1. INTENDED USE
The kit has been designed for the quantitative determination of Thyroid-Stimulating Hormone (TSH) in human serum. The method can be used for samples over the range of 0-100 μIU/ml. The test has to be performed on the Maglumi fully auto analyzer (including Maglumi 1000, Maglumi 2000, Maglumi 2000 plus).

2. SUMMARY AND EXPLANATION OF THE TEST
Thyroid-stimulating hormone (TSH) is a glycoprotein hormone with a molecular weight in the range of 28,000-30,000 Dalton and is composed of the two non-covalently bound subunits αTSH and βTSH. A characteristic feature of the glycoproteins TSH, lutreinising hormone (LH), follicle-stimulating hormone (FSH), and human chorionic gonadotropin (hCG) is their relatively high carbohydrate content as well as the nearly identical sequential homology of their β-subunits (3, 4). On the other hand, the α-subunit has a different amino acid sequence in all four hormones. TSH release and synthesis in the anterior pituitary is stimulated by the hypothalamic thyrotropin releasing hormone (TRH). The TSH released stimulates the thyroid re release of the hormones thyroxine (T4) and triiodothyronine (T3) whose binding to transport proteins in the bloodstream exceed 99.9% and 99.7% respectively. Only the free hormones FT3 and FT4, which are not bound to binding proteins, are physiologically active in peripheral tissues and regulate the thyroid function via a pituitary feedback mechanism.

3. PRINCIPLE OF THE TEST
Sandwich immunoluminometric assay; Use an anti-TSH monoclonal antibody to label ABEI, and use another monoclonal antibody to label FITC. Sample, Calibrator, or Control, ABEI Label, FITC Label and magnetic microbeads coated with sheep anti-FITC are mixed thoroughly and incubated at 37 °C, forming a sandwich; after sediment in a magnetic field, decant the supernatant, then cycle washing for 1 time. Subsequently, the starter reagents are added and a flash chemiluminescent reaction is initiated. The light signal is measured by a photomultiplier as RLU within 3 seconds and is proportional to the concentration of TSH present in controls or samples.

4. KIT COMPONENTS
4.1 Material supplies
Reagent Integral for 100 determinations
Nano magnetic microbeads: TRIS buffer, 1.2%(W/V), 0.2%NaNO3, coated with sheep anti-FITC polyclonal antibody. 2.5ml
Calibrator low 3.0ml
Calibrator high 3.0ml
ABEI Label: anti-TSH monoclonal antibody labeled ABEI, contains BSA, 0.2%NaNO3. 6.5ml
FITC Label: anti-TSH monoclonal antibody labeled FITC, contains BSA, 0.2%NaNO3. 6.5ml
All reagents are provided ready-to-use.

4.2 Preparation of the Reagent Integral
Before the sealing is removed, gentle and careful horizontal shaking of the Reagent Integral is essential (avoid foam formation!). Remove the sealing and turn the small wheel of the magnetic microbeads compartment to and fro, until the colour of the suspension has changed into brown. Place the Integral into the reagent area and let it stand there for 30 mins. During this time, the magnetic microbeads are automatically agitated and completely resuspended.

4.3 Storage of the Reagents Integral
* Do not interchange magnetic microbeads from different lots

4.4 Storage of the Reagents Integral
- Sealed: Stored at 2-8 °C until the expiry date.
- Opened: Stable for 4 weeks. After this period, it is still possible to keep on using the Reagent Integral provided that the controls are found within the expected ranges.
- Keep upright for storage
13. Limitations of the procedure

13.1 Certain drugs may influence both basal TSH and TRH values. In pregnant women or in those taking ovulation, a normal TSH basal level reflects an euthyroid metabolic state, although total thyroxine(T4) and total triiodothyronine (T3) may be elevated. Serum TSH levels alone give no evidence of the presence or absence of thyroid disease. They must always be interpreted in context with the clinical picture and other diagnostic procedures.

13.2 HAMA Patient samples containing human anti-mouse antibodies (HAMA) may give falsely elevated or decreased values. Although HAMA-neutralising agents are added, extremely high HAMA serum concentrations may occasionally influence results.

13.3 High-Dose Hook No high-dose hook effect was seen for TSH concentrations up to 500 μIU/ml. If RLU value of the samples is higher than point J on the curve, the concentration calculated by the instrument is not necessarily accurate. For those samples, it is recommended to dilute them until a RLU value ranging between point A and point J is exhibited. After that, send them for a measurement. The output multiplied by dilution ratio equals the final RLU value of samples.

14. Performance Characteristics

14.1 Accuracy Consider calibrator high of known concentration as a sample, dilute it by 1:2 ratio with diluents, and measure its diluted concentration for 10 times. Then calculate the recovery of measured concentration and expected concentration. The recovery should be within 90% - 110%.

