

Phenotype versus genotype reporting for DNA polymorphisms

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Abstract. In the College of American Pathologists' (CAP) 2001 PI-A survey, consensus was not achieved for the child's phenotype at the HUMTH01 locus. Two alleles (6 and 9.3) were reported by 37 laboratories, and only one allele (9.3) by 30 laboratories. Analysis of the results showed that this discrepancy was related to the kits used. This was attributed to a primer binding site mutation. The mother's phenotype was 8,9.3 and the tested man's 9,9.3. Participants who detected the "6" allele in the child reported a PI of 0, while participants who did not detect it reported a median PI of 1.43. Two participants reported the inferred genotype of 9.3,9.3. This is clearly an incorrect response. This case illustrates that for DNA polymorphisms the phenotypes detected may depend on the testing method used. Thus reporting phenotypes only is sound and scientifically accurate. © 2004 Elsevier B.V. All rights reserved.

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1. Introduction

Historically the Parentage Testing Community has consistently reported the phenotype of tested individuals and not their inferred genotype. When testing relied mostly on blood groups and HLA typing, it was very well recognized that a variety of genotypes could be attributed to a single phenotype. Since the publication of the 1st edition of the Standards for Parentage Testing Laboratories in 1990, the AABB has required that the reports display the "phenotypes established for each individual in each genetic system examined".

With DNA polymorphisms testing methodology, it was argued that the testing directly established a genotype. It became relatively common, specifically in the forensic

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community, to report a homozygous genotype (i.e. two identical alleles) when only one allele was detected.

Since the inception of the parentage testing proficiency survey (PI) in 1993, the CAP Parentage Testing Committee has encouraged the reporting of phenotypes by giving direct instructions on reporting format and downgrading a result reported as a genotype to “acceptable” instead of “good”.

2. Methods

The participant summary reports of all three yearly mailings of the CAP Parentage Testing Survey program for years 1998–2002 were reviewed. Apparently homozygous phenotypes at all STR loci tested by PCR technology were examined for reporting format. The total number of single allele phenotypes was tabulated for each mailing as an absolute number and a percent reported as a genotype (i.e. two identical alleles).

Results on the four individuals tested were also examined to determine whether homozygosity determination influenced the interpretation and/or PI calculation.

3. Results

In the 2001 PI-A survey at the HUMTH01 locus the interpretation of a single allele in the child did influence the PI calculation. A tabulation of the results reported by the participants is listed in [Table 1](#).

Participants using the STR kits from Promega Corporation detected the “6” allele in the child and reported a PI of 0. Participants using the STR kits from Applied Biosystems did not detect the “6” allele and reported a median PI of 1.43. Two participants reported a homozygous genotype of 9.3, which is clearly inaccurate.

Table 1
Results at the HUMTH01 locus 2001 PI-A survey

Specimen	Phenotype	Participants		Performance grade
		No.	%	
Mother	8,9,3	65	92.8	Good
	8,9,3/10	2	2.9	Acceptable
	8,10	2	2.9	Unacceptable
	8	1	1.4	Unacceptable
Child	6,9,3	35	50.0	
	6,9,3/10	2	2.9	
	9,3	28	40.0	
	9,3,9,3	2	2.9	Non-Consensus
	6,10	2	2.9	
	6	1	1.3	
Tested man	9,9,3	63	91.4	Good
	9,9,3/10	2	2.9	Acceptable
	9,10	2	2.9	Unacceptable
	9	1	1.4	Unacceptable
	9,3	1	1.4	Unacceptable

Table 2
Phenotype versus genotype reporting of single allele phenotypes

Year	Single allele phenotypes observed	Total no. of reported as genotypes	% Reported as genotypes
1998	1611	116	7.2
1999	1693	71	4.2
2000	2428	210	8.6
2001	2462	168	6.8
2002	2927	165	5.6

The data collected from 15 mailings are depicted in Table 2.

4. Discussion

In the PI 2001A mailing at the HUMTH01 locus the child's allele had a primer site mutation, causing a lack of amplification with one of two manufacturer's kits. The sample did not achieve consensus as 39 laboratories reported two alleles, while 31 reported a single allele. It is clear from the results that the child was heterozygous for 6 and 9.3. The mother's phenotype was 8,9.3 and tested man's was 9,9.3, thus this locus' results should lead to an inconsistency interpretation. For the laboratories that did not detect the "6" allele, it was reasonable to report the child's phenotype as 9.3 and to calculate a non-0 paternity index. However it was inaccurate to report the child's genotype as 9.3,9.3. Lack of detection of alleles due to primer site mutation can result in apparent homozygosity at a locus. This phenomenon is now well recognized and is not restricted to a single manufacturer's reagents [1,2].

There seemed to be a decreasing trend in the frequency of reporting single allele phenotypes as inferred genotypes before 2000. This frequency increased in 2000. The possibility that this was due to an influx of new laboratories into the survey program in 2000 was entertained. A review of the number of laboratories reporting STR results for the most frequently reported locus, VWA31, did not support this theory. After 2000, the frequency is again decreasing.

References

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