

## SCHIZODEME ANALYSIS WITH THE RESTRICTION ENDONUCLEASE *RSA I* DIFFERENTIATES BETWEEN *TRYPANOSOMA RANGELI* AND *TRYPANOSOMA CRUZI*

ANTONIO M. GONÇALVES; NÉDIA S. NEHME; NANCY SARAIVA\*; IRIS SEGURA\* &  
CARLOS M. MOREL

Instituto Oswaldo Cruz, Departamento de Bioquímica e Biologia Molecular, Caixa Postal 926, 20001  
Rio de Janeiro, RJ, Brasil \*Centro Internacional de Entrenamiento e Investigaciones Medicas (CIDEIM),  
Avenida 1a. Norte No. 3-03, Apartado Aereo 5390, Cali, Colombia

The differentiation between the two American trypanosomes *Trypanosoma cruzi* and *T. rangeli* is an important issue in the geographical areas where they coexist. For this purpose serology, morphology and behaviour in various biological systems need to be assessed (A. D'Alessandro, 1976, p. 328-393 in W. H. R. Lumsden & D. A. Evans (eds) *Biology of the Kinetoplastida*, Academic Press, London). Recent approaches have used monoclonal antibodies (L. Hudson et al., 1987, *Acta Tropica*, 44: 387-394) and complement lysis, lectin agglutination and isoenzyme profiles (M. Steindel et al., 1991, *Mem. Inst. Oswaldo Cruz*, 86: 73-79).

While characterizing several isolates of these parasites from Colombia and Brazil by schizodeme analysis (C. M. Morel et al., 1980, *Proc. Nat Acad. Sci. USA*, 77: 6810-6814), we found that they could be easily distinguished using the endonuclease *Rsa I*. The figure shows that

digestion of the kinetoplast DNA minicircles from *T. cruzi* with this enzyme originates a major band of around 350 base pairs (bp) (arrow). This is consistent with the restriction site for *Rsa I* (GT/AC) being located in the *minirepeat*, a constant sequence of 120 bp repeated four times along the 1400 bp minicircle molecule of *T. cruzi* (W. Degraeve et al., 1988, *Mol. Biochem. Parasitol.*, 27: 63-70). By contrast, a similar treatment of *T. rangeli* kinetoplast DNA originates a more complex restriction fingerprint with fragments distributed over a wide range of molecular weight.

