Comparability of RFLP Results Between Laboratories: AABB/ CAP Survey Data

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INTRODUCTION

The American Association of Blood Banks (AABB) and the College of American Pathologists (CAP) jointly sponsor a proficiency testing program for parentage and forensic laboratories. This report summarizes data on band size (Kb) and paternity index based on RFLP analysis.

METHOD

Results from seven quarterly mailings of three whole blood samples collected in ACD were evaluated to determine variability in band size reported by laboratories testing the same locus with similar probe/enzyme combinations. Extraction method, restriction condition, electrophoresis, hybridization , and detection system varied depending on the laboratories routine for testing samples. Standard deviation (SD) and coefficient of variation (CV) were determined for combinations with multiple responses. Each laboratory was requested to provide results on K562 controls, bin limits and frequency, database source, delta value, and the paternity index for the system tested.

RESULTS

Tables 1 shows that laboratories using the same probe/enzyme combination for testing at the D2S44 locus have comparable results. With the exception of the B-93 survey the CV for the results has been below 3.2%. On all of the seven surveys conducted between January 1993 and May of 1995, band size variation has been less than 100 base pairs. Table 2 shows the results from 17 laboratories reporting on the trio mailed in March of 1995. These results are representative of what has been observed with a large number of other probe/enzyme combinations. In all cases the SD and CV of the band size are very similar regardless of the reporting laboratory. As in table 2, it has been observed that the variation in reported band size increases as the marker detected is bigger. Table 3 shows examples of the range of values for the paternity index reported for various probe/enzyme combinations. The widely divergent values reported for similar test results is representative of all the AABB/CAP surveys evaluated to date. There is no apparent correlation of this variation with the databases used, the reported delta values, or the bin limits.

CONCLUSION

Inter laboratory comparison of RFLP testing on seven trios tested by similar

probe/enzyme combinations show that band sizes reported by multiple laboratories are very similar. The observed SD and CV are small although there is an increase in the variability as the band size increases. In contrast to the comparability of band size, the paternity index values reported on similar RFLP results show wide variability.

TABLE 1

Comparability of RFLP Allele Sizes

D2S44/YNH 24, Hae III

Survey (n)	$\bar{X}$ (Kb)	SD (Kb)	CV (%)
A -93 (15)	2.98	0.064	2.16
B -93 (11)	1.14	0.081	7.13
C -93 (17)	1.99	0.062	3.13
A -94 (30)	2.81	0.057	2.02
B -94 (24)	3.08	0.096	3.1
C -94 (26)	1.56	0.024	1.6
A -95 (28)	2.91	0.084	2.9

TABLE 2

RFLP Results: 17 Laboratories

PI -A 95 (Locus/Enz: D4s139/Hae III)

Band	Size (Kb)*	SD (Kb)	CV (%)
Mother	4.89	0.043	0.88
AF	7.66	0.104	1.36
Mother	7.81	0.147	1.88
M/Ch	7.83	0.132	1.68
AF/Ch	8.6	0.163	1.9
AF	8.61	0.159	1.84

*Mean

TABLE 3

Reported Paternity Index: PI -A95

Marker	Enzyme (*)	Low	High
D2S44	Hae III (21)	2.53	11.36
	Hinf1 (4)	4.25	6.43
D4S163	*Pst1 (7)*	16.67	34.94
D7S22	**Hinf1 (5)**	4.83	8.65
D10S28	Hae III (17)	7.94	80.83
D12S11	**Hinf1 (6)**	12.11	21.84
	Pst1 (6)	10.84	28.8

*** Number of participants**