In vitro rapid detection test of antibodies specific to Trypanosoma bruceli gambiense in human whole blood, serum or plasma

FOR IN VITRO USE
FOR PROFESSIONAL USE ONLY
References: K-15SS2, 40 tests per kit, with accessories K-12SS, 40 tests per kit, without accessories

I. INTRODUCTION

Human African trypanosomiasis (HAT) or sleeping sickness is a life threatening neglected tropical infection affecting rural populations in sub-Saharan Africa. In west and central Africa, the chronic form of sleeping sickness is caused by Trypanosoma bruceli (T.b.) gambiense, a protozoan parasite. It is transmitted by the hematophagus Tsetse fly by biting. HAT causes severe neurological disorders often leading to death if not treated. In addition, affected persons can act as reservoir hosts.

With the steadily decreasing prevalence of HAT, an individual rapid detection test with high specificity that is stable at ambient temperature and can be performed after minimal training is needed.

II. PRINCIPLE OF THE TEST

HAT Sero K-Set test is a ready-to-use lateral-flow test based on a membrane technology. A nitrocellulose membrane is sensitized to catch antibodies of the samples and these are revealed with a colloidal gold conjugate. As antigens, it contains Trypanosoma bruceli Variable Antigen Types LiTat 1.3 and LiTat 1.5. The sample must be delivered directly in the sample well of the device. When 2 drops of buffer are subsequently added to the same sample well, migration occurs on the strip. Then the solubilised conjugate migrates with the sample by passive diffusion and the conjugate and sample material come into contact with the reagent adsorbed onto the nitrocellulose. If the sample contains anti-T.b.gambiense antibodies, the conjugate-antibodies complex will remain bound to the test line and a red line will develop. Solution continues to migrate to encounter a second reagent that binds the migration control conjugate, thereby producing a red control line confirming that the test is working properly. The result is visible in the reading window within 15 minutes.

III. REAGENTS AND MATERIALS

1. Gambiense Sero K-Set (40)
   Sealed pouches each containing one device and one desiccant. Each device contains one sensitized strip.
2. Instruction for use (1)
3. Procedure card (1)
4. BL-A buffer (6 mL)
   Saline dilution buffered to pH 7.5 containing Tris, EDTA, NaN3 (<0.1%), a detergent and blocking proteins.
5. Heparinized capillary tubes (50)
6. Capillary micropipette (2)
7. Materials needed (supplied with item K-15SS2)
   - Blood lancets
   - Alcohol prep pads
   - Gloves
   - Sterile cotton pads

IV. SPECIAL PRECAUTIONS

- All operations linked to the use of the test must be performed in accordance with Good Laboratory Practices (GLP).
- The user has to ensure that the technician who is performing the test is trained to handle the provided accessories.
- All reagents are for in vitro diagnostic use only.
- Pouch must be opened with care at the moment of the test.
- Avoid touching nitrocellulose with your fingers.
- Wear gloves when handling samples.
- Never use reagents or buffer from another kit.
- Green lines indicate reagents adsorption sites. Green color disappears during the test.
- Reagents’ quality cannot be guaranteed beyond their shelf-life dates or if reagents are not stored under required conditions as indicated on the pouch containing the device.

V. WASTE DISPOSAL

- Dispose of gloves, lancets, capillary tubes, alcohol wipes and used devices in accordance with GLP.
- Each user is responsible for the management of any waste produced, and must ensure that it is disposed of in accordance with the applicable legislation.

VI. STORAGE

An unopened pouch may be kept at between 4 and 30 °C and used until the shelf-life date indicated on the packaging. Once the pouch is opened, run the test immediately.

- Do not freeze devices and buffer.

VII. SPECIMEN HANDLING AND COLLECTION

Specimen should be tested as soon as possible after collection. The whole blood specimen must be tested extemporaneously after taking. Serum or plasma may be stored at 2-8°C for 1 week or -20°C for longer periods of time. There is no cross-reactivity with EDTA, heparin or citrate.

VIII. PROCEDURE

Preparations of the test:
1. Allow kit components, in unopened packaging, and specimens to reach room temperature before performing a test.
2. Open the pouch. Once opened, run the test immediately.
3. Indicate the patient’s name or specimen number on the device (one device per sample).
4. Check that the two green lines are present in the reading window. If not, take another device.
5. For the next phases of the test, slide the device partially into the pouch so that the sample well is visible but the reading window is hidden.

Preparations of fingertip blood:
1. Prick the previously disinfected patient’s fingertip with a micro-lancet.
2. Wipe away the first drop of blood with a sterile cotton pad.
3. Fill to the end the heparinized capillary tube provided with the kit. Fill volume is approximately 25 μL. Avoid introducing any air bubbles into the capillary tube.

Performing the test:
1. A. Whole blood:
   Take 25 μL of blood with a capillary tube (fingertip or venous blood – capillary micropipette provided) or a pipette (venous blood only - not provided) and dispense into the inner side of the device sample well as illustrated below (annotated “1” area).
2. B. Serum or plasma:
   Take 15 μL of serum or plasma and dispense into the inner side of the sample well of the Gambiense Sero K-Set (sampling device not provided) as illustrated below (annotated “2” area). In order to obtain uniform drops, don’t touch the membrane of the sample well with the vial dispenser and hold it vertically.
3. Slide the whole device into the pouch to avoid evaporation of the buffer and to maximize the test efficiency.
4. Leave to react for 15 minutes into the pouch.
5. After 15 to 20 minutes, remove the device from the pouch. The results are observed in the reading window.

**WARNINGS**

If the sample is taken using a capillary tube, the time between the sampling and the deposit cannot exceed 30 minutes. Test must be read immediately after its removal from the pouch. Do not take the appearance of new lines into account after the reaction time is passed. Do not use contaminated capillary pipette.

**IX. INTERPRETING RESULTS**

The results are to be interpreted as follows:

- **Positive test result:** in addition to a reddish-purple band at the Control line (C), a visible reddish-purple band appears at the Test line position (T). Intensity of the Test line may vary. Any reddish-purple line (T), even weak, should be considered as a positive result.
- **Negative test result:** a reddish-purple line appears across the central reading window at the Control line (C) position. No other band is present.
- **Invalid test result:** The absence of a Control line indicates a failure in the test procedure, even if a Test line is present. Repeat invalid tests with a new test device. Note: during the drying process, which begins after 20 minutes of running, a faint shadow may appear at the Test line position. It should not be regarded as a positive result.

**X. PERFORMANCE**

A. **Sensitivity - Specificity**

If possible, keep the sample in the freezer within a day after the collection. For samples taken using a capillary tube, leave the capillary pipette in the pouch until the test result is observed. Do not take the appearance of new lines into account after the reaction time is passed.

**XII. TECHNICAL PROBLEMS / COMPLAINTS**

If you encounter a technical problem or if performances do not correspond with those indicated in this package insert:

1. Record the kit batch number.
2. If possible, keep the clinical sample in the freezer during the complaint management.
3. Contact Coris BioConcept (client.care@corisbio.com) or your local distributor.

**XIII. BIBLIOGRAPHIC REFERENCES**


---