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ics PROSICS Programa de Salut Internacional de l'ICS

Instituto de Salud Carlos III

Vall d'Hebron Hospital

Molecular characterization of *Trypanosoma cruzi* in five Chagas disease patients who acquired the infection by different routes of transmission

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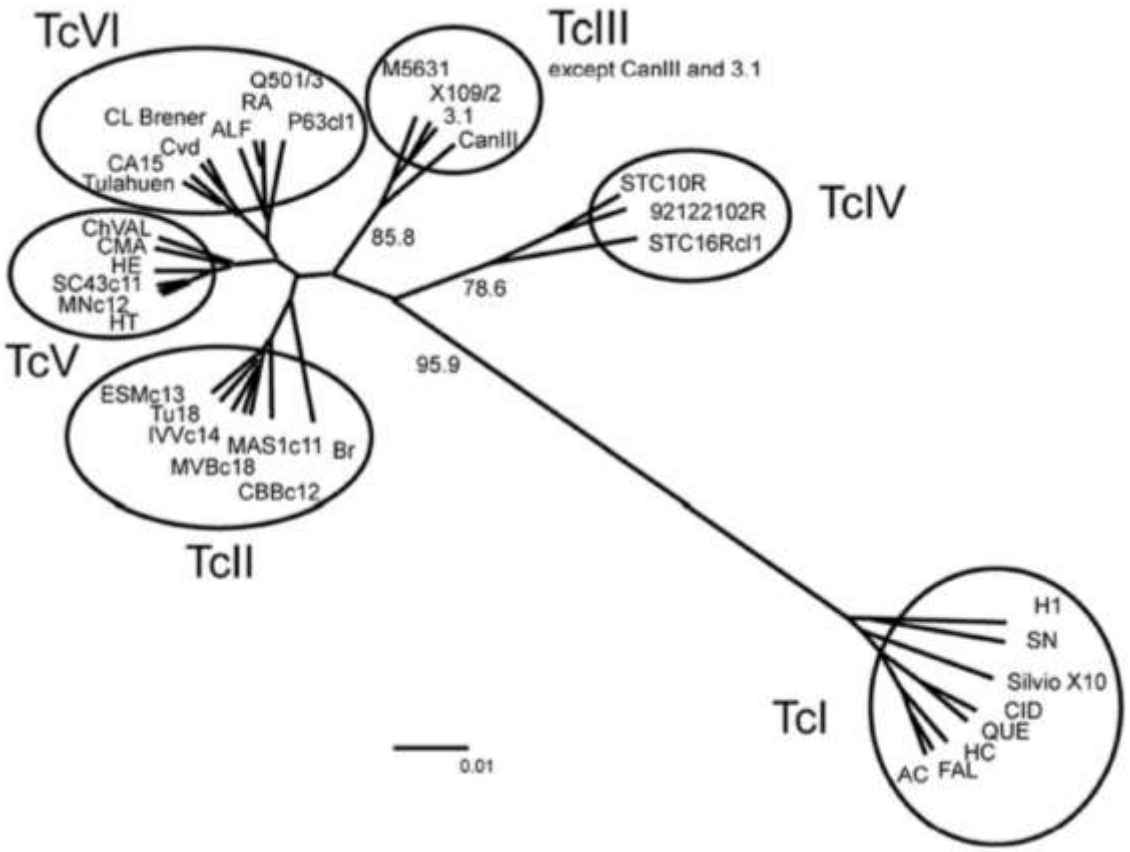
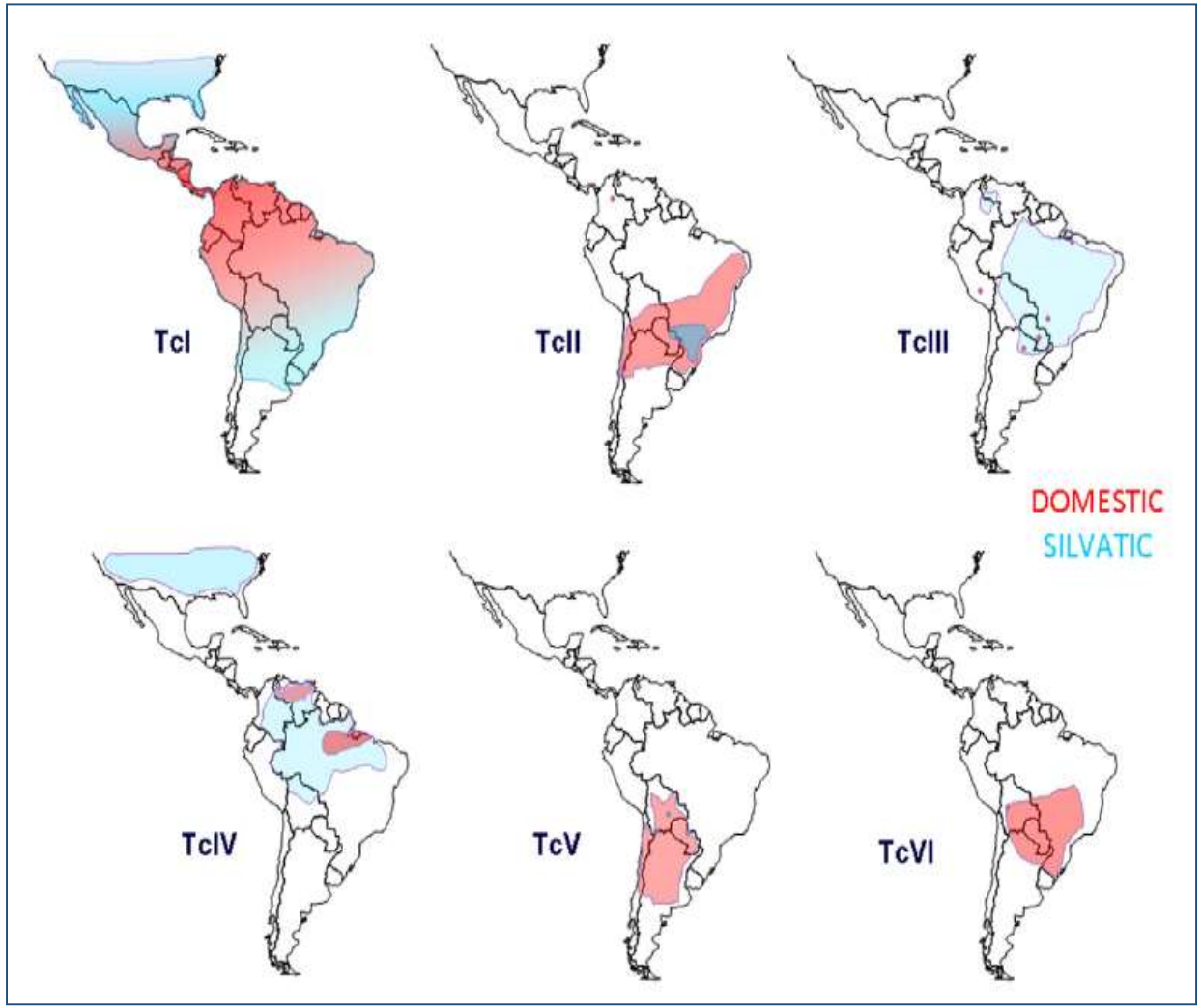
The authors have no conflicts of interest to disclose