14.2 Precision Intra-assay coefficient of variation was evaluated on Calibrator High repeatedly measured 10 times in the same assay, calculating their coefficient of variation, the results should ≤10% for intra-assay coefficient of variation was evaluated on three batches of kit, repeatedly measured 10 times of Calibrator High, calculating three batches of kit for Calibrator High between the measured values of the coefficients of variation, the results should ≤15%

14.3 Sensitivity The sensitivity of the assay defined as the concentration of TSH equivalent to the mean RLU of 20 replicates of the zero standard plus two standard deviations corresponding to the concentration from the standard curve. The sensitivity is typically less than 0.10μIU/ml.

14.4 Specificity The specificity of the TSH assay system was assessed by measuring the apparent response of the assay to various potentially cross reactive analytes. When FSH=150 μIU/ml, the detection results of TSH <0.5μIU/ml When LH=200 μIU/ml, the detection results of TSH <1.0μIU/ml When HCG=500 μIU/ml, the detection results of TSH <5μIU/ml

14.5 Linearity Conduct a logarithmic transform to the RLU value and concentration value of 6 standards. After a double logarithmic fitting, the absolute value of its linearity should exceed 0.9800.

15. References


16. Testing Procedure

To ensure proper test performance, strictly adhere to the operating instructions of the Maglumi Fully Auto analyzer. Each test parameter is identified via a RFID tag on the Reagent Integral. For further information please refer to the Maglumi Fully Auto Operator’s Manual.

17. Quality Control

7. Keep away from direct sunlight.

5. Origin of Calibrators. Calibrators in the Reagent Kit are from Fitzgerald. Biological root: synthetic materials, processed by SDS-PAGE purification, with a purity ≥95%. No HBsAg, anti-HCV, and anti-HIV is found.

6. Calibration

6.1 2 point recalibration

Via the measurement of calibrators, the predefined master curve is adjusted (recalibrated) to a new, instrument-specific measurement level with each calibration.

6.2 Frequency of Recalibration

After each exchange of lot (Reagent Integral or Starter Reagents) after two weeks and/or each time a new Integral is used (recommendation) after each servicing of the Maglumi Fully Auto analyzer if controls are beyond the expected range.

7. Sample Collection, Material and Storage

Collect samples using standard procedures.

Sample material: serum

Store at 2-8℃: 24 hours

For longer storage periods: freeze to below -20℃

Avoid repeated freezing and thawing cycles

Stored samples should be thoroughly mixed prior to use (Vortex mixer)

Vacuum tubes

(a) Blank tubes are recommended type for collecting samples.

(b) If plasma sample is needed, EDTA tube is confirmed has no effect on the results RLU.

(c) Liquiuenin Sodium tube is found to increase the sample RLU and cause test results deviation.

(d) Please ask SNIBE for advice if special additive must be used in the sample blood.

8. Interfering Substances

No interference with test results is seen by concentrations of bilirubin <0.125mg/ml, haemoglobin <16mg/dl or triglycerides <12.5mg/ml.

9. WARNING AND PRECAUTIONS FOR USERS

Keep away from direct sunlight.

For use in IN-VITRO diagnostic procedures only.

Do not interchange reagents from different lots. Do not use kit components beyond their labeled expiry date.

All samples, biological reagents and materials used in the assay must be considered potentially able to transmit infectious agents. They should therefore be disposed of in accordance with the prevailing regulations and guidelines of the agencies holding jurisdiction over the laboratory, and the regulations of each country. Disposable materials must be incinerated; liquid waste must be decontaminated with sodium hypochlorite at a final concentration of 5% for at least half an hour. Any materials to be reused must be autoclaved using an overkill approach (USP 24,2000,p.2143). A minimum of one hour at 121℃ is usually considered adequate, though the users must check the effectiveness of their decontamination cycle by initially validating it and routinely using biological indicators.

The calibrators in this kit are prepared from bovine serum products. However, because no test method can offer complete assurance that HIV, Hepatitis B Virus or other infectious agents are absent, these reagents should be considered a potential biohazard and handled with the same precautions as applied to any serum or plasma specimen.

10. Test Procedure

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11. Quality Control

Observe quality control guidelines for medical laboratories.

Use suitable controls for in-house quality control.

12. Results

12.1 Calculation of Results

The analyzer automatically calculates the TSH concentration in each sample by means of a calibration curve which is generated by a 2-point calibration master curve procedure. The results are expressed in μIU/ml. For further information please refer to the Maglumi Fully Auto Operator’s Manual.

12.2 Interpretation of Results

Reference values: 0.4-4.5 μIU/ml

Results may differ between laboratories due to variations in population and test method. Each laboratory should establish its own reference range.

12.3 Accuracy

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12.4 Precision

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