



## **Trypanosoma cruzi Ikiakarora (TcIII) Draft Genome Sequence**

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**ABSTRACT** Trypanosoma cruzi shows a genetic diversity that has been associated with the variability of clinical manifestations, geographical distribution, and preferential parasite-vector interactions. In an effort to better understand this genetic variability, here, the draft genome of T. cruzi strain Ikiakarora (discrete typing unit Tclll), which has been associated with the sylvatic cycle, is reported.

*T*rypanosoma cruzi is a protozoan parasite that causes Chagas disease, one of the biggest public health problems in Latin America, with more than 10,000 deaths annually [\(1\)](#page-1-0). Currently, the dynamics of migration of people from countries of endemicity has favored the spreading of Chagas disease to the United States, Canada, Europe, and the western Pacific. [\(2](#page-1-1)[–](#page-1-2)[4\)](#page-1-3). This parasite presents a high level of genetic variability, being classified into six discrete typing units (DTUs), TcI to TcVI [\(5\)](#page-1-4). TcIII is concentrated in South America and is associated mainly with the sylvatic cycle and the terrestrial niche [\(6\)](#page-1-5). Here, the draft genome of T. cruzi strain Ikiakarora (IRHO/CO/95), which belongs to DTU III as characterized by 24S $\alpha$  rRNA, miniexon, and 18S rRNA markers, is reported [\(7\)](#page-1-6). This strain was isolated in Catatumbo (North Santander, Colombia) from the sylvatic vector Rhodnius prolixus, the second most frequent transmitting vector of Chagas disease [\(8\)](#page-1-7). Analyses of the H2A gene units of different strains isolated in Colombia showed that the genome of strain Ikiakarora has a high degree of plasticity [\(9\)](#page-1-8).

Epimastigote forms were cultivated at 28°C in liver infusion tryptose (LIT) medium supplemented with 10% fetal bovine serum. When the parasites reached log phase (10  $\times$ 10<sup>6</sup> to 20  $\times$  10<sup>6</sup> parasites/ml), they were collected and lysed in 1% NP-40. Nuclei were lysed by addition of 1% SDS, and genomic DNA was purified by phenol-chloroform extraction and ethanol precipitation. Sequencing was carried out using Ion Torrent technology (Thermo Fischer Scientific, Inc.). Library construction, size selection, quality filtering, DNA concentration, and processing were performed as previously described [\(10\)](#page-1-9), obtaining 3,928,712 raw reads (average read length, 254 bp).

Sequence reads were analyzed using FastQC v0.10.1 (default settings) [\(http://www](http://www.bioinformatics.babraham.ac.uk/projects/fastqc) [.bioinformatics.babraham.ac.uk/projects/fastqc\)](http://www.bioinformatics.babraham.ac.uk/projects/fastqc), and Prinseq v0.20.4 [\(11\)](#page-1-10) was used iteratively for quality filtering using the following parameters: -derep 14, -ns\_max\_p 1 -ns\_max\_n 3 -trim\_ns\_left 1 -trim\_ns\_right 1, -trim\_qual\_right 20 -trim\_qual\_type mean -trim\_qual\_window 5 -trim\_qual\_step 1, -trim\_qual\_right 20 -trim\_qual\_type mean -trim\_qual\_window 1 -trim\_qual\_step 1, -trim\_qual\_left 20 -trim\_qual\_type mean -trim\_qual\_window 5 -trim\_qual\_step 1, -trim\_qual\_left 20 -trim\_qual\_type mean -trim\_qual\_window 1 -trim\_qual\_step 1, -lc\_method entropy -lc\_threshold 50, -min\_ qual\_mean 25, and -min\_len 50. The obtained 3,338,764 quality-filtered reads (average read length, 266 bp) were assembled into 11,096 contigs totaling 18,492,845 bp, with an  $N_{50}$  value of 2,193 bp and an average contig size of 1,666 bp using CLC Genomics Workbench software v8.0 (Qiagen) (length fraction, 0.90; similarity fraction, 0.97; **Citation** Gómez I, Rastrojo A, Lorenzo-Díaz F, Sánchez-Luque FJ, Macías F, Aguado B, Valladares B, Requena JM, López MC, Thomas MC. 2020. Trypanosoma cruzi Ikiakarora (TcIII) draft genome sequence. Microbiol Resour Announc 9:e00453-20. [https://doi.org/10.1128/](https://doi.org/10.1128/MRA.00453-20) [MRA.00453-20.](https://doi.org/10.1128/MRA.00453-20)

