ULTRA-TSH-CHECK-1

Ultra Sensitive Quantitative determination of Thyroid Stimulating Hormone in plasma or serum samples

FOR EASY READER® AND EASY READER® USE ONLY

Ref: U21091

I- PRINCIPLE

Human thyroid stimulating hormone (TSH) is a glycoprotein secreted by the thyrotroph cells in the anterior pituitary (1).

The primary function of TSH is to regulate the release and to control the synthesis of the major thyroid hormones. When circulating thyroid hormone levels fall below normal, the pituitary secretes TSH. This in turn acts on the thyroid gland to produce and release more thyroid hormones. However, when circulating thyroid hormone levels rise above normal, the pituitary responds by releasing less TSH, causing the thyroid gland to decrease production and secretion of thyroid hormones.
Measurement of basal serum concentration of TSH is an essential test in the investigation of suspected hypothyroidism and hyperthyroidism. A raised concentration of TSH confirms a primary cause of the hypothyroidism, while a low concentration indicates hyperthyroidism.
Since the development of highly sensitive TSH immunometric assays, it is widely believed that measurement of TSH in serum probably represents the best single assessment of thyroid function. As well, measurement of TSH after injection of exogenous TRH is useful in the differentiation of secondary and tertiary (hypothalamic) hypothyroidism (3, 4, 5).

The ULTRA-TSH-CHECK-1 is a rapid quantitative assay for the detection of TSH in serum or plasma to be used as a screening test for hypothyroidism or hyperthyroidism diagnosis. The method employs a unique combination of monoclonal dye conjugate and polyclonal-solid phase antibodies to identify TSH in the test samples with a high degree of specificity.

As the sample flows through the absorbent device, the labelled antibody-dye conjugate binds to the TSH forming an antibody-antigen complex. This complex binds to the anti-TSH antibody in the reaction zone (T) and produces a pink colour band. In the absence of TSH, there is no line in the reaction zone (T). The mixture continues flowing through the absorbent device past the reactive zone (T) and control zone (C). Unbound conjugate binds to the reagents in the control zone (C), producing a pink colour band and demonstrating that the reagents are functioning correctly.

II- ULTRA-TSH-CHECK-1 KIT COMPONENTS

Each kit contains everything needed to perform 10 or 20 tests.

1- ULTRA-TSH-CHECK-1 reaction devices:	10	20
2- Disposable plastic pipettes:	10	20
3- Instruction leaflet:	1	1

4- Controls (Optional):

Positive control (ref. V2500U) and Negative control (ref. V2501U): a freeze-dried preparation of a non-infectious compound in diluted Human serum tested and found negative for anti-HIV, anti-HCV and HBs antigen, containing 0.05 % sodium azide and optionally available as a positive and negative control (1x 0.25 mL). The concentration range is indicated on the vial label.

III- STORAGE AND STABILITY

1- All ULTRA-TSH-CHECK-1 kit components should be stored at room temperature (+4°C to +30°C) in the sealed pouch.
2- ***Do not freeze the test kit.***
3- The ULTRA-TSH-CHECK-1 kit is stable until the expiry date stated on the package label.

IV- PRECAUTIONS

1- This test is designed for *in vitro* diagnostic use and professional use only.
2- Read carefully the instructions before using this test.
3- Handle all specimens as if they contained infectious agents. When the assay procedure is completed, dispose of specimens carefully after autoclaving them for at least one hour. Alternatively, they can be treated with 0.5% to 1% solution of sodium hypochlorite for one hour before disposal.
4- Wear protective clothing such as laboratory coats and disposable gloves while assaying samples.
5- Do not eat, drink or smoke in the area where specimens and kit reagents are handled.
6- Avoid any contact between hands and eyes or nose during specimen collection and testing.
7- Do not use beyond the expiry date which appears on the package label.
8- Do not use a test from a damaged protective wrapper.

V- SPECIMEN COLLECTION AND PREPARATION

1-ULTRA-TSH-CHECK-1 test is to be performed on human serum or plasma.
2- The specimen should be collected under the standard laboratory conditions (aseptically in such a way as to avoid haemolysis).
3- If anticoagulant is needed, only citrate or EDTA should be used.
4- Each specimen should be treated as if potentially infectious.
5- If the test is to be run within 48 hours after collection the specimen should be stored in the refrigerator (+2°C to +8°C). If testing is delayed more than 48 hours, the specimen should be frozen. The frozen specimen must be completely thawed, thoroughly mixed and brought to room temperature prior to testing. Avoid repeated freezing and thawing.
6- In case of cloudiness, high viscosity or presence of particulate matter into the serum specimen, it should be diluted with equal volume (V/V) of diluting buffer (not provided but available upon request) before testing.