Discrete Typing Unit (DTU). Concept and Distribution



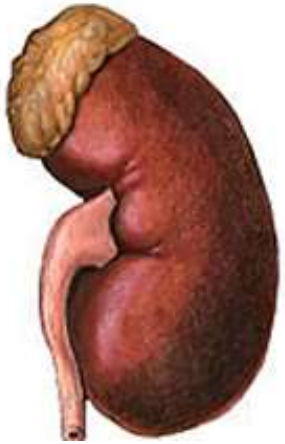
Burgos JM, et al. (2013) PLOS ONE 8 (2013) 3



B. Zingales et al. / Infection, Genetics and Evolution 12 (2012) 240–253

Main Routes of Transmission

VECTORIAL (endemic countries)



ORGAN TRANSPLANTATION

BLOOD TRANSFUSION

VERTICAL TRANSMISION

OBJECTIVE

The aim of this study was to determine the *T.cruzi* DTU of 5 patients who acquired the infection by different routes of transmission

- Male, 103 days
- Bolivian mother
- Asymptomatic

Case 1

- Male, 20 days
- Bolivian mother
- Fever and cholestasis on the newborn

Case 2

- Male, 28 years
- Platelet transfusion Spanish recipient
- Brazilian donor
- Fever, multiple organ failure

Case 3

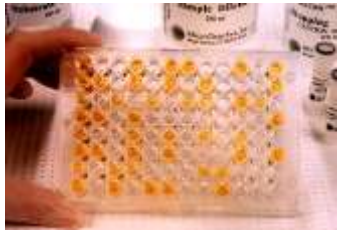
- Male, 37 years
- Bolivia
- VIH co-infection, meningoencephalitis

Case 4

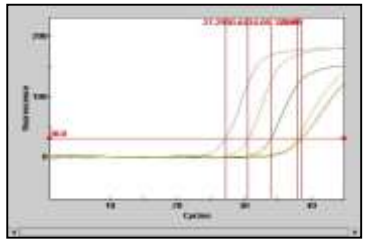
- Male, 58 years
- Bolivia
- Kidney transplant 2014
- Focal neurologic symptoms, brain chagoma

Case 5

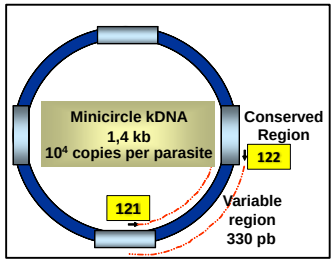
DIAGNOSTIC PROCEDURES



2 Serological tests. ELISA:
✓1 based on recombinant antigens
✓1 based on native antigens



qPCR amplifying a fragment of 166 bp of satellite DNA



Conventional PCR amplifying a fragment of 330 bp of kinetoplastid DNA

ALL SAMPLES SHOWED A HIGH PARASITE LOAD

SAMPLES AND EXTRACTION METHODS FOR GENOTYPING

CLINICAL SAMPLES

Guanidine-preserved Blood (case 2)

DNA/RNA extractor NucliSENS® easyMAG (Biomérieux)

