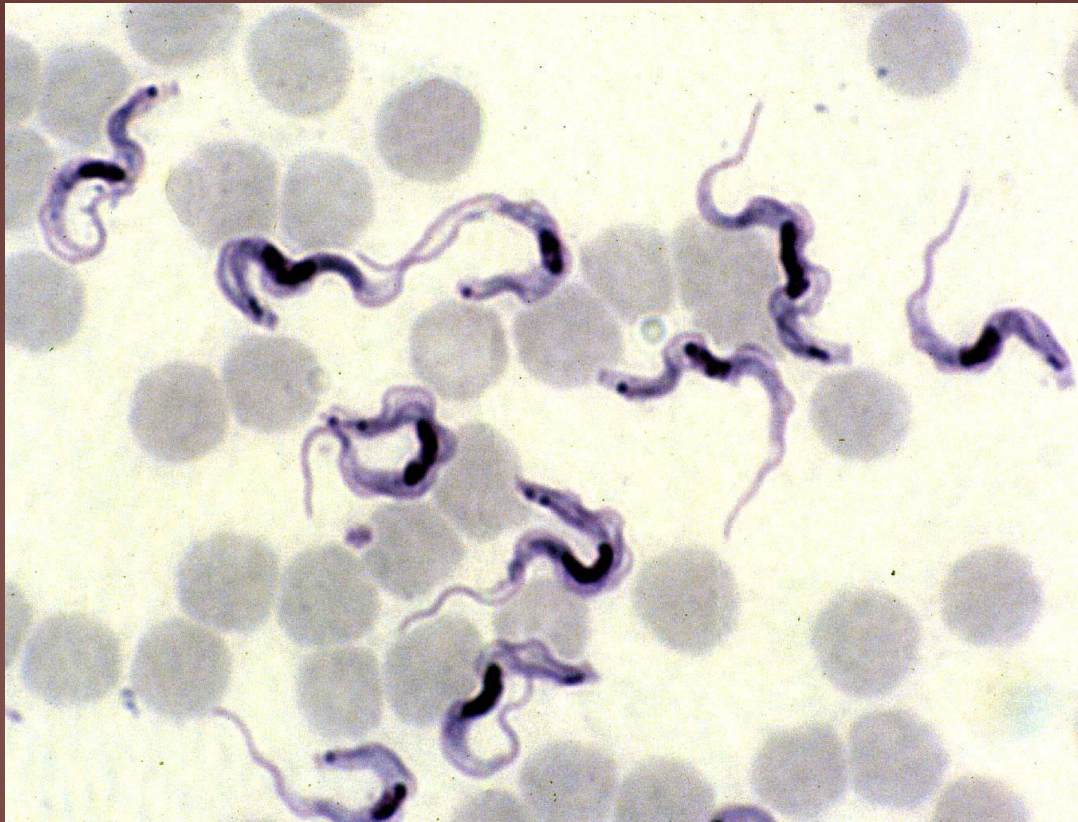


การวินิจฉัยโรค SURRA ในม้า ด้วยวิธี Card Agglutination Test



INTRODUCTION

“SURRA”

- Causative agent: *Trypanosoma evansi*
- Species exhibiting → camels, horses, deer, buffalo, pigs, cattle, dogs, jaguars and tigers
- disease Vector → Horseflies (Tabanus sp.)
Stable flies (Stomoxys sp.)
- Distribution → Africa,
Middle East
Asia
America



Horseflies

INTRODUCTION

Clinical sign → acute, subacute or chronic

weight loss, progressive anemia
fluctuating body temperature
abortion, hind limb weakness,
ventral and peripheral edema.

