INDICATION FOR A SILENT ALLELE OF PROPERDIN FACTOR B POLYMORPHISM (BF*Q0) IN A PATERNITY CASE

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INTRODUCTION

The structural gene of factor B (BF), a component of the alternative complement pathway, is located in the class III region of the major histocompatibility complex (MHC) centromeric to the C2 gene and between the C4 and HLA B genes. Alpern *et al* (1972) first reported polymorphism of BF, by using immunofixation agarose gel electrophoresis: they identified two common (F and S) and two less common (F1 and S1 = S0.7) alleles, codominant at the same locus. Subsequently, a number of rare variants have been described and a nomenclature has been proposed (Geserick *et al* 1990). Further isoelectric focusing (IEF) studies revealed the occurrence of BF subtypes (Teng *et al* 1982; Geserick *et al* 1983) which could not be demonstrated by standard electrophoresis.

Until now, only a few families with an apparent silent allele of the factor B polymorphism (BF*Q0) have been published (Weidinger *et al* 1979; Sociu-Foca *et al* 1980; Tokunaga *et al* 1984; Bertrams *et al* 1985 and 1986; Weidinger *et al* 1989; Stanekova *et al* 1993). BF*Q0 allele frequency is estimated close to 0.001 (Polesky *et al* 1983). Recently, hypomorphic gene products of assumed BF*Q0 alleles were detected by means of advanced methods of IEF (Siemens *et al* 1992). In this paper we present further indication for a silent BF allele, in a case of disputed paternity.

MATERIALS AND METHODS

Blood samples were drawn from four apparently healthy individuals (mother [M], child [C] and two alleged fathers [AF1 and AF2, separate husband and current partner of M, respectively]) for paternity testing, after a private request.

The subjects were investigated - according to the protocol used in 1992 in our laboratory - in the following genetic systems: AB0, RH, MNS, HP, IGHG, IGK, GC, TF, PI, BF, ACP, PGM1, AK, ADA, GPT, GLO, ESD, DQA1, D1S80. BF typing was carried out by immunofixation with a monospecific BF antiserum (New Scientific Company) immediately after agarose gel electrophoresis, as described by Domenici *et al* (1986). Several electrophoretic runs were performed on the same blood samples, in order to confirm the results.

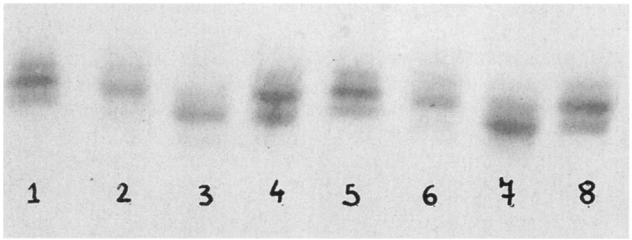
As new DNA polymorphisms were added to our protocol - after 1992 - also YNZ22, APOB, TH01, VWA and FES systems were determined on previously extracted and stored DNA.

RESULTS AND DISCUSSION

The first man [AF1] was excluded as the father of the child [C] in MNS, HP, PI and GLO systems. Only a single indirect exclusion in the BF polymorphism was found in the case of the second man [AF2]. AF2 was BF S, C was BF F (Fig. 1). Biostatistical evaluation of combined data (BF + other 18 systems) yielded, for AF2, a paternity probability of W = 96.73% (Fig. 2).

A request of new blood sample was rejected by mother and AF2, because the probability of paternity was "high enough" for them. So it was not possible to perform the second step of

paternity analysis, including HLA system. Immunochemical and functional levels of factor B were not determined either.



Legend:
1: AF1 = F
2: C = F
3: AF2 = S
4: M = FS
5: AF1 = F
6: C = F
7: AF2 = S
8: M =FS

Fig. 1: BF allotypes in the investigated family, after agarose gel electrophoresis and specific immuno-fixation.

After the study of five "new" DNA polymorphisms, the paternity of AF1 was excluded also in YNZ22, TH01 and VWA systems. On the other hand, the probability of paternity of AF2 raised to W = 99.995% (Fig 2). It is, therefore, very probable that he is the real father.

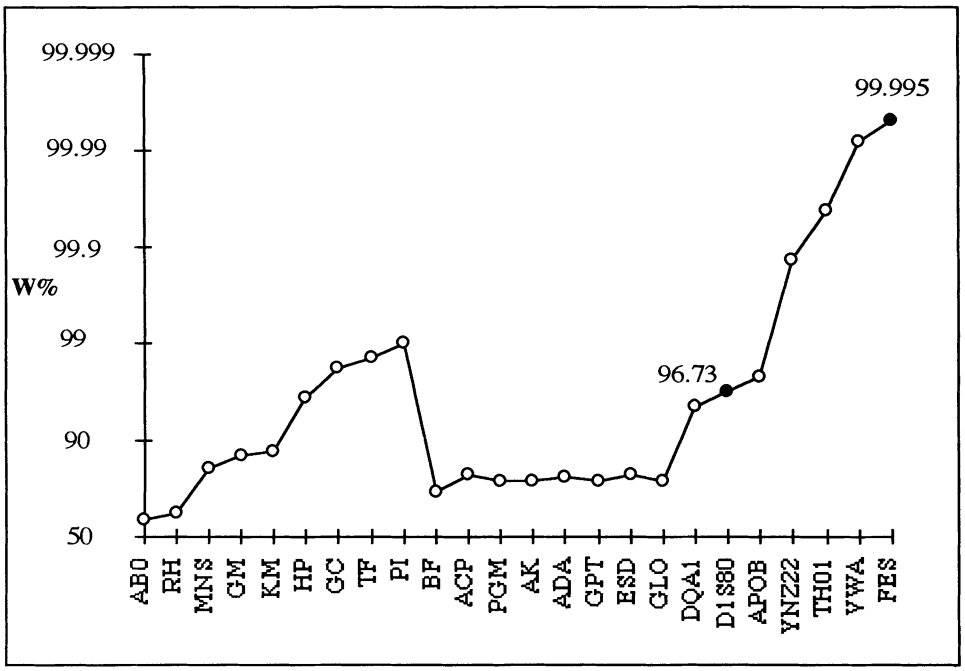


Fig. 2: Cumulative probability of paternity (W) of AF2 (logarithmic scale)

Our findings suggest that the inverse homozygosity between AF2 and C could be due to an (apparently) non-expressed BF allele.

Direct evidence on the nature of abnormal allele was not achieved. In this connection, quantitative analysis are believed to be useful, even if lowered plasma levels of factor B protein are not a reliable criterion, since factor B levels may vary considerably, due to its function as

acute phase reactant (Bertrams *et al* 1985). Recently, the application of advanced methods of IEF for the determination of BF F subtypes showed that different hypomorphic BF products (BF QL = quantitatively low), with functional haemolytic activity, were expressed by assumed BF*Q0 alleles (Siemens *et al* 1992).

When a single indirect exclusion occurs in a polymorphic protein system, in case of disputed parentage, more extensive studies are needed: in our opinion biostatistical evaluation plays an important role, especially if other analyses (quantitative and functional essays, advanced determination methods) or pedigree studies are no practicable. Moreover, the introduction of DNA polymorphisms resulted in a decisive increasing of the power of the biostatistical tool.

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