

# ULTRA-TSH-CHECK-1

## Ultra-Sensitive Quantitative determination of Thyroid Stimulating Hormone in serum samples

FOR EASY READER® AND EASY READER+® USE ONLY

Ref.: U21091 (20 tests) & Ref.: U21091-10T (10 tests)

### I- INTENDED PURPOSE

ULTRA-TSH-CHECK-1 is a rapid screening test for the detection of TSH in serum samples to be used as an aid by medical healthcare professionals in assessing thyroid function. The sole measurement of TSH concentration is not sufficient to diagnose thyroid disorders nor to predict thyroiditis evolution. Determination of other thyroid secreted hormones (T3, T4...) and additional clinical investigation are necessary to confirm the diagnosis.

### II- PRINCIPLE

Human thyroid stimulating hormone (TSH) is a glycoprotein secreted by the thyrotrope 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 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.

### III- 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  |
| <b>4- Controls (Optional): Positive control (ref. V2500U) and Negative control (ref. V2501U):</b> 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 0.5 mL). The concentration range is indicated on the vial label. |    |    |

### IV- STORAGE AND STABILITY

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

### V- PRECAUTIONS

- This test is designed for *in vitro* diagnostic use and professional use only.
- Read carefully the instructions before using this test.
- 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.
- Wear protective clothing such as laboratory coats and disposable gloves while assaying samples.
- Do not eat, drink or smoke in the area where specimens and kit reagents are handled.
- Avoid any contact between hands and eyes or nose during specimen collection and testing.
- Do not use beyond the expiry date which appears on the package label.
- Do not use a test from a damaged protective wrapper.

### VI- SPECIMEN COLLECTION AND PREPARATION

- ULTRA-TSH-CHECK-1 test is to be performed on human serum only.
- The specimen should be collected under the standard laboratory conditions (aseptically in such a way as to avoid haemolysis).
- Each specimen should be treated as if potentially infectious.
- 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.**
- 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.



## VII- ASSAY PROCEDURE

**IMPORTANT:** Switch the reader on and allow it to warm up for at least 30 minutes before performing any measurements.

### a) Control testing

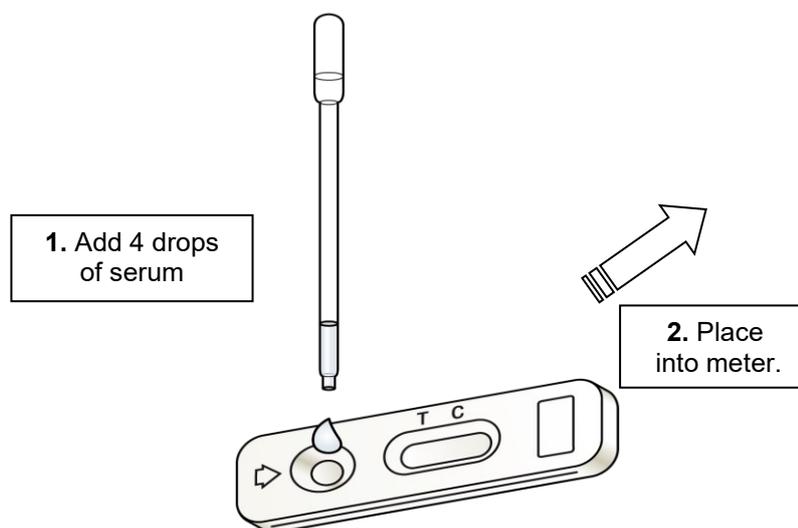
- Wait for 15 minutes after freeze-dried dissolving.
- Add the requested volume (150µL) with **lab pipette (disposable tips)** into the sample well of the cassette and proceed in the same way as for a patient's sample.
- The concentration range (in mIU/L) is indicated on the vial label and obtained result 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 7 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 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

## VIII- PERFORMANCES CHARACTERISTICS

### a) Linearity

A study has been performed using serum samples obtained from dilutions of TSH W.H.O reference material (International Standard n° 81/565) covering a range of 0 to 60 mIU/L. The dose response obtained with the ULTRA-TSH-CHECK-1 quantitative test fits a linear regression in the range of 0.2 to 50 mIU/L:

$$Y = 1.0017x - 0.0888$$

$$\text{Linear regression coefficient (R}^2\text{)} = 0.9998$$

The measuring range is 0.2– 50 mIU/L.

For TSH concentration below 0.2 mIU/L, the result will be given as “< 0.2 mIU/L”.

For TSH concentration over 50 mIU/L, the result will be given as “> 50 mIU/L”.

For samples whose concentration is higher than 50 mIU/L, dilute with saline and repeat the assay as per instructions of Part VII.

### b) Accuracy

Serial dilutions of TSH W.H.O. reference material (International Standard n°81/565) in TSH negative serum pool have been tested using ULTRA-TSH-CHECK-1 test (5 replicates each).

The obtained results are summarized in Table 1.

International standard theoretical concentration	TSH concentration (mIU/L)						
	0.47	0.94	1.88	3.75	7.5	15	30
Obtained results with ULTRA-TSH-CHECK-1 (Mean/5 replicates)	0.52	0.98	1.78	3.48	7.42	14.78	30.06
CV*	14.4 %	13.6 %	9.0 %	12.4 %	6.9 %	11.6 %	7.5 %
Bias	+10.6%	+4.3%	-5.4%	-7.2%	-1.1%	-1.5%	+0.2%

Table 1: accuracy

\*CV: Coefficient of Variation

The bias between nominal and measured values is statistically (95% t-test) non-significant and TSH concentrations determined using ULTRA-TSH-CHECK-1 test are accurately measured when compared to W.H.O. reference material.