Diagnosis → - Identification of the agent by direct microscopic examination

- Serological examination;

- ab-ELISA

- card agglutination test (CATT)

- immunofluorescent antibody test (IFAT)

The card agglutination test

- The Card Agglutination Test for Trypanosomiasis (CATT)
- developed at the Laboratory of Serology, Institute of Tropical Medicine, Antwerp
- for the detection of antibodies against several surface antigens of parasite
- Can demonstrate in plasma or serum
- diluted serum or plasma are mixed with antigen
- agglutination will show if antibodies present in the sample

CHAPTER 2.1.17. TRYPANOSOMA EVANSI INFECTIONS (INCLUDING SURRA)

Infection gives rise to specific antibody responses and a variety of antibody detection tests have been introduced for laboratory and field use. Among those that are used regularly in the laboratory are immunoenzyme assays, card agglutination tests and latex agglutination tests. For field use both card agglutination tests (CATT) for *T. evansi* and latex can be applied.

Assays for detection of circulating antibodies have high measures of validity. Estimates of predictive values of different serological tests indicate that enzyme linked immunosorbent assays (ELISA) for detecting IgG antibodies are more likely to classify correctly uninfected animals, and CATT are more likely to classify correctly truly infected animals.

In situations where there is overt disease, CATTs can be used to target individual animals for treatment with trypanocidal drugs. For declaring a disease-free status, serial testing – ELISA followed by re-testing of suspect samples by CATT – is recommended.

