CHROMOSOME POLYMORPHISMS IN LEGAL PATERNITY CASES.

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Examination for chromosome variants has been used alongside with HLA typing in unsolved paternity cases in Denmark since 1977. Preliminary results have been presented in 1977, 1979 and 1981.

Cultures of lymfocytes obtained from blood samples are used for the preparation of metaphase chromosomes from the parties. The preparations are stained with quinacrine mustard. By use of an UV photomicroscope (Zeiss III) and Agfachrom positive color film (CT 18) at least five color slides of the preparations are produced for each person. The use of color slides makes it possible to recognize the brilliancy of intensely stained chromosomal markers.

The polymorphism of chromosome 3 and 4 concerns the brilliant juxtacentromeric band q 11 which may occur on the long arm of these chromosomes. They are classified as N, S, M, L, vL, and INV.

N denotes a chromosome without a brilliant q 11 band. S denotes a chromosome for which the size of the brilliant q 11 band is smaller than that of the band q 31 on the long arm of chromosome 1. M denotes a chromosome for which the size of the brilliant q 11 band is bigger than that of the q 31 band on chromosome 1 but smaller than that of the band q 31 on the long arm of chromosome 7. L denotes a chromosome for which the size of the brilliant q 11 band is bigger than that of the q 31 band on chromosome 7 but smaller than the short arm of chromosome 16 and vL denotes a chromosome for which the size of the brilliant q 11 band is bigger than the short arm of chromosome 16. INV stands for partial inversion.

The polymorphism of the chromosomes 13, 14, 15, 21, and 22 concerns the intensely stained or brilliant satellite and also the brilliant band p 11 which may occur on the short arm of these chromosomes.

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The size of the satellite as well as the size of the brilliant p 11 band is compared with that of the q 31 band on chromosome 1 and chromosome 7 and with the short arm of chromosome 16 in order to classify the chromosome variants as NN, NS, NM, SN, SM, MN, MS, MM, LN, or vLM. The symbol for the satellite is placed in front of the symbol for the p 11 band. For example SM denotes a chromosome with an intensely stained or brilliant satellite which is smaller than the q 31 band on chromosome 1 and a brilliant p 11 band which is bigger than the q 31 band on chromosome 1 but smaller than the q 31 band on chromosome 7.

The polymorphism of chromosome 1 and 9 concerns the size of the juxtacentromeric band qh which by staining with quinacrine mustard remains without any fluorescense and therefore occurs as a secondary constriction. The variants are classified as SM, L, and vL. SM denotes a chromosome on which the secondary constriction is shorter than the short arm of chromosome 16. L denotes a chromosome on which it is longer than the short arm of chromosome 16 but shorter than the short arm of chromosome 9, and vL denotes a chromosome on which it is longer than the short arm of chromosome 9.

The polymorphism of the Y choromosome concerns the size of the fluorescent distal part of the long arm and is classified as O, S, ML, or vL. O denotes an Y chromosome without a fluorescent band on the long arm. S denotes an Y chromosome which is smaller than chormosome 21 (only small fluorescent spots), ML denotes an Y chromosome which is bigger than chromosome 21 but smaller than chromosome 16, and vL denotes an Y chromosome which is bigger than chromosome 16 (three fluorescent bands).

The frequency of the different chromosome variants in the Danish population was obtained from a material of 1119 unrelated Danish males and females (Niebuhr and Gürtler, 1981). The observed distribution of the different chromosome variants in the kariotypes of the invenstigated 1119 persons was in perfect agreement with the expected Hardy-Weinberg proportions.

Chromosomes which have passed on from parents to their

children are known to maintain their structure. Thus the paternity of non-fathers may be excluded according to one or both of the following two rules:

1) A chromosome variant shall not occur in a child unless it is present in at least one of the parents.

2) A child must have at least one variant of each of the nine heteromorphic autosomes in common with each of the two parents.

