

TECHNICAL NOTE

Pilar Nievas Marco,¹ Bs.C.; Begoña Martínez-Jarreta,¹ Ph.D.; Emilio Abecia Martínez,¹ Ph.D.; Amparo Prades Sanchis,¹ Bs.C.; and Rafael Hinojal Fonseca,² Ph.D.

Allele Frequency Distribution of the STR Loci HUMTPOX, HUMTH01 and HUMVWA in Asturias (North Spain)

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ABSTRACT: In order to use genetic loci in forensic identity testing, some population data are needed. This paper presents a report of allele frequency data for the loci HUMTH01, HUMTPOX and HUMVWA in a population sample from Northern Spain.

No deviation from the Hardy-Weinberg equilibrium was detected in any of the three markers investigated and there was no evidence of association between the alleles of these loci.

Statistical analysis was also carried out to obtain some parameters of medicolegal interest and comparative studies were carried out with other populations studied to date for these five loci. The Asturian sample does not differ substantially from other Caucasian and Spanish populations.

KEYWORDS: forensic science, DNA typing, short tandem repeats, HUMTH01, HUMTPOX, HUMVWA population genetics, Spain

Short tandem repeat (STR) loci are a subgroup of a highly polymorphic variable number of tandem repeat (VNTR) loci (1,2) that have proved to be very useful for the analysis of forensic DNA evidence. Some tetrameric STR systems like HUMTPOX, HUMTH01 and HUMVWA have become widely used in forensic casework analyses, and population data are needed to estimate the rarity of a DNA profile (3–13).

This survey was performed to test the allele frequencies of these systems in a Northern Spanish population (Asturias), which has been poorly studied from a genetic point of view (14). Asturias has experienced some migration, not including the Arabic invasions of the Iberian peninsula, and a better genetic characterization of the Spanish region is interesting.

Material and Methods

Blood samples from 117 healthy unrelated individuals from Asturias were collected. DNA was extracted as previously described (14).

¹ Research scientist, research scientist and lab director, and research scientists, respectively, Department of Legal Medicine, University of Zaragoza, Spain.

² Professor of legal medicine, Department of Legal Medicine, University of Oviedo (Asturias), Spain.

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Amplification of HUMTH01, HUMTPOX and HUMVWA was achieved using the primers described by Edwards et al. (2), Anker et al. (15), and Kimpton et al. (16), respectively. The reaction assay and the amplification conditions were carried out as described by Gené et al. (3,4) and Pestoni (9).

All reactions, together with positive and negative control samples, were performed in a Perkin-Elmer DNA Thermal Cycler 480. Separation was carried out on 6% (w/v acrylamide/bisacrylamide) polyacrylamide denaturing high-performance DNA sequencing gel (Ready Mix Gel ALF grade, Pharmacia). All fresh PCR products were typed twice. The electrophoresis was carried out on the Automated Laser Fluorescent (ALF) DNA Sequencer (Pharmacia) at 1450 V, 38 mA, 45 W and 50°C with laser power at 3 mW for 220 min.

Amplified DNA was mixed with internal fluorescent-labeled size standards, and external lane ladders were also used for adjustment. Sequenced allelic ladders were used for each system as recommended by the DNA Commission of the International Society of Forensic Haemogenetics (17,18). Statistical analyses were performed as described previously (see Refs 5, 14 and 19 for details).

Results and Discussion

The distributions of observed allele frequencies for the three loci are summarized in Table 1. No deviation from the Hardy-Weinberg equilibrium was detected in any of the loci investigated (Tables 2 and 3) and there was no evidence of association detected between the alleles of the loci (Table 4).

Some statistics of medicolegal interest, such as the allelic diversity value and the chance of exclusion in cases of paternity, were also obtained (Table 5).

The Asturian data were compared with other sample populations (3–13) using a RxC contingency table χ^2 test for homogeneity (see Table 6).

