MAGLUMI ACTH (CLIA)

REF 130298003M

Σ 100

EC REP

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CE

FOR PROFESSIONAL USE ONLY
Store at 2-8 °C

COMPLETELY READ THE INSTRUCTIONS BEFORE PROCEEDING

en

SYMBOLS EXPLANATIONS

EC REP	Authorized Representative in the European community
	Manufacturer
	Consult instructions for use
CONT	Contents of kit
IVD	In vitro diagnostic medical device
LOT	Batch code
REF	Catalogue number
	Use by
8°C 2°C	Temperature limitation (store at 2-8 °C)
Σ	Sufficient for
	Keep away from sunlight
	Keep upright for storage

INTENDED USE

The kit has been designed for the quantitative determination of Adrenocorticotropic Hormone (ACTH) in human serum or plasma.
The method can be used for samples over the range of 3.0-2000pg/ml.
The test has to be performed on the Fully-auto chemiluminescence immunoassay (CLIA) analyzer MAGLUMI (Including Maglumi 600,Maglumi 1000,Maglumi 1000 Plus, Maglumi 2000,Maglumi 2000 Plus,Maglumi 3000 and Maglumi 4000).

SUMMARY AND EXPLANATION OF THE TEST

Adrenocorticotropic hormone (ACTH) is a polypeptide hormone which exists principally as a chain, 39 amino acids long, with a molecular mass of appro. 4500 daltons. It is produced in the pituitary and serves to stimulate steroid production by the adrenal cortex. ACTH secretion is in turn controlled by the hypothalamic hormone corticotrophin releasing factor (CRF) and by negative feedback from cortisol.
ACTH determinations are valuable in the differential diagnosis of adrenal insufficiency and hypersecretion. In Addison's disease (primary adrenal insufficiency), elevated levels are typical, whereas low levels are the rule when adrenal insufficiency is secondary to pituitary dysfunction. ACTH determinations can also help to identify the cause of cortisol hypersecretion in Cushing's syndrome. ACTH levels are typically low when this is due to lesions or hyperplasia of the adrenal cortex, and high when it is due to ectopic ACTH production or hypersecretion of ACTH by the pituitary.
Plasma levels of ACTH exhibit a significant diurnal variation. It is important, therefore, to standardize the time of collection: reference ranges have typically been established for approximately 9 in the morning.

PRINCIPLE OF THE TEST

Sandwich immunoluminometric assay;
Use an anti-ACTH monoclonal antibody to label ABEI and use another monoclonal antibody to label microbeads. Sample, Calibrator, or Control with ABEI Label and magnetic microbeads coated with monoclonal antibody are mixed thoroughly and incubated at 37℃, 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 ACTH present in samples.

CONT KIT COMPONENTS

Material Supplies

Reagent Integral for 100 determinations	
Nano magnetic microbeads: coated with anti-ACTH monoclonal antibody, contains BSA, 0.2%NaN_3	2.5ml
Calibrator Low: bovine serum, 0.2%NaN_3	3.0ml
Calibrator High: bovine serum, 0.2%NaN_3	3.0ml
ABEI Label: anti-ACTH monoclonal antibody labeled ABEI contains BSA, 0.2%NaN_3.	22.5ml
All reagents are provided ready-to-use.	

Reagent Vials in kit box	
Internal Quality Control: containing BSA,	2.0ml

0.2%NaN_3. (target value refer to Quality Control Information date sheet)	

Internal quality control is only applicable with MAGLUMI system. Instructions for use and target value refer to Quality Control Information date sheet. User needs to judge results with their own standards and knowledge.

Accessories Required But Not Provided

MAGLUMI Reaction Module	REF: 630003
MAGLUMI Starter 1+2	REF: 130299004M
MAGLUMI Wash Concentrate	REF: 130299005M
MAGLUMI Light Check	REF: 130299006M

Please order accessories from SNIBE or our representative.

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 color of the suspension has changed into brown. Place the Integral into the reagent area and let it stand there for 30 min. During this time, the magnetic microbeads are automatically agitated and completely resuspended.

Do not interchange integral component from different reagents or lots!

Storage and Stability

- Sealed: Stored at 2-8℃ until the expiry date.
- Opened: Stable for 4 weeks. To ensure the best kit performance, it is recommended to place opened kits in the refrigerator if it's not going to be used on board during the next 12 hours.
-  Keep upright for storage.
-  Keep away from sunlight.

CALIBRATION AND TRACEABILITY

1)Traceability

To perform an accurate calibration, we have provided the test calibrators standardized against the Non WHO Reference Material Corticotrophin (ACTH), Human 74/555.

2) 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.

3) Frequency of Recalibration

- After each exchange of lot (Reagent Integral or Starter Reagents).
- Every week and/or each time a new Integral is used (recommendation).
- After each servicing of the Fully-auto chemiluminescence immunoassay (CLIA) analyzer MAGLUMI..
- If controls are beyond the expected range.
- The room temperature has changed more than 5 ℃ (recommendation).

