A new consensus for *Trypanosoma cruzi* intraspecific nomenclature: second revision meeting recommends TcI to TcVI

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In an effort to unify the nomenclature of Trypanosoma cruzi, *the causative agent of Chagas disease, an updated system was agreed upon at the Second Satellite Meeting. A consensus was reached that* T. cruzi *strains should be referred to by six discrete typing units (*T. cruzi *I-VI). The goal of a unified nomenclature is to improve communication within the scientific community involved in* T. cruzi *research. The justification and implications will be presented in a subsequent detailed report.*

Key words: Chagas disease - *Trypanosoma cruzi* strains - taxonomy - discrete typing units

The biological, biochemical and genetic diversity of *Trypanosoma cruzi* strains has long been recognised, along with their eco-epidemiological complexity, which has been reviewed extensively elsewhere (Macedo & Pena 1998, Campbell et al. 2004, Miles et al. 2009). Over the years, numerous approaches have been used to characterise the population structure of *T. cruzi*, aiming at defining the number of relevant subgroups. Accordingly, these subgroups received different designations, including zymodemes (Miles et al. 1977, 1978, 1981, Romanha et al. 1979), schizodemes (Morel et al. 1980), biodemes (Andrade 1974, Andrade & Magalhães 1997), clonets (Tibayrenc & Ayala 1991), lineages (Souto et al. 1996), clades (Kawashita et al. 2001) and, more recently, discrete typing units (DTUs) (Tibayrenc 1998) and haplotypes (Freitas et al. 2006, Herrera et al. 2007).

In a Satellite Meeting held at Fiocruz in 1999, an expert committee reviewed the available knowledge that indicated a convergence toward clustering *T. cruzi* strains into two major groups. Recommendations were issued that can be summarised as follows (Anonymous 1999). *T. cruzi* strains characterised by biological and biochemical features (e.g., biodemes and zymodemes) and molecular techniques [e.g., multilocus enzyme electrophoresis (MLEE), random amplification of polymorphic DNA (RAPD), mini-exon and 24Sα ribosomal DNA sequences] should be classified into two principal groups, named *T. cruzi* I and *T. cruzi* II. The classification of apparent hybrid strains and strains equivalent to Zymodeme 3 (Miles et al. 1978, 1981) and Biodeme Type I (Andrade 1974) would be decided later after further studies.

In the 10 years that followed the meeting at Fiocruz, the scientific community has advanced in the knowledge of *T. cruzi* diversity. Multilocus genotyping has revealed six distinct DTUs, which partition into two major subdivisions, termed DTU I and DTU II. DTUs are defined as ''sets of stocks that are genetically more related to each other than to any other stock and that are identifiable by common genetic, molecular or immunological markers'' (Tibayrenc 1998). *T. cruzi* DTU II was further split into five DTUs, IIa-e (Brisse et al. 2000, 2001), based on congruent phylogenetic information from MLEE and RAPD markers. DTUs I and IIb correspond, respectively, to the *T. cruzi* I and *T. cruzi* II groups recommended by the original expert committee in 1999 (Table I). Current studies indicate that four subdivisions have emerged within DTU I as well (Herrera et al. 2007, Falla et al. 2009), although these have not been integrated into the nomenclature revision.

Although the major genetic variability of *T. cruzi* was proposed initially to have resulted from predominant clonal evolution (Tibayrenc et al. 1986), increasing evidence indicates that genetic exchange between parasites has contributed to the present popu-

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TABLE I

2009 nomenclature for *Trypanosoma cruzi* divisions

DTU designation	Abbreviation	Equivalence to former *T. cruzi* grouping schemes
T. cruzi I	TcI	*T. cruzi* I[a, b] and DTU I[c]
T. cruzi II	TcII	*T. cruzi* II[a] and DTU IIb[c]
T. cruzi III	TcIII	Z3/Z1 ASAT[d], Z3-A[e], DTU IIc[c] and *T. cruzi* III[f]
T. cruzi IV	TcIV	Z3[d], Z3-B[e] and DTU IIa[c]
T. cruzi V	TcV	Bolivian Z2[d], rDNA 1/2[g], clonet 39[h] and DTU IId[c]
T. cruzi VI	TcVI	Paraguayan Z2[i], Zymodeme B[j] and DTU IIe[c]

a: Anonymous 1999; *b*: Falla et al. 2009; *c*: Brisse et al. 2000; *d*: Miles et al. 1981; DTU: discrete typing units; *e*: Mendonça et al. 2002; *f*:Freitas et al. 2006; *g*: Souto et al. 1996; *h*: Tibayrenc and Ayala 1991; *i*: Chapman et al. 1984 ; *j*: Carneiro et al. 1990.

