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Short Communication

Fixed bin frequency distribution for the VNTR Loci D2S44, D4S139, D5S110, and D8S358 in a population sample from Minas Gerais, Brazil

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Abstract

Fixed bin frequencies for the VNTR loci D2S44, D4S139, D5S110, and D8S358 were determined in a Minas Gerais population sample. The data were generated by RFLP analysis of *HaeIII*-digested genomic DNA and chemiluminescent detection. The four VNTR loci have met Hardy-Weinberg equilibrium, and there was no association of alleles among VNTR loci. The frequency data can be used in forensic analyses and paternity tests to estimate the frequency of a DNA profile in the general Brazilian population.

Key words: fixed-bin frequency, VNTR loci, Brazilian population.

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Introduction

DNA polymorphisms detection through restriction fragment length polymorphism (RFLP) analysis has been used as a powerful tool for identity testing in forensic science (Budowle and Baechtel, 1990). Individuals are distinguished by VNTR (variable number of tandem repeats) variation, and their frequency distribution may be classified into a small number of fixed bins, a common technical approach that allows genotyping unambiguously. However, to perform identity tests through VNTRs, a database of fixed bin frequency is required. This investigation describes the RFLP allelic frequencies of the Minas Gerais population, Brazil, distributed in fixed bins for the *HaeIII*-generated VNTR loci D2S44, D4S139, D5S110, and D8S358.

Materials and Methods

The source of DNA consisted of blood samples collected randomly from both unrelated parents of trios that underwent paternity tests. These samples were chosen from a blood bank of a Private Reference Laboratory of Minas Gerais State, which were collected in a three-year period (1997 to 1999). Samples were taken from White and Mulatto individuals; however, due to the great racial mixture in Brazil, ethnic composition was not properly annotated, but mulattoes represented the majority of the sample (80%). DNA was extracted organically from peripheral blood samples, digested with *HaeIII*, and separated in 0.8% agarose gel electrophoresis. Southern transferred to nylon membrane (Byodine A) were hybridized to alkaline phosphatase-conjugated probes (ACES 2.0 plus Kit), according to the supplier's protocol (Life Technologies, Gibco-BRL, Gaithersburg, MD). The probes PH30 (D4S139), LH1 (D5S110), and CEB42 (D8S358) were purchased from Life Technologies (Gibco-BRL, Gaithersburg, MD). The probe YNH4 (D2S44) probe was supplied by LifeCodes Corp. (Valhalla, NY). The estimated base pair sizing of the digested DNA was performed based on a Gibco-BRL molecular weight marker (*i.e.* sizing ladder) in a video documentation system (ImageMaster VDS System, Pharmacia Biotech, San Francisco, CA). The allele size data were distributed in fixed bins according to Budowle *et al*. (1991a).

Hardy-Weinberg equilibrium (HWE) was demonstrated by the likelihood ratio test (Edwards *et al.*, 1992) and by the exact test (Guo and Thompson, 1992). An interclass correlation criterion for two-locus associations was used to detect disequilibrium between VNTR loci (Karlin *et al*., 1981). Independence across the four VNTR loci was determined by examining whether the observed variance of the number of heterozygous loci in the population sample was outside its confidence interval under the assumption of independence (Chakraborty, 1984).

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Results and Discussion

Table I presents the 31 fixed bin frequency distributions for the four highly polymorphic VNTR loci in the Minas Gerais population. This is the first report on the distribution of bin frequencies for the D8S358 VNTR locus in the general Brazilian population. Statistical analysis demonstrated that allelic frequencies at all four loci are in HW equilibrium, and there was no correlation among alleles in any of the loci pairs based on the interclass correlation tests. Furthermore, there was no evidence of association among the four loci described in this population sample using the S_k^2 criterion ($S_k^2 = 0.302$, confidence interval 0.162-0.435).

Population data (bins) of the four VNTR loci for Caucasians in the United States (Budowle *et al.*, 1994) were compared to our Brazilian data. There were very few instances in which substantial differences in fixed bin frequencies could be observed between the two population samples, even considering the great difference of in ethnical composition of both countries. Under the assumption of independence, this result suggests that there would

Table I - Fixed bin frequencies for four VNTR loci in Minas Gerais, Brazil.

Bin	Size (bp)	D2S44	D4S139	D5S110	D8S358
1	1-639	0.000	0.000	0.000	0.002
2	640-772	0.019	0.000	0.002	0.005
3	773-871	0.004	0.000	0.002	0.000
4	872-963	0.016	0.000	0.002	0.000
5	964-1077	0.008	0.000	0.004	0.000
6	1078-1196	0.027	0.000	0.005	0.009
7	1197-1352	0.086	0.000	0.007	0.023
8	1353-1507	0.057	0.000	0.030	0.040
9	1508-1637	0.105	0.002	0.021	0.023
10	1638-1788	0.103	0.000	0.030	0.028
11	1789-1924	0.055	0.000	0.027	0.019
12	1925-2088	0.047	0.000	0.047	0.042
13	2089-2351	0.076	0.010	0.086	0.053
14	2352-2522	0.035	0.000	0.056	0.035
15	2523-2692	0.033	0.010	0.066	0.065
16	2693-2862	0.074	0.002	0.066	0.070
17	2863-3033	0.078	0.002	0.059	0.084
18	3034-3329	0.057	0.020	0.079	0.081
19	3330-3674	0.053	0.033	0.099	0.105
20	3675-3979	0.027	0.037	0.054	0.056
21	3980-4323	0.018	0.047	0.045	0.056
22	4324-4821	0.002	0.084	0.072	0.044
23	4822-5219	0.002	0.069	0.018	0.030
24	5220-5685	0.006	0.074	0.047	0.040
25	5686-6368	0.008	0.141	0.020	0.049
26	6369-7241	0.004	0.166	0.023	0.028
27	7242-8452	0.000	0.111	0.023	0.005
28	8453-10093	0.000	0.097	0.009	0.007
29	10094-11368	0.000	0.027	0.000	0.000
30	11369-12829	0.000	0.022	0.000	0.002
31	12830-25000	0.000	0.047	0.000	0.000
Number of chromosomes		512	596	558	430
Individuals-single band		16	23	22	15
HWE/Likelihood ratio (p =)		0.866	0.081	0.127	0.167
HWE/Exact test (p =)		0.742	0.093	0.148	0.141

be little difference in multiple loci profile frequency estimates using both databases.

In conclusion, this article provides data for estimated frequencies in fixed bins of D2S44, D4S139, D5S110 and D8S358 VNTR loci. Results demonstrated that the fixed bin frequencies of the four loci can be used in forensic analysis and paternity tests for the Brazilian population.

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