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Correspondence

Genetic polymorphism of 22 autosomal STR markers in Paraguay

Dear Editor,

Autosomal Short Tandem Repeats markers (aSTRs) are widely used for human identification, paternity, crime scene caseworks and for population genetics studies [1–3]. Typically 15 aSTR were included in commercial formats for years, but more recently additional markers have been added to new validated kits gaining valuable information of forensic interest [4,5].

In this letter we report the results of 22 autosomal STR markers included in the Powerplex® Fusion kit in a sample of 548 individuals of admixed population from the most populated geographical regions of Republic of Paraguay.

Information on aSTR markers in Paraguayan population has been scarce; to the best of our knowledge there only exists one scientific publication presenting frequencies of 15 aSTR analyzed in 168 individuals [6].

Paraguay has a population about of 6,500,000 inhabitants according to the population census of 2012 projecting a population about of 6,850,000 inhabitants by 2016 [7]. Most of the population resides in urban areas (61% urban against 39% rural) [7]. Paraguay has two official languages spoken by most of the population: Spanish and Guaraní, according to the strong legacy of Guaraní culture in the country [8].

Paraguay borders Bolivia at North, Brazil at East and Argentina at south. The country is politically distributed in 18 departments. The population distribution along the country is very uneven; ten of the 18 departments account for 80% of the population and are located to the East of the country, where Asunción, the Capital city, is located [7].

To the West of the country there is the lowest population density with less than 3% of the population distributed in 60% of the whole territory (Supplementary Fig. 1) [7]. The admixed population represents more than 95% of general population, having a minor proportion of people who self-identify as Native American ethnic groups representing less than 3% of population [7,8].

Peripheral blood samples from 548 of volunteer donors, 540 males and 8 females, were collected from 10 eastern departments where more than 80% of the Paraguayan population is located. Samples gathered from the different departments were distributed as follows: 218 from Alto Paraná (region 1), 31 from Asunción (region 2), 61 from Central (region 3), 113 de Guairá, Caaguazú and Amambay (region 4), 79 from Itapuá (region 5), 38 from Paraguarí, Misiones and Cordillera (region 6) (Supplementary Fig. 2).

This study followed the ethical principles of the 2000 Helsinki Declaration of the World Medical Association (http://www.uma.net/e/policy/b3.htm) and written informed consent was obtained from participants for cooperation under strictly confidential conditions.

DNA was extracted by silica based method. All samples were amplified for 22 autosomal STR markers plus DYS391 locus presented in the PowerPlex® Fusion Kit (Promega Corp, Madison, Wisconsin) following manufacturer's instructions.

Forensic parameters (i.e. Typical Paternity Index (TPI), Power of Discrimination (PD) and Power of Exclusion (PE)) were calculated using an *in home* modified version of the Powerstats Version 1.2 software package (Promega, Madison, WI, USA) [9].

Exact tests for Hardy–Weinberg equilibrium were performed with Arlequin Version 3.5 [10], as well as the inter-population comparisons. A significance level of 0.05 was used for all the statistical tests. Bonferroni's correction was used for Hardy–Weinberg equilibrium and linkage disequilibrium tests between pairs of loci, which assume that a 0.05 significance level obtained for 22 tests (one per locus) yields an actual significance threshold of 0.0023 [11,12].

Two samples showed a tri-allele pattern (0.36%) as follows: one sample showed alleles 11,12,13 in Penta D locus, although no tri-allele nor peak imbalance was found in D21S11. The other sample presented alleles 8,10,11 in TPOX locus. Both samples were re-extracted, re-amplified and their genotypes were replicated (data not shown).

The allele frequencies and statistical parameters of forensic interest for the 22 autosomal markers are shown in Supplementary Table 1. The most polymorphic system was Penta E (PIC = 0.8879) whereas the less one was D22S1045 (PIC = 0.6003). The PD ranged from 0.9798 (Penta E) to 0.8219 (D22S1045) with PE values spanning from 0.7715 (Penta E) to 0.3305 (D22S1045). The He extended from 0.8544 (D2S1338) to 0.6543 (TPOX). No departures from HWE were observed in all loci after Bonferroni's correction for multiple testing (0.05/22 = 0.0023), (Supplementary Table 1).

The combined Power of Exclusion (PE) and Combined Power of Discrimination (PD) for the 22 aSTR set were 0.9999999993 and 0,99999999999999999999999993 respectively.

Tests for sample differentiation among the 6 sampled Paraguayan regions (region 1 to region 6) showed no differentiation for the global test. Thus, the differentiation tests showed that the regions sampled are homogeneous among them for these 22 aSTR set.

Test of differentiation (Supplementary Table 2) shows the results of single locus comparisons for the 22 aSTR markers included in the PowerPlex® Fusion System with published data from other relevant populations of America, Europe and Asia [13–19]. Low differentiation with Argentina, south of Brazil and Hispanic American was found as expected.

This paper shows that the population studied in the most populated departments of Paraguay does not present significant

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population substructure among the different geographical locations sampled, allowing the use of these genetic frequencies for forensic genetics purposes in Paraguay. Furthermore, information on 22 aSTR presented in this study increases the resolution in database searching, it is useful for resolution of deficient paternity cases or paternity cases with inconsistencies, increasing both, the Power of Discrimination and the Power of Exclusion.