lation structure (Sturm & Campbell 2009). This was first documented by the existence of hybrid organisms in sylvatic *T. cruzi* populations and sympatric clinical strains and, later, experimentally (Gaunt et al. 2003 and cited references). The prevailing view is that DTU I and DTU IIb are ancient lineages and that DTU IId and DTU IIe strains are the products of a minimum of two hybridisation events (Westenberger et al. 2005, Freitas et al. 2006, Tomazi et al. 2009). The evolution of DTU IIa and DTU IIc strains is insufficiently understood for the moment, although these DTUs may also have a hybrid origin (Sturm et al. 2003, Westenberger et al. 2005). Based on microsatellite and mitochondrial DNA analyses, DTU IIc may represent a third ancestral lineage, which was named *T. cruzi* III (Freitas et al. 2006).

The advances in the understanding of *T. cruzi* population structure indicate that it is time to revise the nomenclature of *T. cruzi* strains. The standardisation of nomenclature will facilitate communication among researchers working with *T. cruzi* aimed at characterisation of its eco-epidemiological features, pathogenicity and questions of basic biology.

A Second Satellite Meeting was held in Buzios, Brazil, on August 23, 2009, preceding the XIII International Congress of Protistology, the XXV Annual Meeting of the Brazilian Society of Protozoology and the XXXVI Annual Meeting on Basic Research in Chagas Disease. By consensus, the expert committee recognised that the nomenclature for *T. cruzi* strains should be classified into six DTUs, *T. cruzi* I-VI and issued recommendations accordingly. Detailed justification and implications of these decisions will be presented in a future publication.

Recommendations of the Second Satellite Meeting - (i) The known isolates and strains of *T. cruzi* should be assigned to one of six DTUs (*T. cruzi* I-VI). Additional variants may arise in the future; (ii) DTUs *T. cruzi* I and *T. cruzi* II correspond to the two groups originally defined in the First Satellite Meeting (Anonymous 1999). A notable exception is the CL Brener strain, classified at that time as *T. cruzi* II and now reclassified as *T. cruzi* VI; (iii) The designation of the six DTUs, their abbreviations and equivalence are summarised in Table I; (iv) Authors and reviewers of manuscripts describing studies on *T. cruzi* are encouraged to use the new nomenclature; (v) Editors of sci-

TABLE II

Representative strains and corresponding discrete typing units (DTUs)

Strain[a]	DTUs	Country	Host/vector
12 SF	*T. cruzi* II	Bahia, Brazil	*Homo sapiens*
21 SF	*T. cruzi* II	Bahia, Brazil	*Homo sapiens*
3663	*T. cruzi* III	Amazonas, Brazil	*Panstrongylus geniculatus*
3869	*T. cruzi* III	Amazonas, Brazil	*Homo sapiens*
4167	*T. cruzi* IV	Amazonas, Brazil	*Rhodnius brethesi*
4182	*T. cruzi* III	Amazonas, Brazil	*Rhodnius brethesi*
92.80 cl2	*T. cruzi* V	Santa Cruz, Bolivia	*Homo sapiens*
92101601P cl1	*T. cruzi* I	Georgia, USA	*Didelphis marsupialis*
92122102R	*T. cruzi* IV	Georgia, USA	*Procyon lotor*
Bug2148 cl1	*T. cruzi* V	Rio Grande do Sul, Brazil	*Triatoma infestans*
Bug2149 cl10	*T. cruzi* V	Rio Grande do Sul, Brazil	*Triatoma infestans*