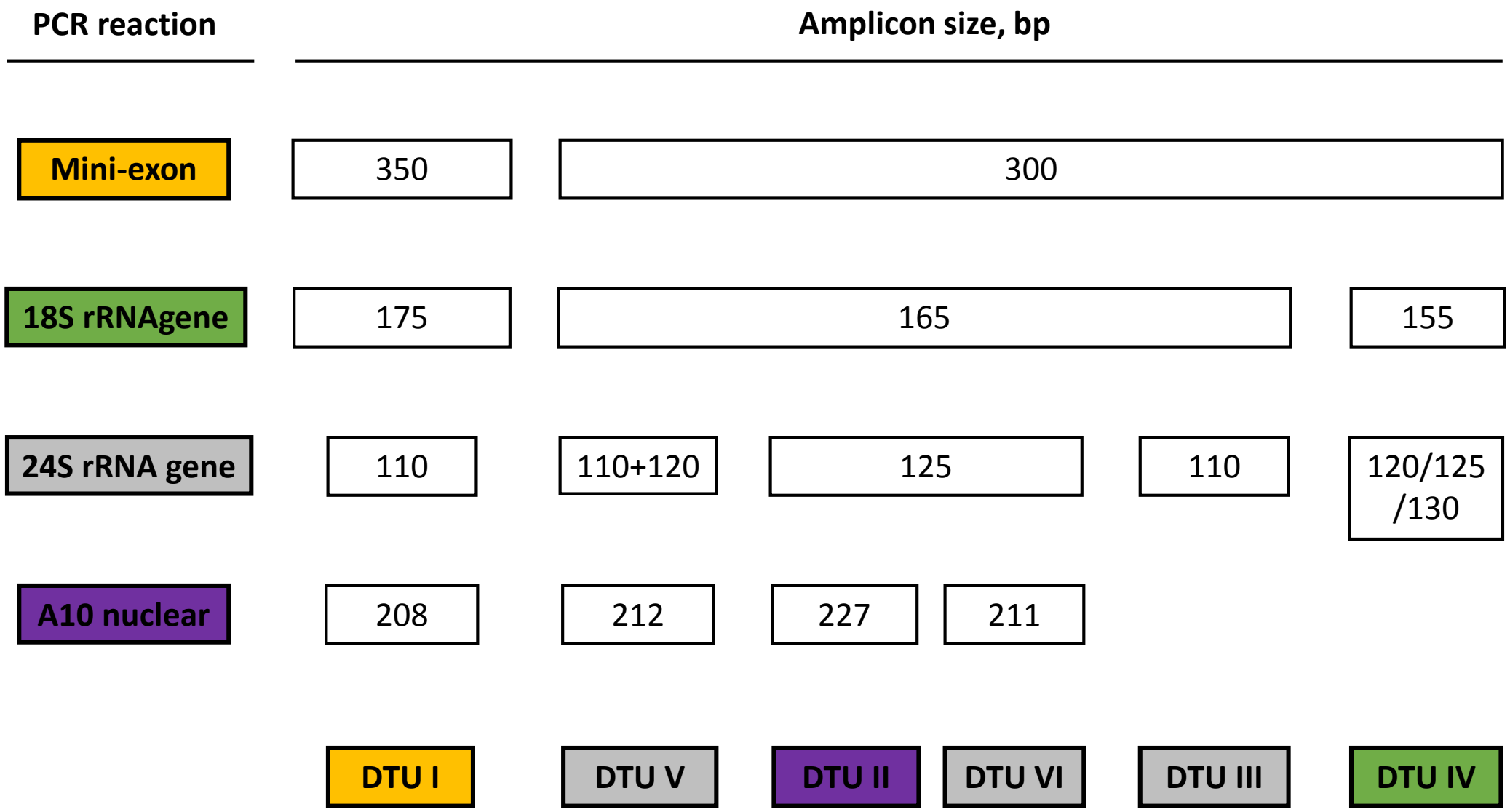
Brain biopsy -Parafin-fixed Tissue (case 5)

DNA Extraction QIAsymphony® SP DNA/RNA Purification (QIAGEN)

Isolated parasites from blood by culture in NNN/LIT medium (Cases 1, 3 and 4)

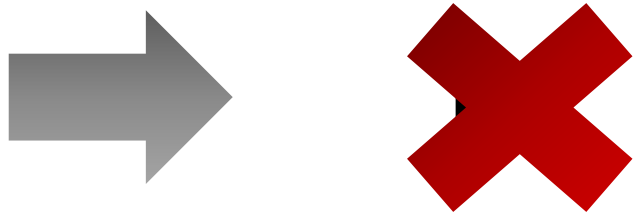
Chelex® 100 Resin (CNM protocol)
High Pure PCR Template Preparation Kit (Roche)

Algorithm for molecular characterization of *T. cruzi* DTUs based on four molecular markers