LOT

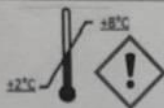
KU-2017-1



INSTITUTE
OF TROPICAL
MEDICINE
ANTWERP

CATT / *T.evansi*
REACTIFS / REAGENTS

IVD



LOT

KU-2017-1



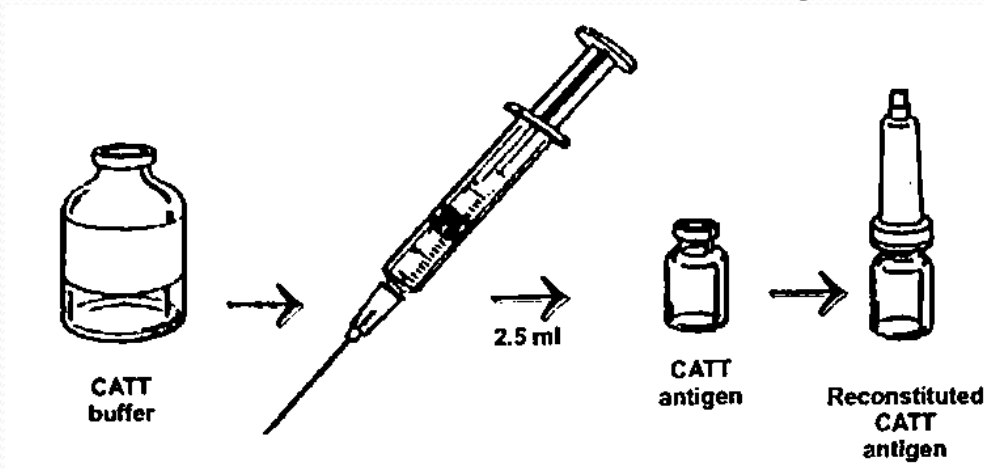
INSTITUTE
OF TROPICAL
MEDICINE
ANTWERP

CATT / *T.evansi*
ACCESSOIRES / ACCESSORIES



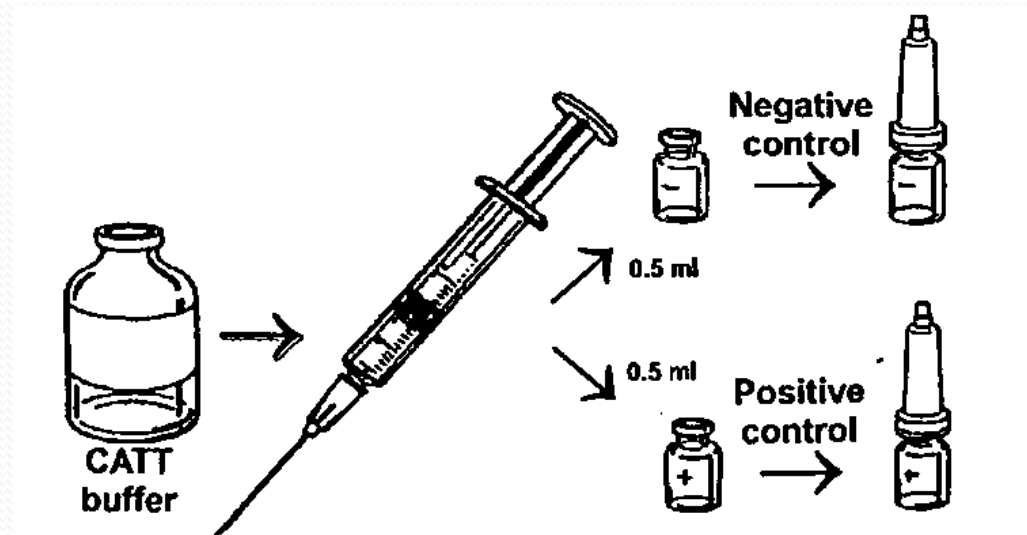
EXECUTION OF THE TEST