SPECIMEN COLLECTION AND PREPARATION

Sample material: serum

Collect samples using standard procedures.

Collect 5.0ml venous blood into tube, and stored in ice-bath, use low-temperature centrifuge to centrifuge to separate the serum from the rest and put the serum into - 20℃ for storage. Avoid hemolysis, which may affect the result.

Serum specimen was stable at room temperature for 2 hours, And store at 2-8℃: 5 hours, for longer storage periods(4 weeks): freeze to below - 20℃. Serum sample can only be frozen for once, avoid freezing repeatedly.

Sample material: plasma

Collect blood with anticoagulation blood tube (EDTA-K_2), then put the tube into ice-bath, use low-temperature centrifuge to centrifuge to separate the plasma from the rest and put the serum into - 20℃ for storage.

Plasma specimen was stable at room temperature for 3 hours, And store at 2-8℃: 8 hours, for longer storage periods（4 weeks）: freeze to below - 20℃.

Avoid repeated freezing and thawing cycles, stored samples should be thoroughly mixed prior to use (Vortex mixer).

Please ask local representative of SNIBE for more details if you have any doubt.

Vacuum Tubes

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

(b) Please ask SNIBE for advice if special additive must be used in sample collecting.

Specimen Conditions

- Do not use specimens with the following conditions:
  (a) heat-inactivated specimens;
  (b) Cadaver specimens or body fluids other than human serum;
  (c) Obvious microbial contamination.
- Use caution when handling patient specimens to prevent cross contamination. Use of disposable pipettes or pipette tips is recommended.
- Inspect all samples for bubbles. Remove bubbles with an applicator stick prior to analysis. Use a new applicator stick for each sample to prevent cross contamination.
- Serum specimens should be free of fibrin, red blood cells or other particulate matter.
- Ensure that complete clot formation in serum specimens has taken place prior to centrifugation. Some specimens, especially those from patients receiving anticoagulant or thrombolytic therapy, may exhibit increased clotting time. If the specimen is centrifuged before a complete clot forms, the presence of fibrin may cause erroneous results.

Preparation for Analysis

- Patient specimens with a cloudy or turbid appearance must be centrifuged prior to testing. Following centrifugation, avoid the lipid layer (if present) when pipetting the specimen into a sample cup or secondary tube.
- Specimens must be mixed **thoroughly** after thawing by **low** speed vortexing or by gently inverting, and centrifuged prior to use to remove red blood cells or particulate matter to ensure consistency in the results. Multiple freeze-thaw cycles of specimens should be avoided.
- All samples (patient specimens and controls) should be tested within 3 hours of being placed on board the MAGLUMI System. Refer to the SNIBE service, for a more detailed discussion of onboard sample storage constraints.

Storage

- If testing will be delayed for more than 8 hours, remove serum or plasma from the serum or plasma separator, red blood cells or clot. Serum specimens removed from the separator gel, cells or clot may be stored up to 5 hours at 2-8°C. Plasma specimens removed from the separator gel, cells or clot may be stored up to 8 hours at 2-8°C.
- Serum specimens can be stored up to 4 weeks frozen at -20°C or colder.

Plasma specimens can be stored up to 8 weeks frozen at

-20°C or colder.

Shipping

- Before shipping specimens, it is recommended that specimens be removed from the serum or plasma separator, red blood cells or clot. When shipped, specimens must be packaged and labeled in compliance with applicable state, federal and international regulations covering the transport of clinical specimens and infectious substances. Specimens must be shipped frozen (dry ice). Do not exceed the storage time limitations identified in this section of the package insert.

WARNING AND PRECAUTIONS FOR USERS

IVD

- For use in *IN-VITRO* diagnostic procedures only.
- Package insert instructions must be carefully followed. Reliability of assay results cannot be guaranteed if there are any deviations from the instructions in this package insert.

Safety Precautions

CAUTION: This product requires the handling of human specimens.

- 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.
- 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. 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.
- It is recommended that all human sourced materials be considered potentially infectious and handled in accordance with the OSHA Standard on Bloodborne Pathogens 13. Biosafety Level 214 or other appropriate biosafety practices should be used for materials that contain or are suspected of containing infectious agents.
- This product contains Sodium Azide; this material and its container must be disposed of in a safe way.
- Safety data sheets are available on request.

Handling Precautions

- Do not use reagent kits beyond the expiration date.
- Do not mix reagents from different reagent kits.
- Prior to loading the Reagent Kit on the system for the first time, the microbeads requires mixing to re-suspend microbeads that have settled during shipment.
- For microbeads mixing instructions, refer to the KIT COMPONENTS, Preparation of the Reagent Integral section of this package insert.
- To avoid contamination, wear clean gloves when operating with a reagent kit and sample.
- Over time, residual liquids may dry on the kit surface, please pay attention the silicon film still exists on the surface of the kit.
- For a detailed discussion of handling precautions during system operation, refer to the SNIBE service information.