CE

VI- ASSAY PROCEDURE

a) Control testing

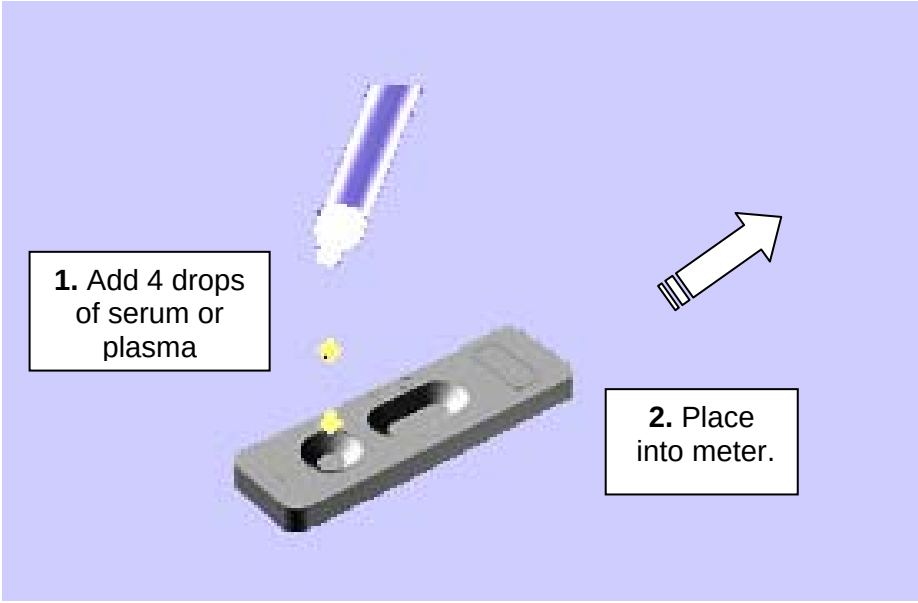
- Wait for 15 minutes after freeze-dried dissolving.
- Add the requested volume (150µL) with **lab pipette (disposable tips)** into the sample well of the cassette and proceed in the same way as for a patient's sample.
- The concentration range (**in mIU/L**) is indicated on the vial label and obtained result must be within the specified range. The confidence range can change slightly depending on lot number.
- **The reconstituted vial should be kept between +2°C and +8°C and should be used within 14 days after reconstitution**.

b) Samples testing

Follow the below instructions or refer to the picture n°1.

1. Allow specimen and ULTRA-TSH-CHECK-1 test device to come to room temperature prior to testing.
2. Remove the reaction device from its protective wrapper by tearing along the split.
3. Label device with the patient's name or control number.
4. Fill the serum dropper with specimen and by holding it vertically, dispense drop-wise into sample well (▷). Add exactly 4 drops (150µL without air bubble) of plasma or serum in the sample well (▷).
5. Read the result **(in mIU/L**) at 20 minutes exactly either using the immediate or countdown reading mode (see corresponding leaflet). In case the result reading is made at different time, wrong results will be obtained.

For general instructions describing how to use the VEDALAB's rapid test readers, refer to the corresponding leaflet.



Picture n°1

VII- PERFORMANCES CHARACTERISTICS

a) Linearity

The measuring range is 0.2– 50mIU/L.
For TSH concentration below 0.2mIU/L, the result will be given as "< 0.2mIU/L".
For TSH concentration over 50mIU/L, the result will be given as "> 50mIU/L".
For samples whose concentration is higher than 50 mIU/L, dilute with saline and repeat the assay as per instructions of Part VI.

b) Accuracy

A study has been performed using serum samples spiked with TSH W.H.O. reference material n°81/565 and preassayed samples covering a range of 0 to 50 mIU/L. Optical densities expressed as a function of TSH concentrations are described by following polynomial curve:

$$Y = -8.90 + 42.49\ X\ -1.40x^2 + 0.015x^3$$

The results show a good correlation (r > 92) of the values obtained with ULTRA-TSH-CHECK-1 on VEDALAB's reader.

c) Analytical sensitivity and abnormal values

The analytical sensitivity of the ULTRA-TSH-CHECK-1 test is 0.2mIU/L.
Levels higher than 5mIU/L could indicate hypothyroidism.
Levels lower than 0.3mIU/L could indicate hyperthyroidism.

d) Precision

Comparative studies were performed on a panel of 50 human sera using either the Roche Modular® or Cobas Elecsys® analysers. Results showed a coefficient of correlation 98.6% with the Roche instruments (COBAS/Modular®).

e) Cross reaction with homologous hormones

Homologous hormones were tested either without any added TSH or with added TSH (33mIU/L). Results are summarized in table I and showed no cross reaction.

