Forensic Science International: Genetics xxx (2016) xxx–xxx

Contents lists available at ScienceDirect

Forensic Science International: Genetics

journal homepage: www.elsevier.com/locate/fsig

Correspondence

Genetic polymorphism of 22 autosomal STR markers in a Han population of Southern China

ARTICLE INFO

Article history:
Received 12 April 2016
Received in revised form 21 June 2016
Accepted 22 June 2016
Available online xxx

Keyword:
Short tandem repeats (STR)
Population data
Linkage disequilibrium
Kinship analysis
Chinese Han population

ABSTRACT

Population genetic data and forensic statistics of 22 autosomal short tandem repeat (STR) loci (D1S1656, D2S1338, D3S3045, D4S2366, D5S2500, D6S477, D7S3048, D8S1132, D9S925, D10S1435, D11S2368, D12S391, D13S325, D14S608, D15S659, D16S539, D17S1290, D18S535, D19S253, D20S470, D21S1270 and GATA198B05) were determined for a sample of 515 unrelated individuals from Han population in Southern China. The expected heterozygosity and the discrimination power varied from 0.7358 to 0.8733 and 0.8915 to 0.9702, respectively. The probability of excluding an unrelated man as the true father (assuming no background relatedness in the population) for trios and for duos ranged from 0.5126 to 0.7415 and 0.3331 to 0.5864, respectively. The studied STRs appear to provide a significant improvement in the statistical power of kinship analysis.

Dear Editor,

In recent years, extended short tandem repeat (STR) markers from the European standard set (ESS) or Combined DNA Index System (CODIS) have been introduced into the forensic community to increase the discrimination power [1,2] and standardize the analysed markers used in different countries. However, supplementary STRs are required to significantly evaluate pairwise, distant and/or complex relationships [3–5]. Additionally, additional STRs may be needed to confirm paternity in cases in which genetic mutations occur between the parents and progeny [6], null alleles are present [7] or a relative of an alleged father is involved [8–10].

The requirement of additional STRs has resulted in the development of a few new forensic autosomal multiplex STR systems in China, such as the Microreader^{TM}23sp ID System (Suzhou Microread Genetics Co., Ltd, Suzhou, Jiangsu Province, China). This kit was used to simultaneously amplify 22 STR loci (D3S3045, D4S2366, D6S477, D7S3048, D8S1132, D9S925, D10S1435, D11S2368, D13S325, D14S608, D15S659, D17S1290, D18S535, D19S253, D20S470, D21S1270, GATA198B05, D1S1656, D2S1338, D5S2500, D12S391 and D16S539) and the amelogenin gender marker. Most of these loci have not been widely applied in the forensic community. We obtained genetic data for the reference samples from a Chinese Han population in southern China.

After obtaining informed consent, oral swab samples were collected from 515 unrelated individuals whose forefathers have resided in the Guangdong Province (405 samples) or Guangxi Province (110 samples) in southern China for at least 3 generations. DNA extraction was carried out using the Chelex-100 method [11] or DNA IQ system (Promega Corporation, Madison, WI, USA).

All DNA samples were amplified using the Microreader^{TM}23sp ID System following the manufacturer's protocol. PCR reactions were performed in a GeneAmp 9700 PCR system Thermal Cycler (Applied Biosystems). Amplified products were analysed and detected using capillary electrophoresis in a 3500xL Genetic analyser (Applied Biosystems, Foster City, CA), and allele calling was obtained using the GeneMapper ID-X 1.2 software (Applied Biosystems). Allele assignment was determined by comparison with referenced ladders that were provided with the kit. Quality control was performed according to the laboratory internal control standards and kit controls.

Arlequin software v 3.5 [12] was used to calculate the allele frequencies, observed heterozygosity (H_o) and expected heterozygosity (H_e) as well as to assess departures from the Hardy–Weinberg equilibrium (HWE), linkage disequilibrium (LD) and population differentiation. The Power of Discrimination (PD) and polymorphic information content (PIC) were computed as previously described [13]. Probabilities of excluding the relatives of the true father from paternity for trios (TPE_{k_0}) and for duos (DPE_{k_0}) were calculated as previously described [9], assuming no background relatedness in the population ($\theta = 0$) and considering only non-inbred alleged father and child. The parameter k_0 is the probability of the alleged father and the child not sharing any identity by descent alleles: $k_0 = 1$ for unrelated individuals, $k_0 = 3/4$ for third degree relatives (first cousins, great grandparent-great grandoffspring, half-avuncular or grand-avuncular), $k_0 = 1/2$ for second degree relatives (half-sibs, grandparent-

http://dx.doi.org/10.1016/j.fsigen.2016.06.017
1872-4973/

grandchild or avuncular), $k_0 = 1/4$ for full sibs and $k_0 = 0$ for parent/child [14,15].