The obtained estimate of the frequencies of the different chromosome variants in the Danish population may be used for calculating the expected rate of exclusions of paternity among non-fathers (Gürtler and Niebuhr, 1981) as shown in table 1. The expected rate of exclusions obtained by use of all the heteromorphic chromosomes is o.72 for paternity cases concerning girls and o.74 for cases concerning boys.

Table 2 concerns 37o unsolved Danish paternity cases with 515 men in which examination for chromosome variants have been used alongside with HLA typing. Exclusions of paternity according to the HLA system were obtained for 159 men. The expected rate of exclusions of paternity among non-fathers according to the HLA system during the periode within which the material was collected has been calculated as o.89. Thus the expected number of non-fathers among the 515 men shall be 159/o.89 = 178.65 and the expected number of exclusions of paternity according to chromosome variants shall be 178.65 x o.7333 = 131. The observed number was 125.

In table 3 the observed number of exclusions of paternity obtained by use of variants of the different heteromorphic chromosomes has been compared with the calculated expected number of exclusions. There is a general agreement between the observed and the expected numbers. However for chromosome 13 and chromosome 22 the observed number of exclusions is significantly lower than the expected values.

This deficit refers to cases in which the difference between the child and the mother and/or the alleged father is too small to support an exclusion. A double blindtest including

732 persons has shown rates of discordance between o.3% and 1.7% for the different heteromorphic chromosomes and has clearly demonstrated that in order to avoid mother/child exclusions and false exclusions of paternity only clearcut differences should be considered conclusive (Niebuhr and Gürtler, 1981).

Among the 515 men in 37o paternity cases exclusion of paternity according to the HLA-system and / or chromosome variants was obtained for 172 men and futher 5 men were excluded according to other type systems. 33 cases were left without a possible father, 336 cases were left with only one possible father, and one case was left with two possible fathers, one with a paternity index of 563 and the other with a paternity index of 15.

Any close linkage between serological type systems and chromosome variants has so far not been observed. Paternity indices obtained from serological typeresults and from chromosome variants may therefore be multiplied in order to obtain a combined paternity index which include all the obtained information concerning the paternity of the alleged father.

The distribution of the combined paternity indices obtained for the remaining 338 men is shown in figur 1. A combined paternity index exceeding lo ooo was obtained for 59% of the men, a combined paternity index exceeding 1 ooo for 86% of the men, and a conbined paternity index exceeding loo for 98% of the men.

The distribution of the paternity indices obtained from the serological typeresults including the HLA type results but without the chromosome variants is drown with a dotted line in figur 1. It occurs that the chromosome variants have increased the mean of the obtained paternity indices with a factor lo.

The observed distribution of the combined paternity indices deviates from the expected normal distribution as shown in figur 2. However, as shown in figur 3 the distribution obtained for the 2o8 men from one man cases is in accordance with the expected Normal distribution. The remaining 13o men belongs to

paternity cases in which the ordinary blood- serum, and enzym typing has left more than one possible father. Such cases represents a selection of mother/child type constellations with a low exclusion-possibility and therefore also with a low value of the paternity index for the biological father. This is in agreement with the observed distrbution of the combined paternity indices for the 13o men.

The first exclusion of paternity according to chromosome variants (the Y chromosome) was published by de la Chapelle et al. in 1967. Examination for chromosome variants has proved usefull within different fields of human genetics. It should be emphasized that the hetermorphism described above is strongly simplified. Olson et al. have recommended the use of several staining methods i order to improve the efficiency of the examination for chromosome variants in paternity cases. However, even in the simplified version discribed above chromosome examination has proved usefull in paternity testing.

Refferences:

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THEORETICAL RATE OF EXCLUSIONS.

CHROMOSOME	RATE OF EXCLUSIONS	TOTAL
1	0,0348	0,0348
9	0,0682	0,1006
3	0,2430	0,3191
4	0,0137	0,3284
13	0,2933	0,5254
14	0,1177	0,5813
15	0,1339	0,6373
21	0,1036	0,6749
22	0,1440	0,7217
Y	0,0831	0,7448

TABLE 1.