**Editor** Jason E. Stajich, University of California, Riverside

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**Received** 22 April 2020 **Accepted** 9 June 2020 **Published** 2 July 2020

minimum contig length, 500 bp). The longest contig was 33,607 bp, and the  $G+C$ content was 48.72%. To assess genome assembly completeness, BUSCO (Benchmarking Universal Single-Copy Orthologs) v4.0.5 analysis [\(12\)](#page-1-11) (parameters: -m genome, -l euglenozoa\_odb10) was performed on the assembled genome using the Euglenozoa odb10 orthologue set ( $n = 130$ ). A total of 106 complete BUSCOs (81.6%) and 102 single-copy BUSCOs (78.5%) were identified from 130 BUSCO-searched groups.

**Data availability.** The T. cruzi Ikiakarora assembled genome has been deposited in GenBank (accession number [WWPZ00000000\)](https://www.ncbi.nlm.nih.gov/nuccore/WWPZ00000000), and the raw reads have been deposited in the SRA (accession numbers [SRR11235102,](https://www.ncbi.nlm.nih.gov/sra/SRR11235102) [SRR11235103,](https://www.ncbi.nlm.nih.gov/sra/SRR11235103) and [SRR11235104](https://www.ncbi.nlm.nih.gov/sra/SRR11235104) and BioProject accession number [PRJNA595095\)](https://www.ncbi.nlm.nih.gov/bioproject/PRJNA595095).

## **ACKNOWLEDGMENTS**

We are grateful to Almudena López-Barajas (IPBLN) for her technical collaboration in the identification of the lineage of strain Ikiakarora.

This work was supported by grant SAF2016-80998-R from the Programa Estatal I+D+i (MINECO) and the Network of Tropical Diseases Research RICET (RD16/0027/ 0001, RD16/0027/0005, and RD16/0027/0008) and FEDER.

This work is part of the Ph.D. thesis of student Inmaculada Gómez at the University of Granada in the Biochemistry and Molecular Biology Program.