Electropherograms showing ladders and the genotypes of a DNA sample amplified using the Microreader™23sp ID System are shown in Fig. S1. The allele frequencies, population statistics parameters and forensic statistical parameters for the 22 studied STR loci are presented in Table S1. The number of alleles observed at each locus varied from 8 to 16 with the corresponding allele frequencies ranging from 0.0010 to 0.4107. Applying a 0.05 significance level, all of the loci analysed reached the Hardy-Weinberg equilibrium ($P > 0.05$), except for the D1S1656 ($P = 0.0172$), D10S1435 ($P = 0.0113$) and D13S325 ($P = 0.0075$) loci (Table S1). However, after the Bonferroni's corrected significance level ($P = 0.05/22 = 0.0023$) was employed, the statistical significance was no longer observed. Pairwise linkage disequilibrium tests showed that there were 27 pairs of departure out of a total of 231 interclass correlation tests. However, after the Bonferroni correction was applied, no pairs were still significant in the LD test.

The forensic statistical parameters (Table S1) showed that the most informative locus was D7S3048, which had the highest expected heterozygosity (0.8733). The least informative locus was D10S1435, which had an expected heterozygosity of 0.7358. The average expected theoretical heterozygosity across all loci was 0.8168. The power of discrimination (PD) value was observed from 0.8915 (D10S1435) to 0.9702 (D7S3048). The combined power of discrimination (CPD) of the 22 STR loci was $1–2.4979 \times 10^{-28}$. These results suggest that the 22 STRs contain highly polymorphic information that could be used for forensic purposes.

To assess the genetic informativeness of the STRs for paternity testing, we computed the TPE_{k_0} and DPE_{k_0} for different autosomal kinship between the alleged father and child (Table S2) [9]. The conventional probabilities of excluding an unrelated man as the father for trios and duos were lowest at D10S1435 (0.5126 and 0.3331, respectively) and highest at D7S3048 (0.7415 and 0.5864, respectively). The combined power of exclusion (CPE) of the 22 STR loci for trios and duos was $1–1.090 \times 10^{-10}$ and 0.999999321, respectively. These results indicated that the 22 STRs could provide a significant improvement in the probability of solving paternity cases involving a relative as the father.

In kinship analyses, the forensic DNA community is concerned with the genetic linkage and tendency of alleles at two or more syntenic STR markers to be inherited together during meiosis [16,17]. Hence, we present the mapping positions and genetic locations of the 22 STR loci, as well as common forensic loci on the same chromosome, in Table S3. In addition, the mutation rates of 7 out of the 22 STRs have been reported in References [18] and [19]. Their results indicated that the mutation rates were similar to those of commonly used forensic STR loci.

Allelic frequencies for the twenty-two STRs were compared to the previously published Chinese Han population data estimated from southern China (Guangdong) [20], eastern China (Shanghai, Zhejiang) [19,21,22], northern China (Hebei) [23] and northwestern China (Chengdu) [24]. Only the frequencies for D18S535 were significantly different from the data from Shanghai (Table S4). This difference may be the result of our sampling process.

In summary, the 22 autosomal STR loci contain highly polymorphic genetic information. As supplementary markers, these STRs can improve the statistical power of kinship analysis, such as paternity cases involving a relative as the father and pairwise kinship cases that cannot be sufficiently resolved using routine STRs.

Conflict of interest

The authors declare no conflicts of interest.

Acknowledgements

This work was supported by grants from the National Natural Science Foundation of China (81373245) and the fundamental research funds of Sun Yat-sen University for the medical young teachers' Cultivation Project (14ykpy02).

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.2016.06.017.

