SEROLOGICAL TESTS IMPROVEMENT FOR CHAGAS DISEASE DIAGNOSIS

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Introduction. Chagas Disease is a parasitic infection caused by *Trypanosoma cruzi* and is consider endemic of Latin America. In Mexico there are few studies about the national prevalence. During the chronic phase of the disease serological tests are used for diagnosis. In this kind of techniques expensive equipment is required and a total protein extract is used in order to achive good sensitivity, but cross reactions are also increased.

The objective of this work was to standardize Dot-ELISA as a diagnostic test for *T. cruzi* infection, since it's an easy, inexpensive and accessible test. As well as reduce cross reaction with *Leishmania spp* of serological tests using protein fractions.

Methods. A total of 249 sera were tested with Dot-ELISA(1): 96 from chagasic patients and 153 from healthy individuals. 40 blood samples dripped and eluted from filter paper were also tested. Sensitivity, specificity and kappa index were calculated in order to determine a correlation value between this technique and other diagnostic tests.

Different protein fractions were obtained by molecular size exclusion chromatography from a total extract with a Superose 12 column and immunodominant antigens were determined by western blot(2). High molecular weight proteins were used to reduce cross reaction in western blot and Dot-ELISA.

Results. Dot-ELISA obtained 97% sensitivity, 88% specificity and a kappa index of 0.79 compared to western blot and ELISA previously standardized for diagnosis (Figure 1).

High molecular weight proteins (250-124 kDa) reduce false positive results when used in serological tests (Figure 2 and Figure 3).

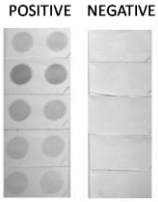

Fig.1 Dot-ELISA evaluation of serum samples from infected patients (positive) and healthy individuals (negative).

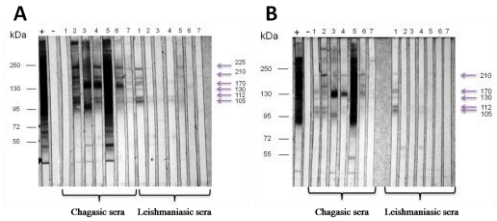

Fig. 2. Western blot diagnostic test of chagasic patients sera using A) Total protein extract and B) High molecular weight proteins fraction, compared to leishmaniasic patients sera.

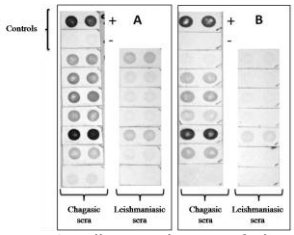

Fig. 3. Dot-ELISA diagnostic test of chagasic and leishmaniasic patients sera using high molecular weight proteins fraction tested with different sample dilution. A) 1:750, B) 1:1000.

Conclusions. Dot-ELISA presented a good correlation with Western blot and ELISA, already standardized for diagnosis of Chagas Disease. Because it is an easy, inexpensive test, it may be useful in field studies.

By reducing the number of proteins used as antigens in serological tests we could increase specificity, maintaining high levels of sensitivity.

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