1. Reconstitution of the CATT antigen



Can be use during 1 week when store between +2 °C and 8 °C

1. Reconstitution of the control



DO NOT FREEZE

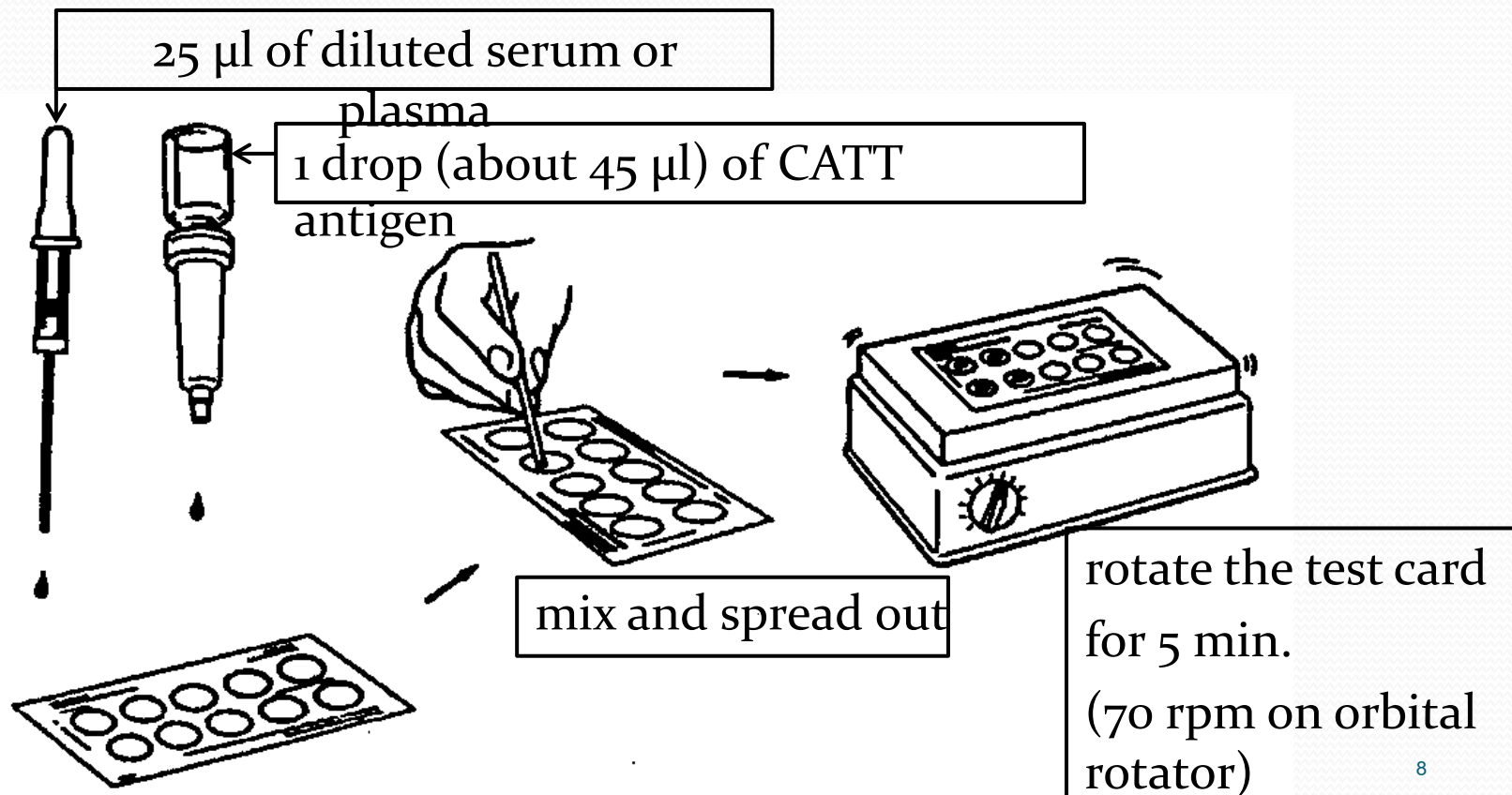
DO NOT FREEZE

EXECUTION OF THE TEST

3. Preparation of test sample

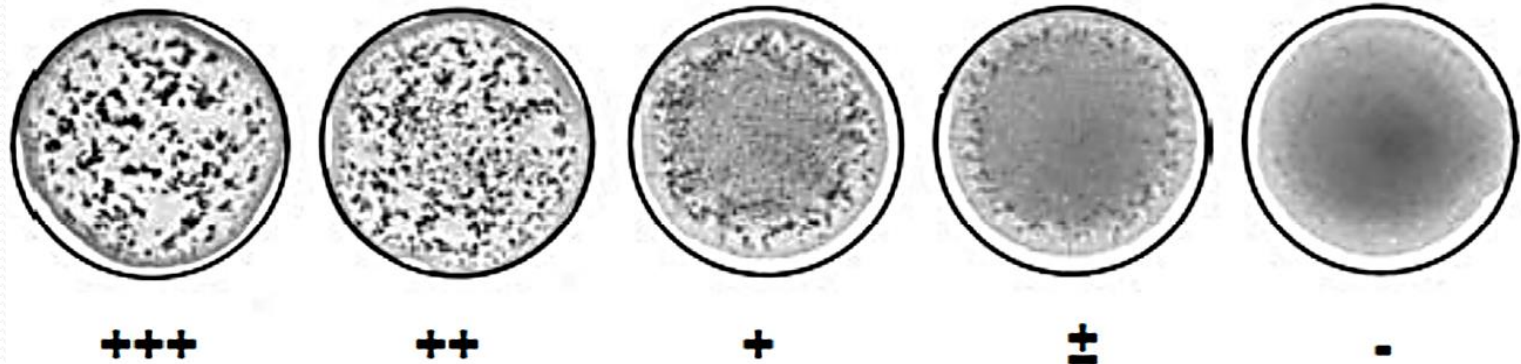
prepare a 1:4 or 1:8 dilution in CATT BUFFER