TEST PROCEDURE

To ensure proper test performance, strictly adhere to the operating instructions of the Fully-auto chemiluminescence immunoassay (CLIA) analyzer MAGLUMI. Each test parameter is identified via a RFID tag on the Reagent Integral. For further information please refer to the Fully-auto chemiluminescence immunoassay (CLIA) analyzer MAGLUMI Operating Instructions.

200μl +200μl +20μl	Sample, calibrator - ABEI Label Nano magnetic microbeads
30 min	Incubation
400μl	Cycle washing
3 s	Measurement

DILUTION

Sample dilution by analyzer is not available in this reagent kit.
Samples with concentrations above the measuring range can be diluted manually. After manual dilution, multiply the result by the dilution factor.
Please choose applicable diluents or ask SNIBE for advice before manual dilution must be processed.

QUALITY CONTROL

- Observe quality control guidelines for medical laboratories
- Use suitable controls for in-house quality control. Controls should be run at least once every 24 hours when the test is in use, once per reagent kit and after every calibration. The control intervals should be adapted to each laboratory's individual requirements. Values obtained should fall within the defined ranges. Each laboratory should establish guidelines for corrective measures to be taken if values fall outside the range.

LIMITATIONS OF THE PROCEDURE

1) Limitations

Patients with malignancies may exhibit ACTH values within the normal range. ACTH concentrations may be elevated in case of liver cirrhosis, hepatitis or tyrosinaemia. Thus, ACTH determination is more suitable for therapeutic monitoring and follow-up as well as for a comparison with histological results. ACTH serum levels may only be interpreted in context with the clinical picture and other diagnostic procedures. The ACTH assay should not be used as the only criterion for cancer screening.

2) Interfering Substances

No interference with test results is seen by concentrations of bilirubin<25mg/dl, haemoglobin<400mg/dl or triglycerides< 1500mg/dl.

3) HAMA

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

4) High-Dose Hook

No high-dose hook effect was seen for ACTH concentrations up to 50,000pg/ml.

RESULTS

1) Calculation of Results

- The analyzer automatically calculates the ACTH 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 pg/ml. For further information please refer to the Fully-auto chemiluminescence immunoassay (CLIA) analyzer MAGLUMI Operating

Instructions.

2) Interpretation of Results

- Reference values：

8:00-10:00, 6-40 pg/ml
16:00, 3-30 pg/ml
24:00, <20 pg/ml

- Results may differ between laboratories due to variations in population and test method. If necessary, each laboratory should establish its own reference range.

PERFORMANCE CHARACTERISTICS

1) Precision

Intra-assay coefficient of variation was evaluated on 3 different levels of control serum repeatedly measured 20 times in the same run, calculating the coefficient of variation.

Intra-assay precision			
Control	Mean(pg/ml)	SD(pg/ml)	CV%
Level 1	61.42	3.35	5.46
Level 2	262.35	12.70	4.84
Level 3	530.75	24.15	4.55

Inter-assay coefficient of variation was evaluated on three batches of kits. Repeatedly measured 3 different levels of control serum 21 times, calculating the coefficient of variation.

Inter-assay precision			
Control	Mean(pg/ml)	SD(pg/ml)	CV%
Level 1	59.35	5.09	8.57
Level 2	260.47	22.19	8.52
Level 3	528.46	45.18	8.55

2) Analytical Sensitivity

The sensitivity is defined as the concentration of ACTH 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 3.0pg/ml.

3) Specificity

The specificity of the ACTH assay system was assessed by measuring the apparent response of the assay to various potentially cross reactive analytes.

Compound	Concentration	Cross reactivity
BSA	50 µg/ml	0.6%

4) Recovery

Consider calibrator high of known concentration as a sample, dilute it by 1:2 ratios 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%.

Expected	Mean Measuring	Recovery
630.6 pg/ml	642.5 pg/ml	102%

5) Linearity

Use ACTH calibrator to prepare the six-point standard curve, measuring all points' RLU except point A, and then do four-parameter linear fitting in double logarithm coordinate, the absolute linear correlation coefficient(r) should be bigger than 0.9800.

Calibrator Point	Concentration pg/ml	Absolute linear correlation coefficient (r)
A	0.0	
B	50.0	r=0.9963
C	200.0	
D	400.0	
E	1000.0	
F	2000.0	

6) Method comparison

A comparison of MAGLUMI ACTH (y) with a commercially available ACTH test (x) using clinical samples gave the following correlations (pg/ml):

Linear regression
y = 1.0996x+20.929
r = 0.9967

Number of samples measured: 100
The sample concentrations were between 0.043 and 1098.39 pg/ml.

REFERENCES

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