370 DANISH PATERNITY CASES WITH 515 MEN.

CHROMOSOME VARIANTS	HLA EXCLUDED	HLA NOT EXCLUDED	TOTAL
EXCLUDED	112	13	125
NOT EXCLUDED	47	343 *)	390
TOTAL	159	356	515

HLA RATE OF EXCLUSION: 0,89

EXP. NUMBER OF NON-FATHERS: 159/0,89 = 178,65

EXP. NUMBER OF CHROMOSOME EXCLUSIONS: = 178,65 x 0,7333

= 131

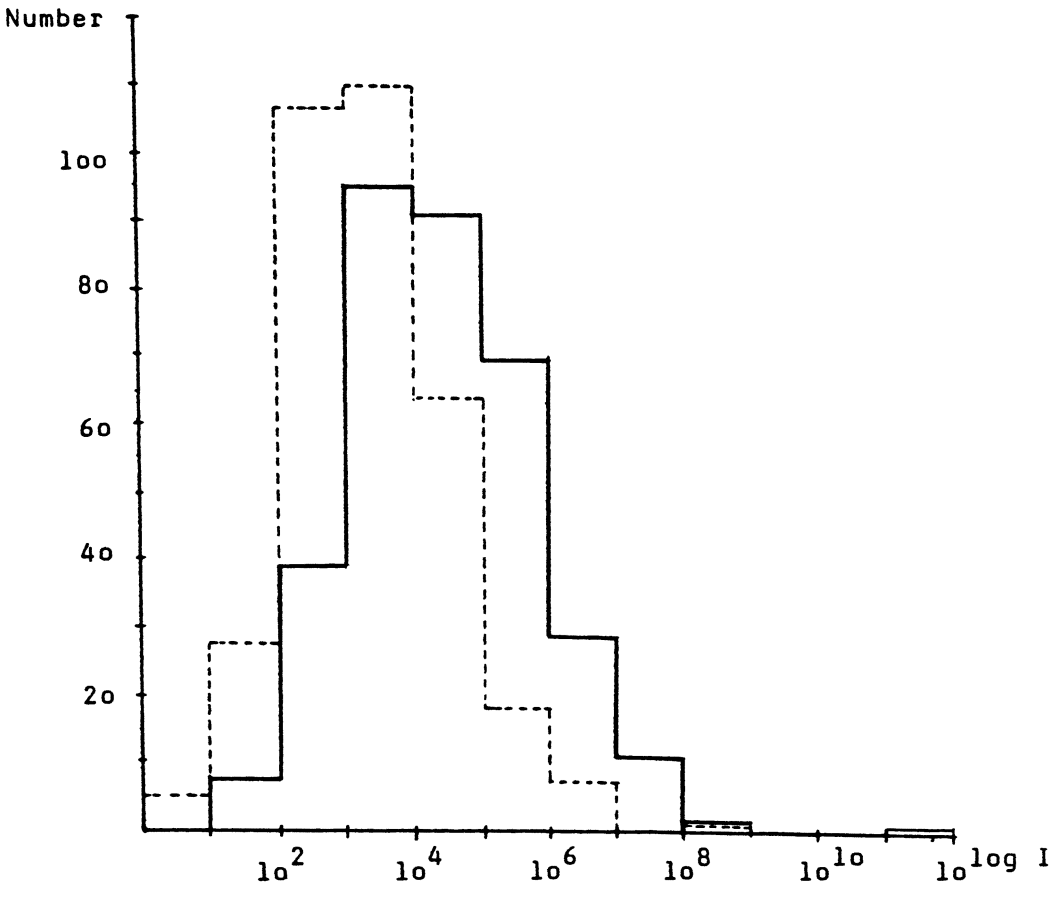
*) 5 EXCLUSIONS ACCORDING TO OTHER TYPE SYSTEMS.

TABLE 2.

NUMBER AF EXCLUSIONS AMONG THE 178 NON-FATHERS.