4. Agglutination reaction

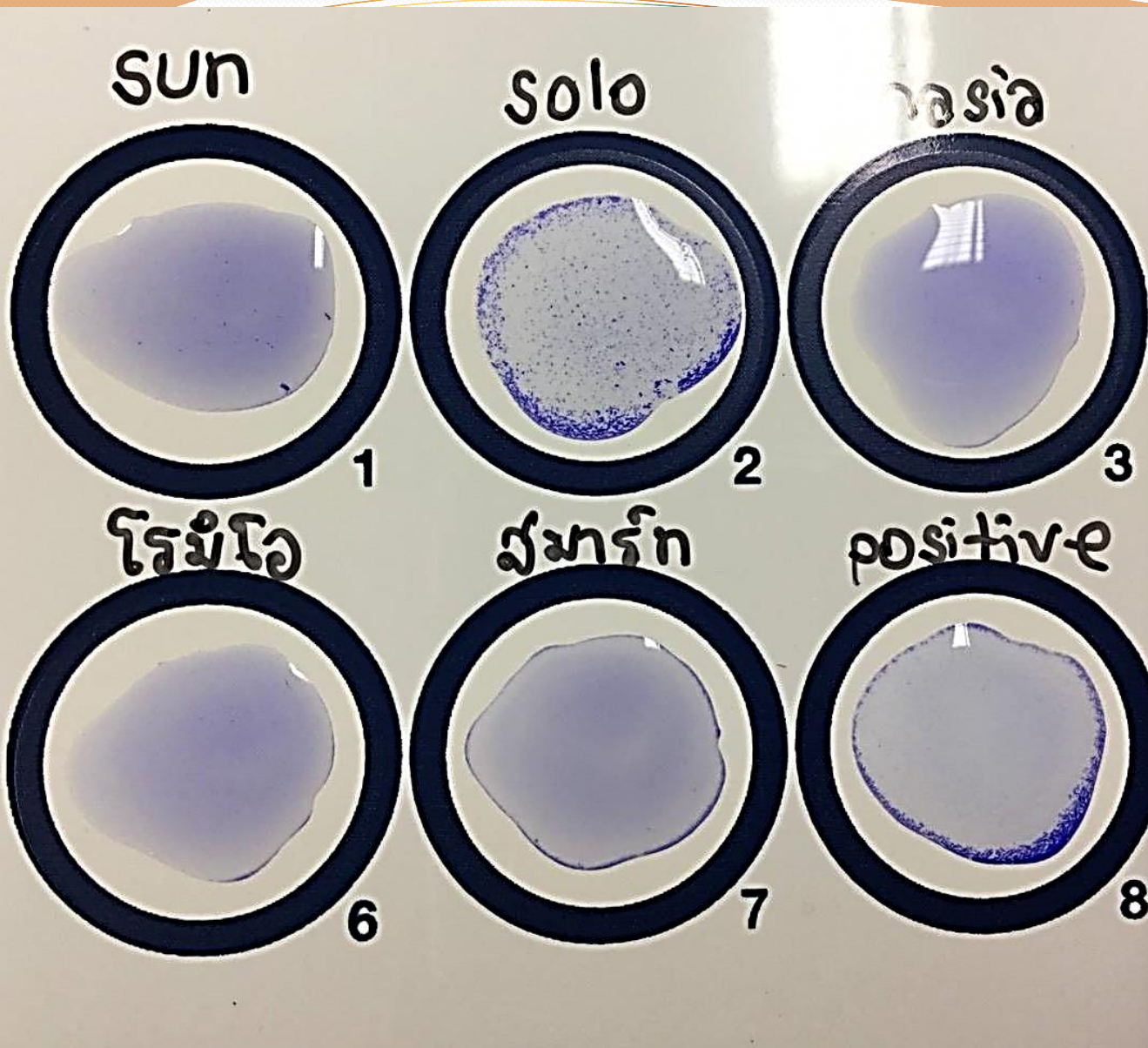


EXECUTION OF THE TEST

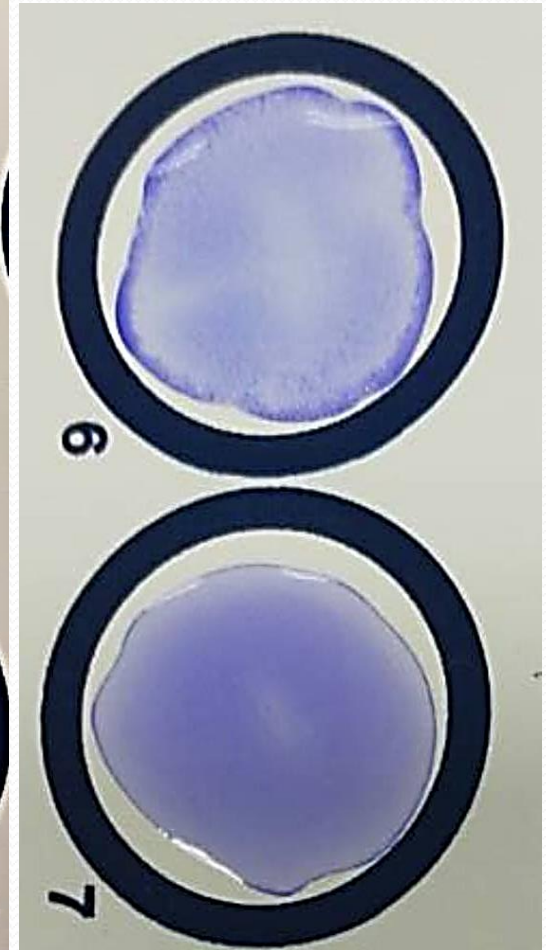
after 5 minutes, read the results before removing card from rotator or stop from tilting
reading the results as follow:



