Brief Communication

Evaluation of Testryp CATT applied to samples of dried blood for the diagnosis of sleeping sickness

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Described is an evaluation of the card agglutination test (Testryp CATT) applied to dried blood collected on filter-paper. The sensitivity of the test was compared for samples of diluted sera, whole blood and dried blood. Sera diluted 1:8 gave similar CATT results to those obtained with dried blood. The false negative rate was 5.8%, and test specificity, 100.0%. Use of CATT with samples of dried blood is recommended for screening populations at risk for trypanosomiasis in situations where specialized surveillance teams are not available to test sera or whole blood.

The card agglutination test (Testryp CATT) was developed for the serological diagnosis of Gambian sleeping sickness (1). In view of its simplicity and efficiency, the test has been recommended as the technique of choice to screen for the disease in the field (6, 7). The test is generally carried out on samples of fresh whole blood, serum, or plasma. However, if it could be performed on dried blood samples, it would be of value for use in mass surveys, when the examination of samples can be delayed, and in population surveillance, as part of primary health care efforts. The objective of the study was to evaluate CATT as a diagnostic technique for sleeping sickness based on the use of dried blood samples.

Materials and methods

Blood and serum samples

Samples of whole blood, collected by finger-prick in a capillary tube and on Whatman No. 1 filter-paper, and of sera were taken from 52 Congolese patients with parasitologically confirmed Gambian trypanosomiasis (group A) and from 118 trypanosomiasis-free Congolese (group B) who lived in the same area as the patients in group A (Bouenza focus). The samples of blood collected on the filter-paper were dried and stored in a dessicator.

CATT test systems

Whole blood was analysed by Testryp CATT according to the method described by Magnus et al. (1). In the quantitative test on serum, sera were titrated at twofold dilutions from 1:2 to 1:16.

When CATT was used with dried blood, two filter-paper discs of dried blood (5-mm diameter) were stacked on each test area of the card and covered with a drop (35 μl) of phosphate-buffered saline (PBS). A contact time of 1 hour was allowed to enable the dried serum to be eluted by the PBS. A drop (about 45 μl) of CATT reagent was then added to each test area of the card without removing the filter-paper discs. The resulting solution was mixed and spread, and the card was placed for 5 minutes on a rotator. Agglutination patterns of 1+ or more were taken to be positive.

Results and discussion

The CATT results for samples of diluted sera, whole blood, and dried blood from the individuals in group A are shown in Table 1. All gave positive CATT results when serum (1:4 dilution) and whole blood were tested. Only three patients (5.8%) exhibited negative results with dried blood; also, serum from these patients was negative when tested at a 1:8 dilution. The specificity of the Testryp CATT for dried blood was 100%, as indicated by the results with the 118 trypanosomiasis-free subjects (group B). The high diagnostic value of the test could enable dried blood samples to be used for the surveillance of populations if itinerant diagnostic teams are not available. The blood samples could be taken by non-specialized health care workers, either at a permanent site or during a vaccination round. Analysis

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Table 1: Testryp CATT results for samples of diluted sera, whole blood, and dried blood from 52 Congolese patients with parasitologically confirmed trypanosomiasis

<table>
<thead>
<tr>
<th>Serum (dilution)</th>
<th>No. of samples</th>
<th>No. positive</th>
<th>No. negative</th>
</tr>
</thead>
<tbody>
<tr>
<td>1:2</td>
<td>52</td>
<td>52 (100)*</td>
<td>0 (0)</td>
</tr>
<tr>
<td>1:4</td>
<td>52</td>
<td>52 (100)</td>
<td>0 (0)</td>
</tr>
<tr>
<td>1:8</td>
<td>52</td>
<td>49 (94.2)</td>
<td>3 (5.8)</td>
</tr>
<tr>
<td>1:16</td>
<td>52</td>
<td>39 (63.5)</td>
<td>19 (36.5)</td>
</tr>
<tr>
<td>Whole blood</td>
<td>52</td>
<td>52 (100)</td>
<td>0 (0)</td>
</tr>
<tr>
<td>Dried blood</td>
<td>52</td>
<td>49 (94.2)</td>
<td>3 (6.8)</td>
</tr>
</tbody>
</table>

* Figures in parentheses are percentages.

of the data obtained could therefore provide epidemiological information about trypanosomiasis and the distribution of at-risk zones. The considerably greater sensitivity of Testryp CATT in this study compared with that found in previous investigations in the Congo (2) can be attributed to the use of a modified Trypanosoma brucei gambiense serotype in the test (T. Vervoort, personal communication). The 5.8% of dried-blood samples that gave false negative reactions arose because of the dilution obtained upon extraction of dried blood from filter-paper (subsequently, this was found to be approximately 1:6). No false positive results were found at this dilution for the trypanosomiasis-free individuals in group B. The CATT requires a dilution greater than 1:4 to permit its use on samples of dried blood (2-4). These findings are consistent also with those reported by Miezan et al. (8).

Acknowledgements
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Résumé
Evaluation du Testryp CATT sur sang sec pour le diagnostic de la trypanosomiasis
Le test d’agglutination sur carte (Testryp CATT) effectué à partir de prélèvements de sang sec sur papier filtre est décrit pour application au dépistage sérologique de la trypanosomiasis. Les sensibilités des tests sur sang sec, sérum dilué et sang total ont été comparées. Les sérum à la dilution 1:8 et les échantillons de sang sec donnaient des réponses identiques avec un taux de faux négatifs égal à 5.8%. La spécificité du CATT sur sang sec était de 100%. Ce test est recommandé comme technique de diagnostic de la maladie du sommeil.

References