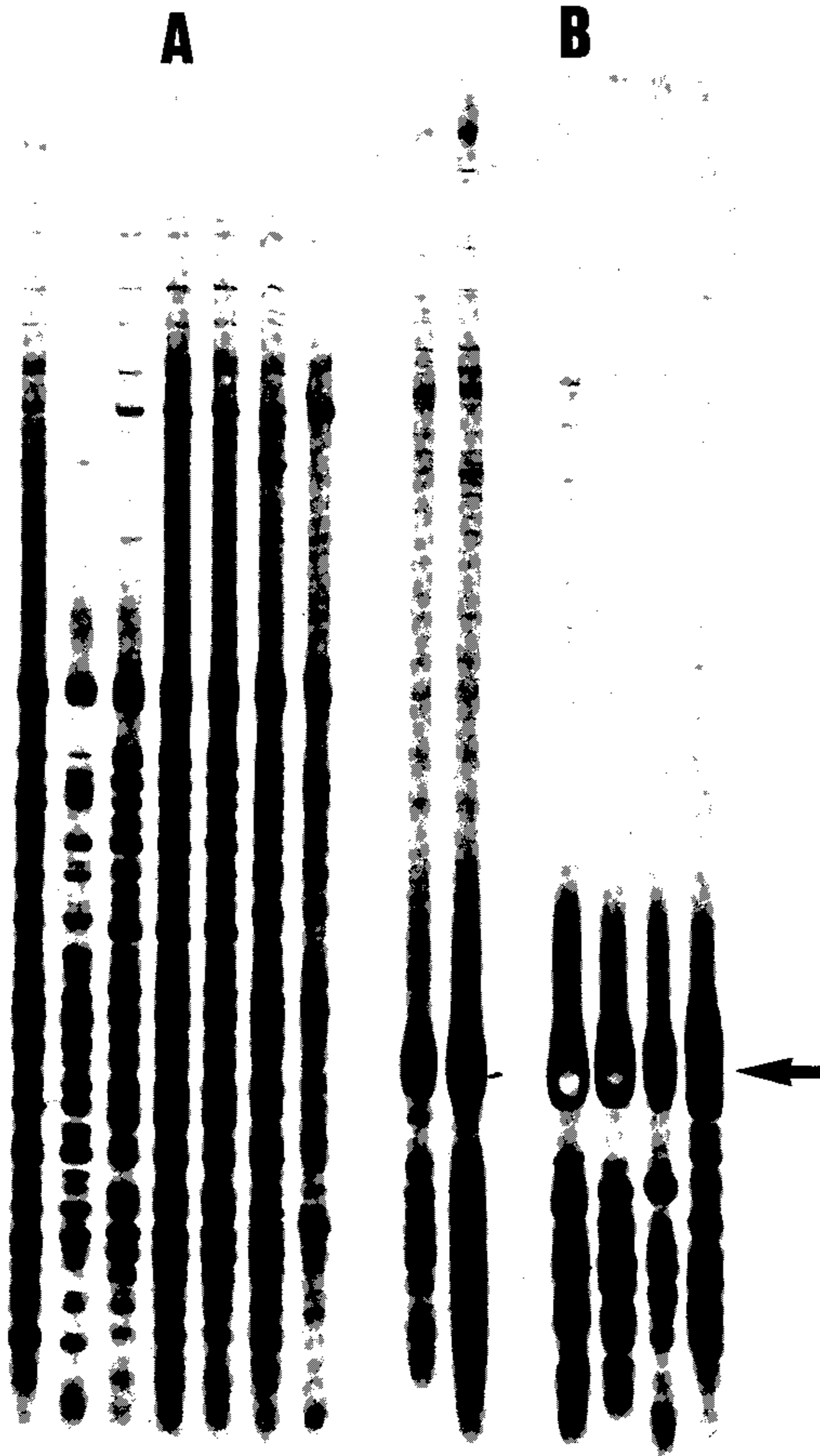
DNA sequence analysis of kinetoplast DNA minicircles from *T. rangeli* is being carried out in our laboratories to investigate their structure and determine the distribution of *Rsa I* sites. The present results, however, already indicate that *Rsa I* schizodeme analysis can be a simple and reliable method for the differentiation of this parasite from *T. cruzi*.

---

This work received financial support from the UNDP/World Bank/WHO Special Programme for Research and Training in Tropical Diseases (TDR), CNPq and COLCIENCIAS.

Received 25 July 1991.

Accepted 4 September 1991.



*Rsa* I schizodeme analysis of isolates of *Trypanosoma rangeli* and *T. cruzi*. Kinetoplast DNA samples were prepared and processed as described by C. M. Morel et al. (1980, *Proc. Nat. Acad. Sci. USA*, 77: 6810-6814) and A. M. Gonçalves et al. (1984, p. 95-109 In C. M. Morel *Genes and Antigens of Parasites*, Oswaldo Cruz Foundation, Rio de Janeiro) using *Rsa* I from New England Biolabs. The restriction fragments were analyzed on a 6-10% polyacrylamide silver-stained gel according to A. M. Gonçalves et al. (1990, *Mem. Inst. Oswaldo Cruz*, 85: 101-106). A: *T. rangeli* (seven isolates from Colombia, two from human cases, five from the insect host *Rhodnius prolixus*). B: *T. cruzi* (three isolates from Brazil – two from human cases, one from a triatomine; three isolates from Colombia – one from a human case and two from *Didelphis marsupialis*). The arrow indicates the fragment of MW = 350 bp (1/4 th the MW of a minicircle).