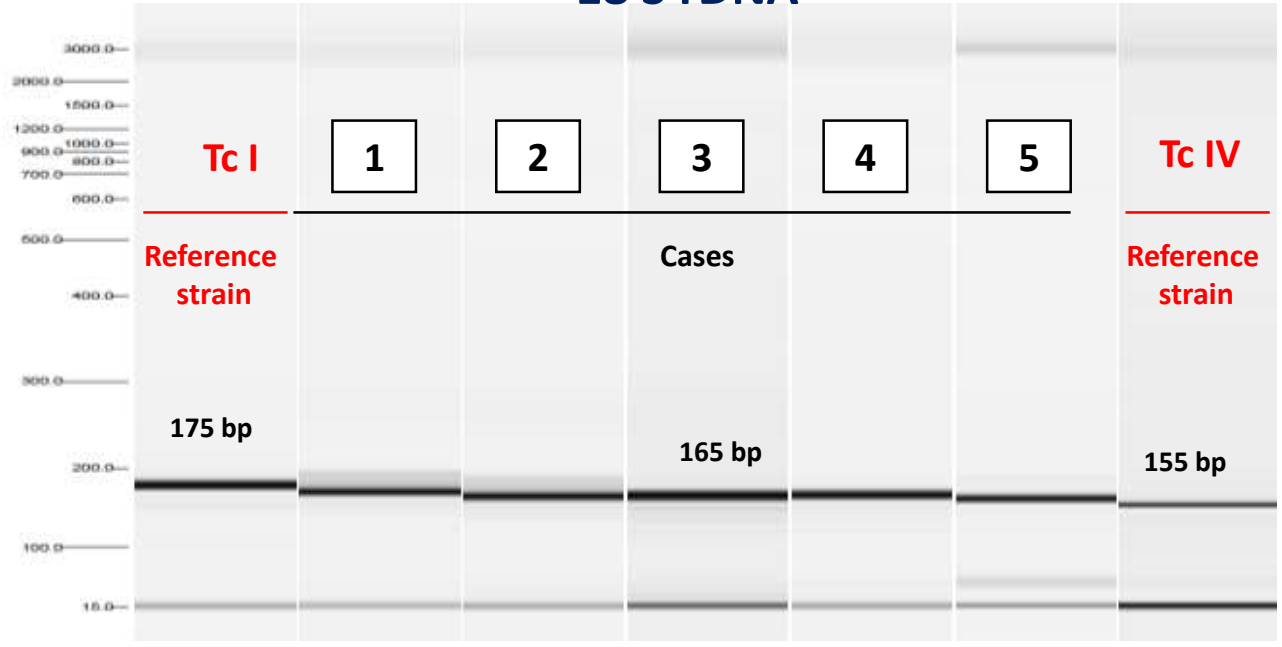


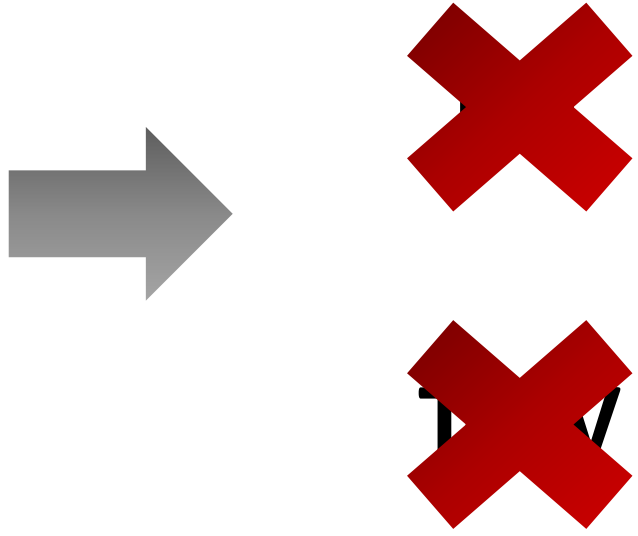
Analysis: Capillary electrophoresis (Qiagen QIAxcel and QIAxcel BioCalculator software version 3.0)