Substances	References	Concentration	TSH Negative: <0.2 mIU/L	TSH Positive: 10 mIU/L
hCG	W.H.O 07/364	500 mIU/mL	<0.2	
		10,000 mIU/mL	<0.2	8.9
FSH	W.H.O 98/704	250 mIU/mL	<0.2	9
LH	W.H.O 80/552	1000 mIU/mL	<0.2	10.1

Table I: Hormones interference results

f) Interferences

1-Anticoagulants

Citrate and EDTA were not showing any interference while **heparin was inhibiting the detection and was therefore strongly interfering**.

2- Substances

Commonly encountered substances were tested, either with or without added TSH, to demonstrate that these substances do not interfere with ULTRA-TSH-CHECK-1 results. (Table II).

Substances	Concentration	TSH Negative < 5 mIU/L	TSH Positive > 5 mIU/L
Acetaminophen	20 mg/dL	Negative	Positive
Acetylsalicyclic Acid	20 mg/dL	Negative	Positive
Ampicillin	20 mg/dL	Negative	Positive
Ascorbic Acid	20 mg/dL	Negative	Positive
Atropin	20 mg/dL	Negative	Positive
Caffeine	20 mg/dL	Negative	Positive
Gentisic Acid	20 mg/dL	Negative	Positive
Glucose	2 mg/dL	Negative	Positive
Paracetamol	20 mg/dL	Negative	Positive
Tetracycline	20 mg/dL	Negative	Positive
Hemoglobin	1 mg/dL	Negative	Positive
Hematocrit Range	20 – 50 %	Negative	Positive

Table II: Common substances interference results

g) Intra-assay reproducibility

Within run precision was evaluated by using 20 replicates of 3 samples containing 0.6, 10 and 21.4mIU/L of TSH as determined with quantitative ULTRA-TSH-CHECK-1 for VEDALAB's reader.

The obtained CVs (coefficient of variation) were respectively equal to 13.79, 7.13 and 13.82%.

h) Hook effect

No hook effect was observed up to a TSH concentration of 1,000mIU/L. The reader result was shown as "> 50mIU/L".

VIII- LIMITATIONS

1- As for any diagnostic procedure, the physician should confirm the data obtained using this test by other clinical methods.

2- ULTRA-TSH-CHECK-1 rapid test is designed to quantify TSH within a range of 0.2mIU/L to 50mIU/L which is suitable for hypothyroidism and hyperthyroidism detection.

3- **Do not use heparinized samples which do not react in the test.**

4- Very rare cases of hypothyroidism with an associated low level of TSH or hyperthyroidism with an associated high level of TSH have been reported.

5- In early pregnancy, high level of TSH could be detected.

6- In case of doubtful results, additional T4 or T3 assays should be performed.

7- As it is true for any diagnostic method or for any measurements through analysers, there is a variability of the obtained result. Therefore, a confidence range of +/- 25% should be considered for the final value and for the clinical significance of the result.

8- High level of RF (Rheumatoid factor) or HAMA (Human anti-mouse antibodies) could give false positive result.

9- High level of CRP (C-reactive protein) indicates inflammatory process related to infection and thus increased concentration in poly-specific antibodies that could give false positive result in some cases.

10- This format of test is to be only used with VEDALAB's rapid test readers.

11- If the reading time (20 minutes) is not strictly respected, wrong results will be obtained.

12- This format of test should not be used for visual reading.

IX- BIBLIOGRAPHY

1- Burger, HG and Patel, YC (1977). Thyrotropin releasing hormone – TSH. Clin. Endocrin Metab, 6, 83-100.

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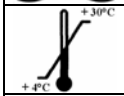
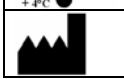
3- Thornes, HM, Mcleod, DT and Carr D (1987), Economy and efficiency in routine thyroid function testing: Use of a sensitive immunometric assay for thyrotropin in a general hospital laboratory. Clin Chem 33, 1635-1638.

4- Klee, GC and Hay, ID (1987), Assessment of sensitive thyrotropin assays for an expanded role in thyroid function testing: Proposed criteria for analytic performance and clinical utility. J Clin Endocrin Metab, **64**, 461 –471.

5- Synder, PJ and Utiger, RD (1972), Response to thyrotropin releasing hormone (TRH) in normal man. J. Clin Endocrin Metab, 3 4, 380-385.

	Read the instructions before use		For *in vitro* diagnostic use
	Temperature limitations		Do not reuse
	Manufacturer		

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