

Subtyping of coagulation factor XIII_A by Isoelectric focussing

G. Arnold, M.J. Prata*, A. Amorim*, J. Kömpf, H. Ritter

Institut für Anthropologie und Humangenetik der Universität
Tübingen, *Instituto de Antropologia Fac. Ciências Univ. do Porto

Introduction

Human blood coagulation factor XIII consists of 2 dimeric molecules, F XIII_A and F XIII_B which are under separate control of loci on chromosome 1 and 6, respectively. F XIII_A is the zymogen for factor XIII_a bearing transglutaminase activity, which forms intermolecular γ -glutamyl- ξ -lysine cross-links between monomeric fibrin strands, thereby stabilizing the fibrin clot.

A genetic polymorphism of factor XIII_A with two common alleles, F XIII_A* 1,2, was first described by Board (1979).

Suzuki et al. (1988) demonstrated the existence of subtypes of each of this alleles, F XIII_A* 1A, 1B, 2A, 2B, by isoelectric focusing.

Material and Methods

EDTA-Plasma samples and diluted crude white cell lysates from 184 families with 513 children from SW Germany and 201 unrelated individuals from North-Portugal were analyzed. Separation was by IEF in a pH gradient from 5-8 with 3 M urea.

(Acrylamidegel-total volume:15 ml, 2,5 ml acrylamide, 1,2 ml bis-acrylamide, 0,8 ml ampholyte 5-6.5, 0,2 ml ampholyte 6-8, 1,95 g saccharose, 2,7 g urea).

Visualization was by Enzyme-Immuno-Assay with anti-human-F XIII_A (rabbit, Behring) and anti-rabbit (goat, POD con-jugated, ATAB) after blotting onto nitrocellulose.

Results and discussion

Fig. 1: Pherogram of F XIII_A subtypes

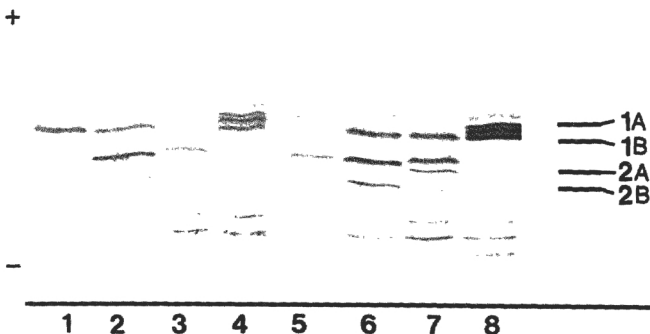


Fig. 1 shows five different phenotypes of F XIII_A
(1) 1B, (2,6) 2B1B, (3,5) 2B1A, (4,8) 1A1B, (7) 2A1B

Fig. 2: Zymogram of Transglutaminases

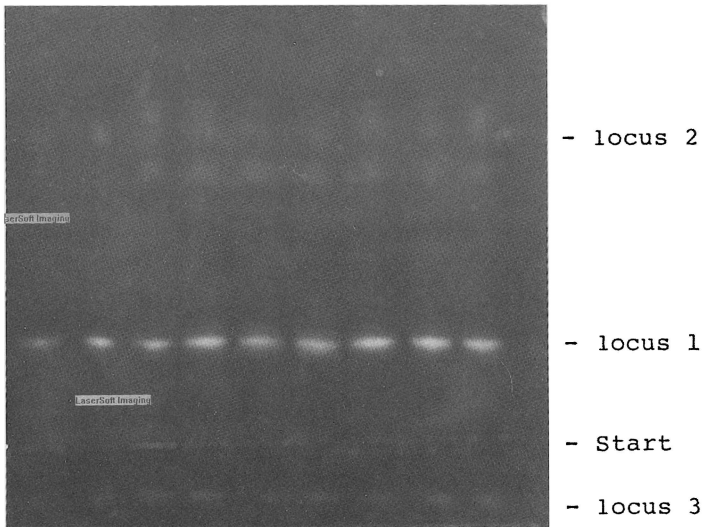


Fig. 2 shows the results after IEF and functional assay (using the transglutaminase activity of activated F XIII_A). There are two more patterns with transglutaminase activity, a result which was first described by Sebetan (1988), he interpreted it as additional loci of F XIII_A.