MINI-EXON

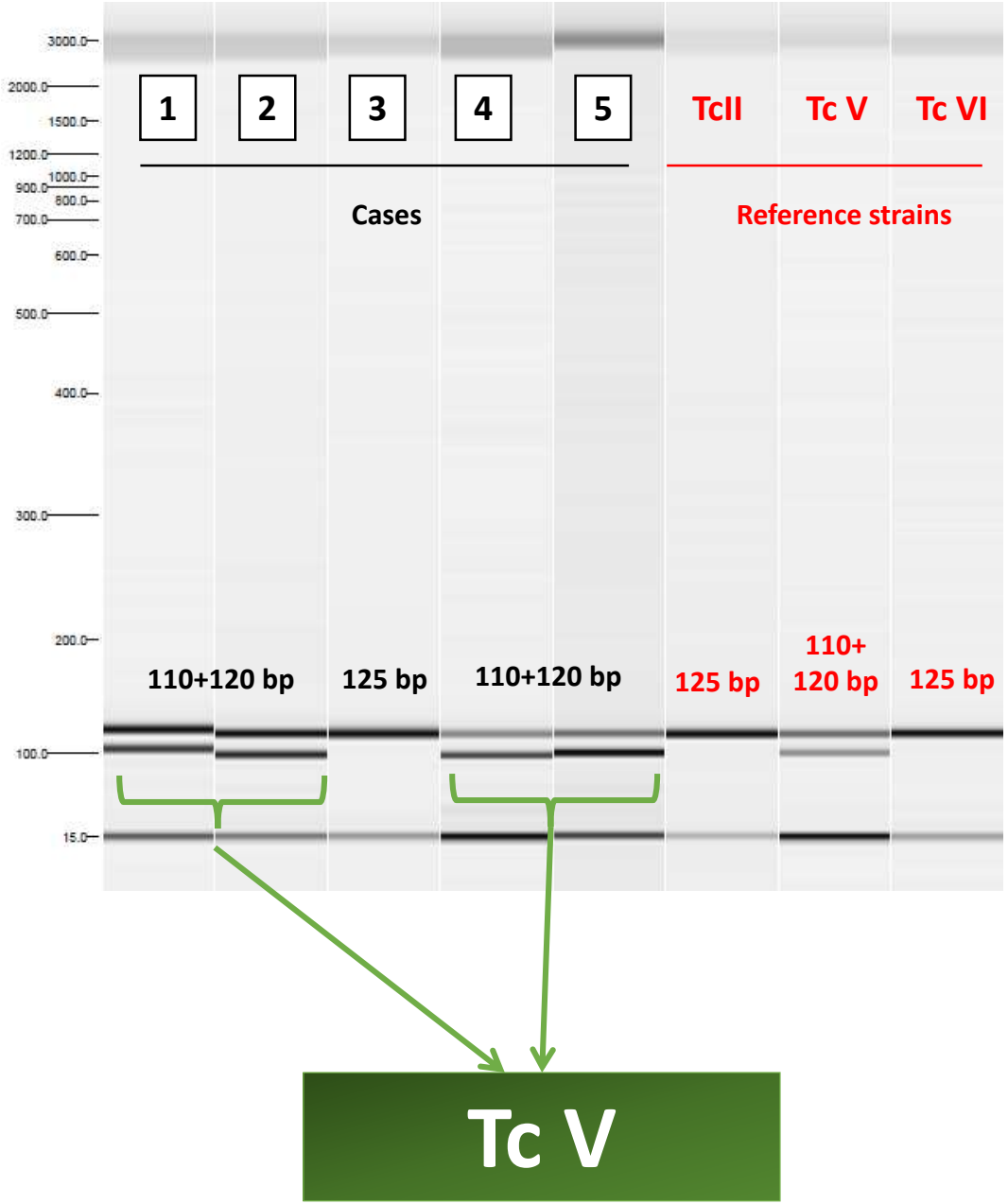


18 S rDNA

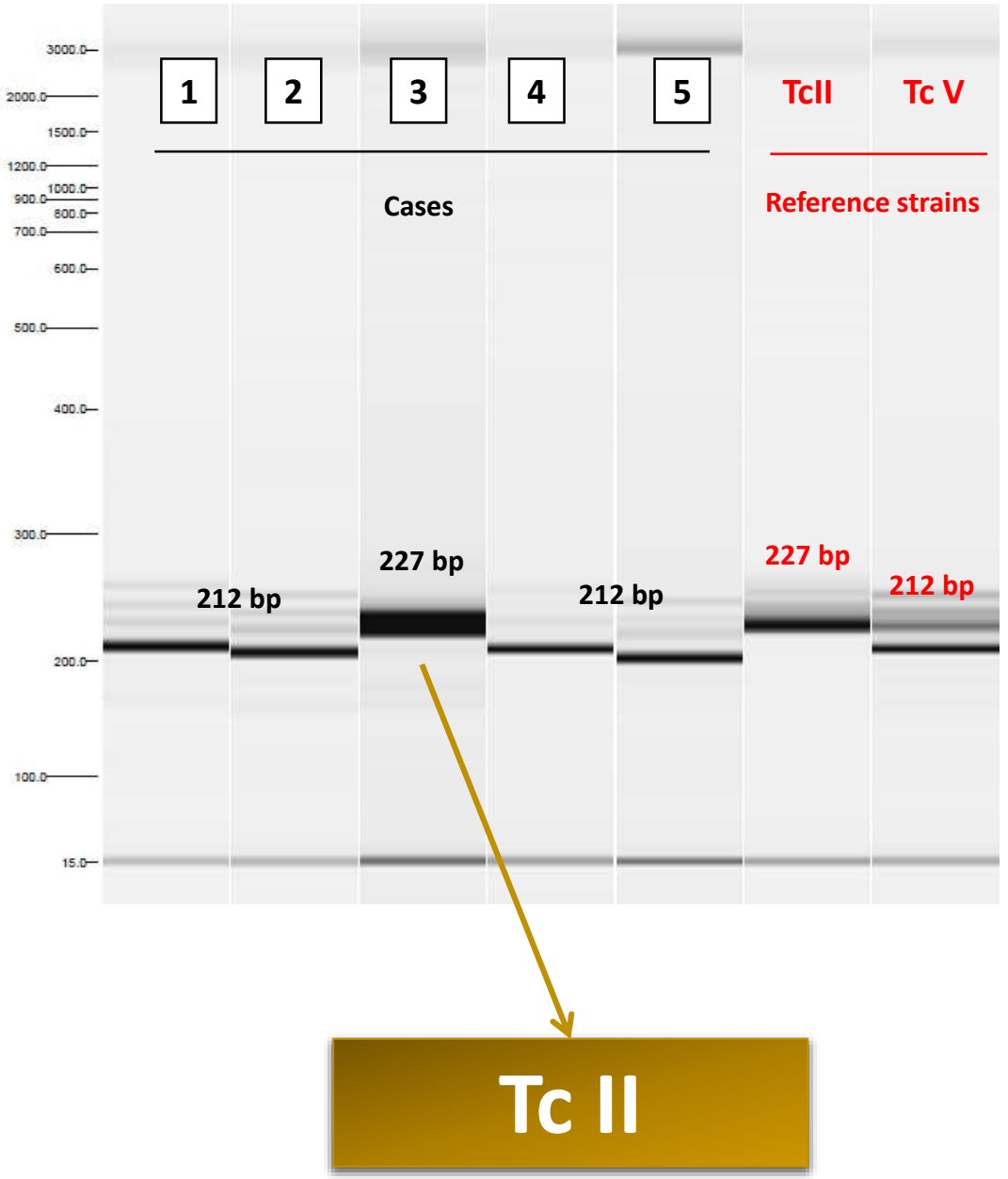




24 S rDNA (D71-D72 primers)



A10 nuclear fragment



CONCLUSIONS

- ✓Molecular characterization can be directly performed in those **clinical samples with high parasite load.**
- ✓**Tc V** was identified in all cases with a common Bolivian origin as source of infection.
- ✓ **TcII** was characterized in the recipient who received platelet transfusion from a Brazilian donor.
- ✓DTUs distribution in migrant population seems to be similar to that observed in the patients' countries of origin.

Thank you for your attention!