This paper follows the guidelines for publication of population genetics data requested by the journal [20].

Proficiency testing of the GHEP-ISFG WG (Spanish–Portuguese Speaking Working Group of International Society for Forensic Genetics) (http://www.gep-isfg.org) and the proficiency testing of the Argentine Society for Forensic Genetics (SAGF) (http://www.sagf.org.ar) are regularly carried out.

Appendix A. Supplementary data

Supplementary data associated with this article can be found, in the online version, at http://dx.doi.org/10.1016/j.fsigen.2016.08.002.

References

[1] B. Budowle, T. Moretti, S. Niezgoda, B. Brown, CODIS and PCR-based short tandem repeat loci: law enforcement tools, Proceedings of the Second European Symposium on Human Identification, Madison, Wisconsin: Promega Corporation, 1998, pp. 73–88 Available at http://www.promega.com/products/pm/genetic-identity/ishi-conference-proceedings/2nd-eshi-oral-presentations/ (accessed 2.07.16).
[2] J.M. Butler, Genetics and genomics of core short tandem repeat loci used in human identity testing, J. Forensic Sci. 51 (2006) 253–265.
[3] J.M. Butler, Advanced Topics in Forensic DNA Typing: Methodology, first ed., Elsevier Academic Press, New York, 2012.
[4] Promega Corporation, Powerplex Fusion kit http://www.promega.com/products/pm/genetic-identity/powerplex-fusion (accessed 2.07.16) (2016).
[5] Thermofisher Scientific, GlobalFiler information https://www.thermofisher.com/ar/es/home/industrial/forensics/human-identification/forensic-dna-analysis/pcr-amplification-forensic-dna-profiling/globalfiler.html (accessed 2.07.16) (2016).
[6] E. Martinez-Espin, F. Fernandez-Rosado, J.C. Alvarez, C. Entrala, J.A. Lorente, M. Oviedo de Duarte, E. Villanueva, B. Budowle, Paraguayan population data on the fifteen STR loci included in the Powerplex 16™ kit, J. Forensic Sci. 48 (2003) 253–255.
[7] Dirección General de Estadística, Encuestas y Censos de la Secretaría Técnica de Planificación de la Presidencia de la República de Paraguay. http://www.dgeec.gov.py/Publication/index.php (accessed 2.07.16).
[8] Ethnologue: Languages of the World, in: M. Lewis, Paul, F. Gary Simons, D. Charles Fennig (Eds.), nineteenth edition, SIL International, Dallas, Texas, 2016.
[9] A. Tereba, Profiles in DNA 3, Tools for Analysis of Population Statistics, Promega Corporation, 1999. http://www.promega.com/geneticidtools.
[10] L. Excoffier, H.E.L. Lischer, Arlequin suite ver 3.5: a new series of programs to perform population genetics analyses under Linux and Windows, Mol. Ecol. Resour. 10 (2010) 564–567.
[11] B.S. Weir, Genetic Data Analysis II: Methods for Discrete Population Genetic Data, Sinauer Associates, USA, 1996, pp. 109–110.
[12] K. Tamura, G. Stecher, D. Peterson, A. Filipski, S. Kumar, MEGA6: molecular evolutionary genetics analysis version 6.0, Mol. Biol. Evol. 30 (2013) 2725–2729.
[13] A. Borosky, U. Toscanini, A. Gómez, M.L. Parolín, N. Basso, C. Vullo, Forensic population data for 20 STR loci in Argentina, Forensic Sci. Int. Genet. 13 (2014) e20–e21.
[14] C. Vullo, A. Borosky, C. Romanini, L. Catelli, T. Yamamoto, Frequency data for 12 mini STR loci in Argentina, Forensic Sci. Int. Genet. 4 (2010) e79–e81.
[15] L. Fernandez-Formoso, C. Phillips, A. Rodriguez, R. Calvo, Allele frequencies of 20 STRs from northwest Spain (Galicia), Forensic Sci. Int. Genet. 6 (2012) e149–e150.
[16] L. Fernández, O. García, P. Martín, C. Albarrán, P. García, M. Sancho, A. Alonso, A Spanish Population Study of the STR Loci D2S1338, D19S433, Penta D, and Penta E, J. Forensic Sci. 48 (2003) 1182.
[17] A. Gastaldo, R. Rodenbusch, N. Da Fré, A. Mardini, C. Alho, Allele frequencies of 20 autosomal STR in the population from Rio Grande do Sul Southern Brazil, Forensic Sci. Int. Genet. 19 (2015) 15–17.
[18] O. Rocabado, P. Taboada, F. Inda, I. Yurrebaso, O. García, Population genetic data for 15 STR loci (IdentifilerTM kit) in Bolivia, Legal Med. 11 (2009) 302–304.
[19] C.R. Hill, D.L. Duewer, M.C. Kline, M.D. Coble, J.M. Butler, U.S. population data for 29 autosomal STR loci, Forensic Sci. Int. Genet. 7 (2013) e82–e83.
[20] A. Carracedo, J.M. Butler, L. Gusmão, A. Linacre, W. Parson, L. Roewer, P.M. Schneider, Update of the guidelines for the publication of genetic population data, Forensic Sci. Int. Genet. 10 (2014) A1–A2.

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