Formal Genetics

Table 1 shows the segregation in 184 families with 513 children in SW-Germany. Observed and expected segregation ratios are in good agreement: $\chi^2 = 14,14$, $0,7 < P > 0,80$, $df = 11$.

Thus, by this extended formal analysis, the hypothesis proposed by Suzuki et al. (1988) is confirmed:

"Four common alleles F XIII_A*1A, 1B, 2A, 2B at an autosomal locus control the expression of 10 phenotypes"

Table 1:

matings		No. of fam.	phenotypes of children									
			1A	1B	1A1B	2B	2A	2A1A	2A1B	2B1A	2B1B	2A2B
1A	x 1B	2			8							
	x 1A1B	3	3		0							
	x 2B1A	2	4							1		
1B	x 1B	18		63								
	x 1A1B	33		38	49							
	x 2B	9									24	
	x 2B1A	14			14						22	
	x 2B1B	43		51							63	
1A1B	x 2A1A	2			3				3			
	x 1A1B	8	8	8	9							
	x 2B	6							8	11		
	x 2A1A	1	1		1		1	1				
	x 2A1B	4		0	2		2	4				
2B	x 2B1A	6	2		6					4	6	
	x 2B1B	12		10	9					10	6	
	x 2B1A	1				2				0		
	x 2B1B	2				3						
	x 2B1B	1		0					2		3	
2A1B	x 2B1A	1	1			0				2	0	0
	x 2B1B	6			5	5				4	5	
	x 2B1B	10		2		7					17	
Total		184	19	172	106	17		3	10	29	157	

Population Genetics

Table 2: Distribution of F XIII^A phenotypes in samples from SW-Germany and N-Portugal

Phenotypes	SW-Germ. observed	exp.	N-Port. observed	exp.
1A	5	7,36	7	7,96
1B	95	96,31	73	65,23
2A	0	0,03*	3	0,72*
2B	14	12,09	7	5,75
1A1B	56	53,26	39	45,57
2A1A	1	0,97*	7	4,78
2B1A	20	18,87	20	13,53
2A1B	6	3,51*	11	13,67
2B1B	65	68,25	33	38,74
2A2B	0	1,24*	0	4,06*
4-1B	1	0,64*		
4-2B			1	0,17*
0-1B	1	0,64*		
others	0	0,83	0	0,82*
total	264		201	
chi ²		1,61		8,30
P (3 df)		>0,30		>0,95
		<0,40		<0,975

*pooled for chi² calculation

Table 3:

Gene frequencies	SW-Germany	Portugal
F XIII ^A *1A	0,167	0,199
1B	0,604	0,570
2A	0,011	0,060
2B	0,214	0,169
4	0,002	0,002
0	0,002	

Table 3 shows the distribution of F XIII^A alleles in a sample of 260 unrelated individuals from SW-Germany and of 201 unrelated individuals from North-Portugal.

References

- Board PG. (1979): Genetic polymorphism of the subunit of human coagulation factor XIII. *Hu. Geneti.* 31:116-124
- Suzuki K., Matsui K., Ito S., Fujita K., Matsumoto H. (1988): Polymorphism of the A subunit of coagulation factor XIII: Evidence for subtypes of the F XIII^A*1 and FXIII^A* alleles. *Hum. Genet.* 43: 170-174
- Sebetan IM., (1988): A new method for F XIII^A genetic variants determination using isoelectric focusing in 1 M urea, evidence for three structural gene loci, F XIII^A1, F XIII^A2 and F XIII^A3.