References

[1] P. Gill, L. Fereday, N. Morling, P.M. Schneider, New multiplexes for Europe-amendments and clarification of strategic development, Forensic Sci. Int. 163 (1–2) (2006) 155–157.
[2] D.R. Hares, Selection and implementation of expanded CODIS core loci in the United States, Forensic Sci. Int. Genet. 17 (2015) 33–34.
[3] M. Nothnagel, J. Schmidtke, M. Krawczak, Potentials and limits of pairwise kinship analysis using autosomal short tandem repeat loci, Int. J. Legal Med. 124 (3) (2010) 205–215.
[4] O. Skare, N. Sheehan, T. Egeland, Identification of distant family relationships, Bioinformatics 25 (18) (2009) 2376–2382.
[5] I. Carboni, S. Iozzi, A.L. Nutini, F. Torricelli, U. Ricci, Improving complex kinship analyses with additional STR loci, Electrophoresis 35 (21–22) (2014) 3145–3151.
[6] R. Chakraborty, D.N. Stivers, Paternity exclusion by DNA markers: effects of paternal mutations, J. Forensic Sci. 41 (4) (1996) 671–677.
[7] M.C. Kline, C.R. Hill, A.E. Decker, J.M. Butler, STR sequence analysis for characterizing normal, variant, and null alleles, Forensic Sci. Int. Genet. 5 (4) (2011) 329–332.
[8] W.K. Fung, Y.K. Chung, D.M. Wong, Power of exclusion revisited: probability of excluding relatives of the true father from paternity, Int. J. Legal Med. 116 (2) (2002) 64–67.
[9] N. Pinto, L. Gusmao, T. Egeland, A. Amorim, Paternity exclusion power: comparative behaviour of autosomal and X-chromosomal markers in standard and deficient cases with inbreeding, Forensic Sci. Int. Genet. 7 (2) (2013) 290–295.
[10] N. Pinto, M. Magalhaes, E. Conde-Sousa, C. Gomes, R. Pereira, C. Alves, L. Gusmao, A. Amorim, Assessing paternities with inconclusive STR results: the suitability of bi-allelic markers, Forensic Sci. Int. Genet. 7 (1) (2013) 16–21.
[11] P.S. Walsh, D.A. Metzger, R. Higuchi, Chelex 100 as a medium for simple extraction of DNA for PCR-based typing from forensic material, Biotechniques 10 (4) (1991) 506–513.
[12] L. Excoffier, H.E. Lischer, Arlequin suite ver 3.5: a new series of programs to perform population genetics analyses under Linux and Windows, Mol. Ecol. Resour. 10 (3) (2010) 564–567.
[13] W. Goodwin, A. Linacre, S. Hadi, Forensic parameters, An Introduction to Forensic Genetics, Wiley-Blackwell, Chichester, West Sussex, UK, 2011, pp. 181–182.
[14] M.S. Blouin, DNA-based methods for pedigree reconstruction and kinship analysis in natural populations, Trends Ecol. Evol. 18 (10) (2003) 503–511.
[15] N. Pinto, P.V. Silva, A. Amorim, General derivation of the sets of pedigrees with the same kinship coefficients, Hum. Hered. 70 (3) (2010) 194–204.
[16] A.A. Westen, H. Haned, L.J. Grol, J. Harteveld, K.J. van der Gaag, P. de Knijff, T. Sijen, Combining results of forensic STR kits: HDplex validation including allelic association and linkage testing with NGM and Identifiler loci, Int. J. Legal Med. 126 (5) (2012) 781–789.
[17] P. Gill, C. Phillips, C. McGovern, J.A. Bright, J. Buckleton, An evaluation of potential allelic association between the STRs vWA and D12S391: implications in criminal casework and applications to short pedigrees, Forensic Sci. Int. Genet. 6 (4) (2012) 477–486.
[18] D. Lu, Q. Liu, W. Wu, H. Zhao, Mutation analysis of 24 short tandem repeats in Chinese Han population, Int. J. Legal Med. 126 (2) (2012) 331–335.
[19] S. Zhang, Z. Zhang, S. Zhao, Z. Zhao, C. Li, Genetic polymorphisms in 12 autosomal STRs in a Shanghai Han population from China, Electrophoresis 34 (4) (2013) 613–617.
[20] D.J. Lu, Q.L. Liu, H. Zhao, Genetic data of nine non-CODIS STRs in Chinese Han population from Guangdong Province, Southern China, Int. J. Legal Med. 125 (1) (2011) 133–137.

[21] S. Zhang, H. Tian, J. Wu, S. Zhao, C. Li, A new multiplex assay of 17 autosomal STRs and Amelogenin for forensic application, PLoS One 8 (2) (2013) e57471.
[22] A. Zhou, W. Wu, Q. Liu, Y. Wu, D. Lu, Population genetic data of the NGM SElect STR loci in Chinese Han population from Zhejiang region, Chin. Int. J. Legal Med. 127 (2) (2013) 377–378.
[23] L. Lei, J. Xu, Q. Du, L. Fu, X. Zhang, F. Yu, C. Ma, B. Cong, S. Li, Genetic polymorphism of the 26 short tandem repeat loci in the Chinese Hebei Han population using two commercial forensic kits, Mol. Biol. Rep. 42 (1) (2015) 217–225.
[24] J.N. Li, H.B. Luo, F. Song, Y.P. Hou, Genetic polymorphisms of 22 autosomal STR loci in Chinese Han population, Forensic Sci. Int. Genet. Supp. Ser. 5 (2015) e45–e47.

Qiu-Ling Liu
Zi-Xiang Chen
Chu-Guang Chen
De-Jian Lu*
Faculty of Forensic Medicine, Zhongshan Medical School of Sun Yat-Sen University, 74 Zhongshan 2nd Road, Guangzhou 510080, PR China

Suzhou Microread Genetics Co., Ltd., China, Room 312, No.15 Building, Biomedical Engineering Park, No.8 Jinfeng Road, SND, Suzhou 215163, Jiangsu, PR China

Faculty of Forensic Medicine, Zhongshan Medical School of Sun Yat-Sen University, 74 Zhongshan 2nd Road, Guangzhou 510080, PR China

* Corresponding author.
E-mail addresses: liuql@mail.sysu.edu.cn (Q. Liu), lanczx@163.com (Z. Chen), mr_ccg@126.com (C. Chen), lvdejian@mail.sysu.edu.cn (D. Lu).

Received 12 April 2016
Received in revised form 21 June 2016
Accepted 22 June 2016
Available online xxx

Please cite this article in press as: Q.-L. Liu, et al., Genetic polymorphism of 22 autosomal STR markers in a Han population of Southern China, Forensic Sci. Int. Genet. (2016), http://dx.doi.org/10.1016/j.fsigen.2016.06.017