- +++ = STRONGLY POSITIVE** (very strong agglutination)
- ++ = POSITIVE** (strong agglutination)
- +** = **POSITIVE** (moderate agglutination)
- ±** = **WEAKLY POSITIVE** (weak agglutination)
- = **NEGATIVE** (absence of agglutination)



Positive control



Negative control

Preliminary Evaluation of Diagnostic Tests Using Horses Experimentally Infected with *Trypanosoma evansi*

U WERNERY ^a, R ZACHARIAH ^a, J.A MUMFORD ^b, T LUCKINS ^c

Seven surra negative horses were intravenously inoculated with $3 \times 10^{(6)}$ *Trypanosoma evansi* parasites derived from a camel. The microhaematocrit centrifugation test (MHCT) was the most sensitive, first detecting parasites between one and three days (x 2.4) post infection (p.i.). The antigen (ag)-ELISA detected antigen between three and ten days (x 6.6) p.i. The latex agglutination test (LAT) first gave positive results on day 3 (x 3.0) p.i. Following the treatment of horses with trypanocidal drugs, the MCHT and the mouse inoculation test (MIT) became negative.

Three antibody detection techniques, ab-ELISA, card agglutination test (CAT), and immunofluorescent antibody test (IFAT) detected antibodies in the blood of all seven infected horses but not in the uninfected control.

Comparison of serological tests for equine trypanosomosis in naturally infected horses from Kazakhstan

In this study, we compared the complement fixation test (CFT), the horse complement fixation test (HCFT) and a card agglutination test for trypanosomosis (CATT/*T. evansi*) for the diagnosis of equine trypanosomosis in the Republic of Kazakhstan.

CFT has a sensitivity of 57.2% (CI 31.5--79.5%) and a specificity of 95.8% (CI 89.2--98.5%), HCFT has a sensitivity of 80.6% (CI 44.1--95.6%) and a specificity of 99.5% (CI 90.7--100%), **CATT has a sensitivity of 80.2% (CI 44.5--95.2%) and a specificity of 98.5% (CI 79.5--99.9%)**. The seroprevalence of equine trypanosomosis in Kazakhstan was estimated at 16.4% (CI 9.4--27.0%). The data suggest that for epidemiological studies and the control of equine trypanosomosis serological tests prove useful since they have a high specificity and a satisfactory sensitivity.

Field applicable tests, such as CATT/*T. evansi* may be used to replace laboratory-based tests, such as CFT and HCFT.

Serological diagnosis of *Trypanosoma evansi* (Steel, 1885) in horses using a direct agglutination test

A direct agglutination test is described to diagnose 'Mal de Caderas' caused by *Trypanosoma evansi*. The antigen used was a suspension of trypsin-treated parasites stabilized with formalin. The test was evaluated in horses with both natural and experimental infections.

Test sensitivity and specificity were 94 and 97%, respectively. Treatment of serum with 2-mercaptoethanol before testing permitted the differentiation of IgM and IgG antibodies, and possible differentiation of current infection from past exposure to the parasite. The antigen was stable over a 6-month evaluation period and also showed good reproducibility between different batches.

The direct agglutination test is proposed as another tool for diagnosis of *T. evansi* in horses, both for detecting clinical cases and for seroepidemiological studies.