## <span id="page-1-0"></span>**REFERENCES**

- <span id="page-1-1"></span>1. Anonymous. 2011. Working to overcome the global impact of neglected tropical diseases—summary. Wkly Epidemiol Rec 86:113–120.
- 2. Bern C, Montgomery SP. 2009. An estimate of the burden of Chagas disease in the United States. Clin Infect Dis 49:e52– e54. [https://doi.org/](https://doi.org/10.1086/605091) [10.1086/605091.](https://doi.org/10.1086/605091)
- <span id="page-1-2"></span>3. Gascon J, Bern C, Pinazo MJ. 2010. Chagas disease in Spain, the United States and other non-endemic countries. Acta Trop 115:22–27. [https://](https://doi.org/10.1016/j.actatropica.2009.07.019) [doi.org/10.1016/j.actatropica.2009.07.019.](https://doi.org/10.1016/j.actatropica.2009.07.019)
- <span id="page-1-3"></span>4. Guerri-Guttenberg RA, Grana DR, Ambrosio G, Milei J. 2008. Chagas cardiomyopathy: Europe is not spared! Eur Heart J 29:2587–2591. [https://](https://doi.org/10.1093/eurheartj/ehn424) [doi.org/10.1093/eurheartj/ehn424.](https://doi.org/10.1093/eurheartj/ehn424)
- <span id="page-1-4"></span>5. Zingales B, Andrade SG, Briones MR, Campbell DA, Chiari E, Fernandes O, Guhl F, Lages-Silva E, Macedo AM, Machado CR, Miles MA, Romanha AJ, Sturm NR, Tibayrenc M, Schijman AG, Second Satellite M, Second Satellite Meeting. 2009. A new consensus for Trypanosoma cruzi intraspecific nomenclature: second revision meeting recommends TcI to TcVI. Mem Inst Oswaldo Cruz 104:1051–1054. [https://doi.org/10.1590/s0074](https://doi.org/10.1590/s0074-02762009000700021) [-02762009000700021.](https://doi.org/10.1590/s0074-02762009000700021)
- <span id="page-1-5"></span>6. Zingales B, Miles MA, Campbell DA, Tibayrenc M, Macedo AM, Teixeira MM, Schijman AG, Llewellyn MS, Lages-Silva E, Machado CR, Andrade SG, Sturm NR. 2012. The revised Trypanosoma cruzi subspecific nomenclature: rationale, epidemiological relevance and research applications. Infect Genet Evol 12:240 –253. [https://doi.org/10.1016/j.meegid.2011.12.009.](https://doi.org/10.1016/j.meegid.2011.12.009)
- <span id="page-1-6"></span>7. Brisse S, Verhoef J, Tibayrenc M. 2001. Characterisation of large and small subunit rRNA and mini-exon genes further supports the distinction of six Trypanosoma cruzi lineages. Int J Parasitol 31:1218 –1226. [https://doi](https://doi.org/10.1016/s0020-7519(01)00238-7) [.org/10.1016/s0020-7519\(01\)00238-7.](https://doi.org/10.1016/s0020-7519(01)00238-7)
- <span id="page-1-7"></span>8. Rodriguez P, Montilla M, Nicholls S, Zarante I, Puerta C. 1998. Isoenzymatic characterization of Colombian strains of Trypanosoma cruzi. Mem Inst Oswaldo Cruz 93:739 –740. [https://doi.org/10.1590/s0074](https://doi.org/10.1590/s0074-02761998000600008) [-02761998000600008.](https://doi.org/10.1590/s0074-02761998000600008)
- <span id="page-1-8"></span>9. Thomas MC, Olivares M, Escalante M, Maranon C, Montilla M, Nicholls S, Lopez MC, Puerta C. 2000. Plasticity of the histone H2A genes in a Brazilian and six Colombian strains of Trypanosoma cruzi. Acta Trop 75:203–210. [https://doi.org/10.1016/S0001-706X\(00\)00061-9.](https://doi.org/10.1016/S0001-706X(00)00061-9)
- <span id="page-1-9"></span>10. Gómez I, Rastrojo A, Sanchez-Luque FJ, Lorenzo-Díaz F, Macías F, Valladares B, Aguado B, Requena JM, López MC, Thomas MC. 2020. Draft genome sequence of Trypanosoma cruzi B. M. López strain (TcIa), isolated from a Colombian patient. Microbiol Resour Announc 9:e00031-20. [https://doi.org/10.1128/MRA.00031-20.](https://doi.org/10.1128/MRA.00031-20)
- <span id="page-1-11"></span><span id="page-1-10"></span>11. Schmieder R, Edwards R. 2011. Quality control and preprocessing of metagenomic datasets. Bioinformatics 27:863– 864. [https://doi.org/10](https://doi.org/10.1093/bioinformatics/btr026) [.1093/bioinformatics/btr026.](https://doi.org/10.1093/bioinformatics/btr026)
- 12. Seppey M, Manni M, Zdobnov EM. 2019. BUSCO: assessing genome assembly and annotation completeness. Methods Mol Biol 1962:227–245. [https://](https://doi.org/10.1007/978-1-4939-9173-0_14) [doi.org/10.1007/978-1-4939-9173-0\\_14.](https://doi.org/10.1007/978-1-4939-9173-0_14)