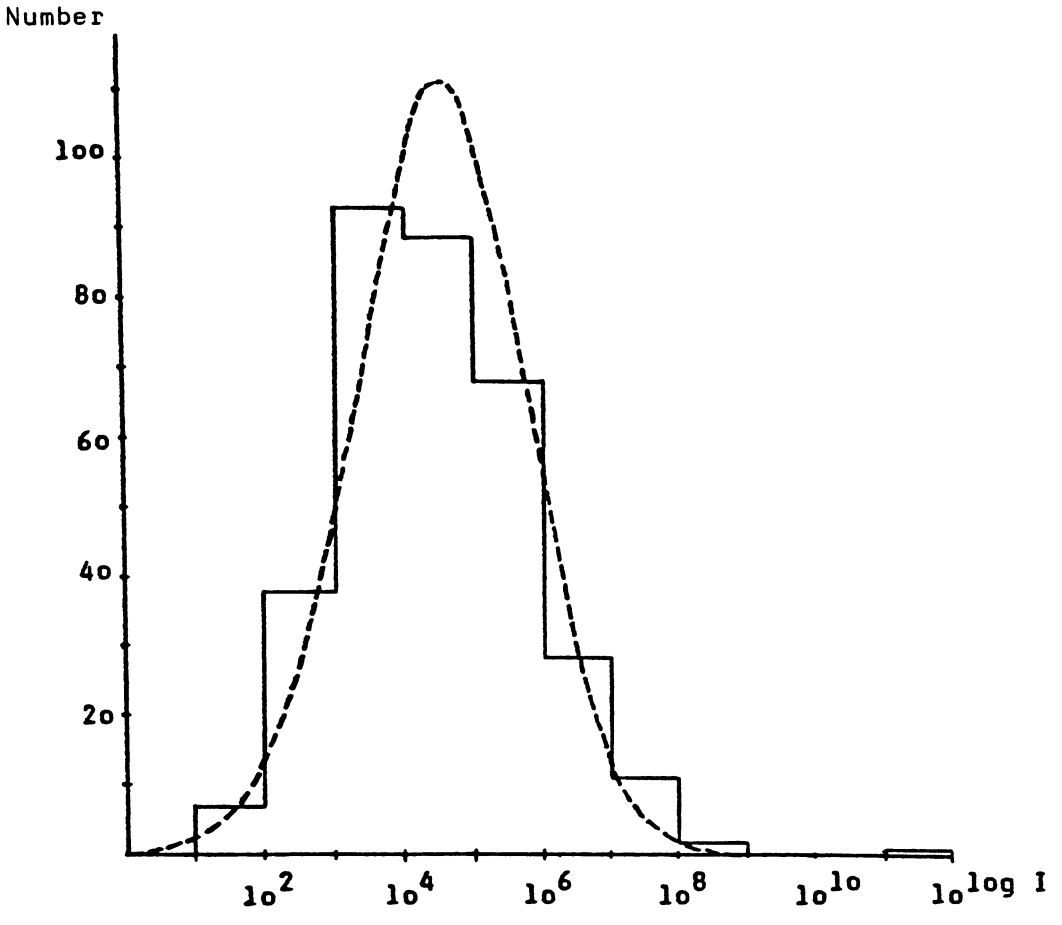
CHROMOSOME	OBSERVED	EXPECTED	χ^2
1	2	6,22	2,97
9	7	12,18	2,36
3	41	43,41	o,17
4	4	2,45	o,99
13	36	52,4o	7,26 *
14	14	21,o3	2,66
15	24	23,92	o,oo
21	15	18,51	o,74
22	15	25,73	5,23 *
Y	11	7,42	1,8o

TABLE 3.