↳

Strain[a]	DTUs	Country	Host/vector
CA-1	*T. cruzi* I	Argentina	*Homo sapiens*
CanIII cl1	*T. cruzi* IV	Pará, Brazil	*Homo sapiens*
CL	*T. cruzi* VI	Rio Grande do Sul, Brazil	*Triatoma infestans*
CL Brener[b]	*T. cruzi* VI	Rio Grande do Sul, Brazil	*Triatoma infestans*
CM17	*T. cruzi* III	Meta, Colombia	*Dasypus sp.*
Colombiana	*T. cruzi* I	Colombia	*Homo sapiens*
Cuica cl1	*T. cruzi* I	São Paulo, Brazil	*Philander opossum*
Cutia cl1	*T. cruzi* I	Espírito Santo, Brazil	*Dasyprocta aguti*
Davis 9.90	*T. cruzi* I	Tegucigalpa, Honduras	*Triatoma dimidiata*
Dm28c[c]	*T. cruzi* I	Carabobo, Venezuela	*Didelphis marsupialis*
Dm7	*T. cruzi* I	Casanare, Colombia	*Didelphis marsupialis*
Dog Theis[d]	*T. cruzi* IV	Oklahoma, USA	*Canis familiaris*
Esmeraldo cl3	*T. cruzi* II	Bahia, Brazil	*Homo sapiens*
G	*T. cruzi* I	Amazonas, Brazil	Opossum
Gambá cl1	*T. cruzi* I	São Paulo, Brazil	*Didelphis azarae*
IVV cl4	*T. cruzi* II	Cuncumen, Chile	*Homo sapiens*
JEM C	*T. cruzi* I	Boyacá, Colombia	*Homo sapiens*
José	*T. cruzi* I	Paraíba, Brazil	*Homo sapiens*
K-98[e]	*T. cruzi* I	Argentina	*Homo sapiens*
M5631 cl5	*T. cruzi* III	Pará, Brazil	*Dasypus novemcinctus*
M6241 cl6	*T. cruzi* III	Pará, Brazil	*Homo sapiens*
MAS cl1	*T. cruzi* II	Minas Gerais, Brazil	*Homo sapiens*
MN cl2	*T. cruzi* V	Region IV, Chile	*Homo sapiens*
NR cl3	*T. cruzi* V	Salvador, Chile	*Homo sapiens*
P63 cl1	*T. cruzi* VI	Makthlawaiya, Paraguay	*Triatoma infestans*
PALC	*T. cruzi* I	Casanare, Colombia	*Rhodnius prolixus*
Peruvian	*T. cruzi* II	Peru	*Homo sapiens*
RA	*T.cruzi* VI	Argentina	*Homo sapiens*
Sc43 cl1	*T. cruzi* V	Santa Cruz, Bolívia	*Triatoma infestans*
SO3 cl5	*T. cruzi* V	Potosi, Bolivia	*Triatoma infestans*
Sylvio[f] X10 cl1	*T. cruzi* I	Pará, Brazil	*Homo sapiens*
Td11C	*T. cruzi* I	Boyacá Colombia	*Triatoma dimidiata*
Tu18 cl1	*T. cruzi* II	Tupiza, Bolívia	*Triatoma infestans*
Tulahuen	*T. cruzi* VI	Tulahuen, Chile	*Homo sapiens*
Tulahuen cl2	*T. cruzi* VI	Tulahuen, Chile	*Homo sapiens*
X10/1[g]	*T. cruzi* I	Pará, Brazil	*Homo sapiens*
X109/2	*T. cruzi* III	Makthlawaiya, Paraguay	*Canis familiaris*
Y	*T. cruzi* II	São Paulo, Brazil	*Homo sapiens*
YuYu	*T. cruzi* I	Minas Gerais, Brazil	*Triatoma infestans*

a: the term cl following the name of the strain indicates it is a clone derived from the original isolate; *b*: CL Brener is a clone derived from the CL strain; *c*: Dm28c is a clone derived from the Dm28 strain; *d*: Dog Theis sometimes shortened to DogT or DogTh; *e*: K-98 is a clone derived from the CA-1 strain; *f*: Sylvio referred to as Silvio; *g*: same as Sylvio (Silvio) X10 cl1.

entific journals are encouraged to adopt the recommended nomenclature for the *T. cruzi* DTUs in their journals; (vi) Typing services should be made available for strains in the existing literature. A list of some reference and "experimental" strains and their corresponding designation is presented to serve as guide for researchers (Table II).

To obtain the greatest effectiveness, the new recommendations for the naming of *T. cruzi* will require a simple and reproducible schema for typing isolates into their respective DTUs. Lewis et al. (2009) described such a schema using currently available markers in the form of a triple assay that employed rDNA PCR (Souto et al. 1996) and PCR-RFLP of the HSP60 and GPI loci (Westenberger et al. 2005). The expert committee is exploring the possibility of a multicentric study to standardise and validate different protocols for genotyping reference and laboratory strains, as well as field isolates. Any multicentric study will make a call for comparative typing protocols that are under development currently in other laboratories.

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