### c) Analytical sensitivity

The analytical sensitivity of the ULTRA-TSH-CHECK-1 test is 0.2 mIU/L.

Levels higher than 5 mIU/L could indicate hypothyroidism.

Levels lower than 0.3 mIU/L could indicate hyperthyroidism.

### d) Diagnostic sensitivity, specificity and overall correlation

A panel of 73 human pre-assayed serum samples (Biomérieux VIDAS analyser) is assayed using the ULTRA-TSH-CHECK-1 quantitative test.

The obtained results (using VEDALAB reader) are reported in the table 1 (negative samples correspond to samples for which TSH concentration is  $\leq 0.2$  mIU/L and positive samples correspond to samples for which TSH concentration is  $> 0.2$  mIU/L).

		VIDAS		
		Positive	Negative	Total
ULTRA-TSH-CHECK-1	Positive	61	0	61
	Negative	2	10	12
	Total	63	10	73

Table 1: Summary of results

Diagnostic sensitivity =

$$\frac{61}{63} \times 100 = 96.8\% \text{ (CI* 95\% [88.4 - 100.0])}$$

63

Diagnostic specificity =

$$\frac{10}{10} \times 100 = 100\% \text{ (CI 95\% [70.79 - 100.0])}$$

10

Overall correlation =

$$\frac{(61+10)}{73} \times 100 = \frac{71}{73} \times 100 = 97.3\% \text{ (CI 95\% [89.9 - 100.0])}$$

73

\*CI 95%: 95% Confidence interval

On the other hand, the coefficient of correlation between quantified results of ULTRA-TSH-CHECK-1 test and VIDAS results is 97.7% (CI 95% [95.5 - 98.8]) for the measuring range of 0.2 to 25 mIU/L and 93.2% (CI 95% [88.6 - 96.0]) for the measuring range of 0.2 to 50 mIU/L.

### e) Analytical specificity (cross-reactivity)

There is no false positive results observed on ULTRA-TSH-CHECK-1 quantitative test for the following hormones (W.H.O international standards concentrations):

- hCG (Standard 99/688): 50,000 mIU/mL
- LH (Standard 81/535): 250 mIU/mL
- FSH (Standard 08/282): 250 IU/L

### f) Interferences

#### 1- Rheumatoid factor (RF)

A serum sample having a RF concentration of 1,862 IU/mL did not show any false positive results. Therefore, there is no interference up to a RF concentration of 1,862 IU/mL.

#### 2- HAMA

HAMA (anti-mouse human antibody) positive serum samples (type 1 or 2) did not show any false positive results. Therefore, there is no interference for HAMA type 1 and type 2 positive samples.

#### 3- Hemoglobin, bilirubin, triglycerides

Negative (0 mIU/L), weak positive (2 and 10 mIU/L) and strong positive (30 mIU/L) TSH samples, spiked with hemoglobin (final concentration: 1 g/L), bilirubin (final concentration: 7 mg/L) or triglycerides (final concentration: 7.5 g/L) did not show any effect on ULTRA-TSH-CHECK-1 quantitative test results (negative or positive).

### g) Hook effect

No hook effect has been observed up to 2,000 mIU/L TSH concentration.

### h) Intra-lot repeatability

Within run precision was evaluated by performing 20 replicates of 4 serum samples with different concentrations (0, 1, 10 and 30 mIU/L) using ULTRA-TSH-CHECK-1 quantitative test and VEDALAB reader. Coefficients of variations obtained for 1, 10 and 30 mIU/L positive samples are respectively 14.4%, 11.6% and 9.6%.

### i) Inter-lot reproducibility

Inter-lot variation was evaluated performing 3 replicates of 3 serum samples having different TSH concentrations (0, 2 and 30 mIU/L) using 3 different lots of ULTRA-TSH-CHECK-1 quantitative test and VEDALAB reader. Coefficients of variations obtained for 2 and 30 mIU/L positive samples are respectively 12.0% and 11.2%.

## IX- 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 0.2 mIU/L to 50 mIU/L which is suitable for hypothyroidism and hyperthyroidism detection.**

**3- Use serum samples only.**

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 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.

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

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

11- Do not use the reader for measurements before at least 30 minutes warm-up after having switched on.

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

13- High colored serum samples may produce weaker results.

## X- BIBLIOGRAPHY

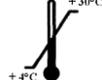
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**4- Klee, GC and Hay, ID (1987),** Assessment of sensitive thyrotropin assays for an expanded role in thyroid function testing: Proposed criteria for analytic performance and clinical utility. J Clin Endocrin Metab, 64, 461 –471.

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	Read the instructions before use		For <i>in vitro</i> diagnostic use
	Temperature limitations		Do not reuse
	Manufacturer		



Manufactured by VEDALAB – France

## CHANGES DESCRIPTION

Changes type:

- N/A Not Applicable (creation)
- Technical change Addition, revision and/or removal of information related to the product.
- Administrative Implementation of non-technical changes noticeable to the end-user.

Changes type	Change description
Technical change	Modifications in Chap VIII a), b), c), d), e), f), g), h) and i). Addition in Chap IX one limitation

**Note:** Minor typographical, grammar, spelling and formatting changes are not reported in the change details.