pharmaceuticals	Acetaminophen (20µg/mL)	104.4	100.3
	Erythromycin (60µg/mL)	102.7	100.2
	Diltiazem (6.24µg/mL)	109.5	91.1
	Verapamil (2µg/mL)	108.2	90.5
	Acetylcysteine (415µg/mL)	102.7	109.4
	Acetylsalicylic acid (250µg/mL)	96.5	98.3
	Amiodarone (6µg/mL)	105.0	99.1
	Ampicillin-Na (50.3µg/mL)	99.7	91.8
	Ascorbic acid (60µg/mL)	100.0	91.9
	Carbimazole (500ng/mL)	90.5	91.2
	Cefoxitin (66µg/mL)	93.5	92.7
	Cyclosporine (3µg/mL)	103.8	96.2
	Doxycycline (30µg/mL)	104.8	95.1
	Fluocortolone (400 ng/mL)	90.1	99.5
	Furosemide (12.5µg/mL)	100.3	101.4
	Heparin (3,000 U/L)	102.2	91.2
	Hydrocortisone (1.8µg/mL)	96.9	90.1
	Ibuprofen (250µg/mL)	98.6	109.0
	Iodide (380µg/mL)	98.0	86.8

Pharmaceuticals	Levodopa (4 mg/mL)	99.2	103.9
	Methyldopa (15µg/mL)	100.0	90.0
	Metronidazole (120µg/mL)	99.0	92.9
	Octreotide (2 ng/mL)	101.7	90.8
	Perchlorate (16 ng/mL)	99.0	91.6
	Prednisolone (3µg/mL)	98.6	91.6
	Propranolol (2µg/mL)	96.9	90.0
	Propylthiouracil (10µg/mL)	90.5	90.8
	Rifampicin (640µg/mL)	91.5	93.9
	Theophylline (400µg/mL)	107.1	94.5
	Thiamazole/Methimazole (500 ng/mL)	102.0	90.7
	Avidin (5 µg/mL)	107.7	90.4
	Au-nanoparticles (5µg/mL)	103.4	97.4
Heterophilic Antibodies	RF (1075 IU/mL)	109.2	93.5
	HAMA (70 ng/mL)	104.4	96.5

The following substances were evaluated for potential cross-reactivity with the FREND™ Free T4 at two concentrations. Testing was done according to the CLSI guideline EP07-A2. No significant cross-reactivity was found except for the L-Thyroxine (Levothyroxine) itself.

Cross-reactant	Cross-reactant Concentration (ng/dL)	% Cross-reactivity	
		Free T4 Low	Free T4 High
Levothyroxine, T4 (1 µg/dL)	1,000	99.44	99.57
Diiodothyronine, T2 (5 µg/dL)	5,000	0.0001	0.0006
Tetraiodothyroacetic Acid (10 µg/dL)	10,000	0.0005	0.0005
Triiodothyroacetic Acid (1 µg/dL)	1,000	0.004	0.0157
Triiodothyropropionic Acid (5 µg/dL)	5,000	0.0019	0.0055
Diiodotyrosine,DIT (1 mg/dL)	1,000,000	0.000001	0.000002
L-Triiodothyronine, T3 (1 µg/dL)	1,000	0.003 7	0.026
Monoiodotyrosine (1 mg/dL)	1,000,000	0.000001	0.000019
Reverse T3 (10 µg/dL)	10,000	0.0009	0.0022

% Crossreactivity = 100 x ((Measured value - true value)/concentration of interferent), absolute value.

References

1) Pierce JG. The subunits of pituitary thyrotropin—their relationship to other glycoprotein hormones. *Endocrinology* 1971, 89, 1331-1344.
2) Sapin R, D'herbomez M. Dosage des hormones thyroïdiennes: thyroxine (T4) et triiodothyronine (T3). In La thyroïde, LeClere J, Orgiazzi J, Rousset B, Schlienger Elsevier, ed. 2001, 268-274.
3) Pearce C.J. Byfield PGH. Free thyroid hormone assays and thyroid function. *Annual Clinical Biochemistry.* 1986, 23, 230-237.
4) Ekins R. Measurement of Free Hormones in Blood - *Endocrine Reviews,* Vol. 11, No. 1, 1990, 5-46.
5) Smith BR, Pyle GA, Peterson VB, Hall R. Interaction of thyrotropin with the human thyrotropin receptor. *J. Endocrinol.* 1977, 75, 391-400.
6) Sterling K, Lazarus JH. The thyroid and its control. *Annu. Rev. Physiol*. 1977, 39, 349-371.
7) Patel YC, Alford FP, Burger HG. The 24-hour plasma thyrotrophin profile. *Clin. Sci.* 1972, 43, 71-77.
8) Morley JE. Neuroendorine control of thyrotropin secretion. *Endocr. Rev.* 1981, 2, 396-436.
9) Wehmann RE, Rubenstein HA, Pugeat MM, Nisula BC. Extended Clinical Utility of a Sensitive and Reliable Radioimmunoassay of Thyroid-Stimulating Hormone. *South. Med.* J. 1983, 76, 969-976.
10) Burger HG, Patel YC. The Value of Serum Thyrotropin Measurement in the Diagnosis and Management of Hypothyroidism. *Med. J. Aust.* 1972, 2, 293-297.
11) Petersen VB, McGregor AM, Belchetz PE, Elkeles RS, Hall R. The Secretion of Thyrotropin with Impaired Biological Activity in Patients with Hypothalimic-Pituitary Disease. *Clin. Endocrinol.* 1978, 8, 397-402.
12) Beck-Peccoz P, Amr S, Menezes-Ferreira MM, Faglia G, Weintraub B. Decreased Receptor Binding of Biologically Inactive Thyrotropin in Central Hypothyroidism. *N. Engl. J.* Med. 1985, 312, 1085-1090.
13) Hay ID, Bayer MF, Kaplan MM, Klee GG, Larson PR, Spencer CA. American Thyroid Association Assessment of Current Free Thyroid Hormone and Thyrotropin Measurements and Guidelines for Future Clinical Assays. *Clin. Chem.* 1991, 37, 2002-2008.
14) Demers LM, Spencer CA. Laboratory Medicine Practice Guidelines: Laboratory Support for the Diagnosis and Monitoring of Thyroid Disease. *The National Academy of Clinical Biochemistry,* 1996.
15) Beastall GH, Beckett GJ, Franklyn J; Frasier WD, Hickey J, John R, Kendall-Taylor P, Nevens B, Vanderpump M. UK Guidelines for the Use of Thyroid Function Tests. *The Association for Clinical Biochemistry,* 2006.

Glossary of symbols

	Caution, warning, Consult accompanying documents
	Catalogue number/Reference number
	Lot number/Batch number
	Use by YYYY-MM-DD or YYYY-MM
	Manufacturer
	Authorized representative in the European Community
	CE marking
	*In vitro* diagnostic medical device
	Temperature limitation
	Contains sufficient for <n> tests
	Do not reuse
	Do not use if package is damaged
	For prescription use only
	Irritant

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Revised on 2021. 01. 06