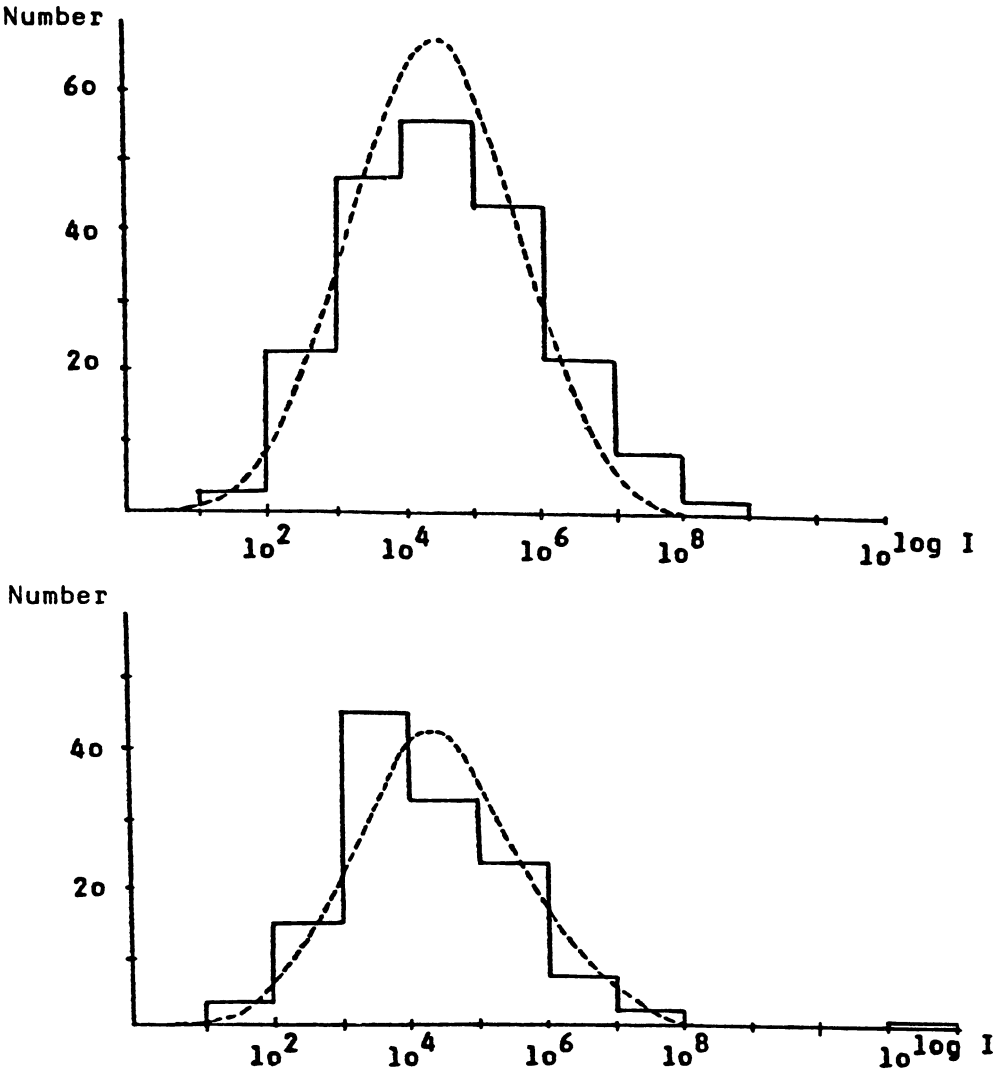
OBSERVED DISTRIBUTION OF THE PATERNITY INDICES AMONG 338 ALLEGED FATHERS BEFORE ''''''''' AND AFTER CHROMOSOME EXAMINATION ————,

FIGUR 1.



OBSERVED AND EXPECTED DISTRIBUTION OF THE PATERNITY INDICES AMONG 338 ALLEGED FATHERS.

FIGUR 2.



OBSERVED AND EXPECTED DISTRIBUTION OF THE PATERNITY INDEX AMONG 208 ALLEGED FATHERS FROM ONE MAN CASES (ABOVE) AND 130 ALLEGED FATHERS FROM CASES INVOLVING MORE THAN ONE MAN (BELOW).

FIGUR 3.

IV. Population Genetics