No significant differences were found between Asturians and other Caucasians like the Swiss and Spanish (Andalucia, Central Spain and Galicia, which is situated next to the region of Asturias). The data suggest that there is a general uniformity for the STR loci in all the Spanish populations studied to date, including Catalonia, the Pyrenees and the Basque Country. However for HUMTH01 slight differences were observed between the Asturian and these other populations. These results are consistent with those from previous studies carried out by us on some other Northern Spanish populations (5), and a better characterization of this should

TABLE 1—Observed allele frequency distribution for the STR loci: HUMTH01, HUMTPOX, HUMVWFA31 in 117 unrelated individuals from Asturias.

	HUMTH01	HUMTPOX	HUMVWA
6	0.222
7	0.158
8	0.175	0.543	...
9	0.154	0.068	...
9.3	0.269
10	0.021	0.081	...
11	...	0.265	...
12	...	0.043	...
13
14	0.104
15	0.104
16	0.292
17	0.288
18	0.162
19	0.043
20	0.007
P_{min}^*	0.0245	0.0223	0.0221

* Minimum allele frequency (20).

TABLE 2—Test of Hardy-Weinberg expectations of genotype frequencies.

	HUMTH01	HUMTPOX	HUMVWA
χ^2	0.285	0.797	0.719
Likelihood ratio test	0.389	0.594	0.163
Exact test	0.294	0.643	0.750

TABLE 3—Observed and expected heterozygous and homozygous classes for the STR loci in the sample studied.

	HUMTH01	HUMTPOX	HUMVWA
Obs. het.*	13	9	15
Exp. het. \pm SE†	13.08 \pm 2.11	8.69 \pm 1.74	14.17 \pm 1.59
Obs. hom.‡	6	3	5
Exp. hom. \pm SE§	4.91 \pm 0.85	3.15 \pm 1.58	4.77 \pm 1.70

* Observed heterozygosity.

† Expected heterozygosity.

‡ Observed homozygosity.

§ Expected homozygosity \pm standard error.

TABLE 4—Correlation test (p-values).

HUMTH01/HUMTPOX	0.246
HUMTH01/HUMVWA	0.591
HUMTPOX/HUMVWA	0.357

be recommended. Unfortunately there is not enough population data available for these genetic markers on Arabic groups. The collection of these data would be very interesting since it could help to a better understanding of the Arabic influence in the Iberian populations.

As could be expected, significant differences were observed between the Asturian sample and non-Caucasian populations such as Caribbean and Japanese (Table 6).

In conclusion, we have obtained allele frequency data that can be used in identity testing to estimate the frequency of a multiple PCR-based profile in the Asturian population. The Asturian sample

TABLE 5—Parameters of forensic efficiency.

	HUMTH01	HUMTPOX	HUMVWA
Obs. H*	0.8034	0.6667	0.7194
Exp. H†	0.8017	0.6248	0.7852
MEC‡	0.5977	0.3798	0.5707
MEP§	0.6023	0.3217	0.5719
PIC	0.7674	0.5711	0.7501
PM¶	0.0797	0.2011	0.0842
PD**	0.9203	0.7989	0.9157

* Observed heterozygosity.

† Expected heterozygosity.

‡ Mean exclusion chance.

§ Mean exclusion probability.

|| Polymorphism information content.

¶ Probability of match.

** Power of discrimination.

TABLE 6—Comparison of different populations for HUMTH01, HUMTPOX and HUMVWFA31 loci.

Populations	HUMTH01	HUMTPOX	HUMVWFA31
Asturias/Catalonia (3,4)	0.0177	0.4054	0.4904
Asturias/Pyrenees (5)	0.0016	0.5480	...
Asturias/Central Spain (6)	0.4955	0.6698	0.5888
Asturias/Andalucia (7)	0.2956	0.4816	...
Asturias/Basque (8)	0.0294	0.9814	0.7027
Asturias/Galicia (9)	0.5448	...	0.6063
Asturias/Switzerland (10,11)	0.3259	0.9300	0.2249
Asturias/Sao Tomé (12)	0.0001	0.0021	0.0001
Asturias/Japon (13)	0.0001	0.0385	0.0001

does not differ substantially from other Caucasian or Spanish populations.

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Additional information and reprint requests:

Prof. Begoña Martínez Jarreta
 Departamento de Medicina Legal
 Facultad de Medicina
 C/Domingo Miral s/n
 50009 Zaragoza, Spain