

A Protocol for Reporting Single Indirect Exclusions.

H. F. Polesky and J. M. Souhrada

Memorial Blood Center of Minneapolis, Minneapolis, Minnesota,  
55404, U.S.A.

It is difficult to reach a conclusion with regard to paternity when the only inconsistency is reverse homozygosity in one system. The finding of a "single indirect exclusion" often occurs when a child and parent have a common null allele. On the other hand, in some cases this observation establishes non parentage. This report will present a protocol for reporting and resolving the question of parentage in such cases.

In testing 23,000 trios of disputed parentage we have observed exclusions of the alleged father in 30% of cases after routine testing (red cell antigens, serum proteins, red cell enzymes with CEP  $\geq .95$ ). In a small percent of cases only a single indirect exclusion was found. In such cases a report is prepared which indicates the following:

- 1) the results do not provide sufficient information to establish whether or not the tested man may or may not be the biologic father.
- 2) an explanation is given of the results in the discrepant system.
- 3) a recommendation to test additional genetic systems including HLA is made.
- 4) Neither the Paternity Index (PI) or likelihood value is reported.

Table one summarizes 115 cases in which additional testing has been done. In 67 cases the additional testing (HLA) excluded the tested man. The original residual PI was less than 50 in 77% of excluded cases. This table indicates that the residual PI (PI calculated after assigning the excluding system a value of 1) is helpful in predicting whether additional testing will provide exclusionary evidence.

If the additional testing (CEP  $\geq .99$ ) fails to exclude a second report is prepared which includes a set of calculations based on the following assumptions:

- 1) the index for the discrepant system equals 1 (residual PI).

- 2) an exclusion is present (index = 0).
- 3) the child and tested man have a "null" allele.
- 4) the alleged father is a random man who may be either homozygous or has the "null" allele. ( $r/q+2r$  where  $r$  is frequency of null and  $q$  is frequency of the normal allele).

Table two shows by system the number of cases observed and excluded by additional testing and the gene frequency of silent alleles we use for our calculations. It is of interest that in all systems except three [FY (6.7%), PGMi (28.6%), and PLG (46.7%)], HLA establishes non paternity for 60% or more of the tested men. This probably reflects the greater frequency of nulls in these systems.

Depending on whether the PI calculated by method # 4 is above or below 20 various conclusions are provided. If the PI value  $\geq 20$  the report included the following:

- 1) Failure to exclude in all other genetic systems tested and calculations made considering various options with regard to the excluding system indicated almost with certainty that the tested man is the biologic father of the child in question.
- 2) Evidence of sexual intercourse between the mother of the child in question and the tested man at a time proximate to the conception of the child in question and ruling out sexual contact between the mother of the child in question and any first degree relative of the tested man could further strengthen the possibility that the tested man is the biologic father of the child in question.

In cases in which the calculated value is less than 20 an additional statement is added to the above;

- 3) To prove with certainty whether the tested man has the rare null gene in the "excluding" system, family members would need to be tested. This may include parents, siblings, and any other children of the tested man.

This approach appears to be a reasonable solution to providing scientific data to the court in difficult cases. It also provides a basis for decisions on whether to pursue family studies.

**Table 1**

**PI Calculated Without  
Single Indirect Exclusion  
Prior To HLA**

Residual Index	Tested #	HLA Excludes #	%
≤ 10	38	30	78.9
10-20	18	14	77.8
20-50	17	8	47.1
50-100	5	3	60.0
> 100	37	12	32.4
<b>Total</b>	<b>115</b>	<b>67</b>	<b>58.3</b>

**Table 2**

**Results of Additional Testing  
In Cases With A  
Single Indirect Exclusion**

System	# Cases	# HLA Excludes	Null Frequency
GC	5	5	0.000
F13A	2	2	0.001
BF	7	6	0.0009
HP	7	6	0.001
ACP	9	7	0.002
GLO	8	6	0.001
ESD	3	2	0.0007
GM	3	2	0.001
JK	13	8	0.001
RH	8	5	0.0007
F13B	2	1	0.001
MNSs	10	6	0.001
PI	1	1	0.0001
PLG	15	7	0.0035
PGM1	7	2	0.001
FY	15	1	0.005