Int J Legal Med (1998) 111 : 97–100

TECHNICAL NOTE

H. R. Schneider · S. Rand · H. Schmitter
G. Weichhold

ACTBP2-nomenclature recommendations of GEDNAP

Received: 18 June 1997 / Received in revised form: 27 August 1997

Abstract A system of nomenclature is proposed for the complex STR system ACTBP2 (SE33) in order to facilitate data exchange between laboratories.The nomenclature conforms to the ISFH recommendations as far as it is possible for such complex systems. A blind trial was carried out between up to 20 laboratories to ascertain the reproducibility of the nomenclature under working conditions. The population studies carried out have established that there are minimal regional differences in the allele frequencies and that the system of nomenclature is robust.

Key words ACTBP2 · SE33 · Nomenclature · Population studies · Blind trial

Introduction

The need for consensus nomenclatures for STR systems currently in use in European forensic laboratories is becoming more and more apparent. The experience gained from recent blind trial studies (Gill et al. 1994; Kimpton et al. 1995; Wiegand et al. 1995; Anderson et al. 1996) and the increasing number of "across the border" cases, underlines the need for a rapid and accurate exchange of information between European forensic laboratories. The first phase of European harmonisation is the selection of suitable core systems (STRs) for the investigation of forensic casework. The second phase is to agree on a consensus nomenclature for the exchange of data and comparison of results. For many systems this has already been achieved at the European level (Gill et al. 1994; Anderson et al. 1996) even for complex systems such as D21S11 and HUMFIBRA (FGA) (Barber et al. 1996; Gill et al. 1997a). However, for the complex but highly informative systems such as ACTBP2 (Moos and Gallwitz 1983; Polymeropoulos et al. 1992; Urquhart et al. 1993) several different nomenclatures have been proposed based on sequencing data (Müller and Brinkmann 1994), the number of repeats (Möller et al. 1995) or the total number of base pairs (Schmitter and Sonntag 1995). This situation is made more complex by the use of different separation systems such as native gels (Möller and Brinkmann 1994) and denaturing gels (Möller et al. 1995) whereby alleles are classified by mobility and fragment length respectively.

The aim of this report was to propose a consensus nomenclature for the system ACTBP2 based on a sequenced ladder (Möller et al. 1995) which is in accordance with the ISFH recommendations. The robustness of the nomenclature was tested in a blind trial involving laboratories from the GEDNAP group and by population studies from four different regions of Germany.

Materials and methods

A total of 13 samples of blood, saliva, seminal stains, and hairs were sent to participating laboratories within the GEDNAP blind trial survey for analysis. For the population studies, blood samples from unrelated individuals from four regions of Germany were analysed. These consisted of 562 from the Institute of legal medicine Munich (ILM-M), 416 from the Bundeskriminalamt Wiesbaden (BKA), 599 from the Institute of legal medicine Münster (ILM-MS) and 529 from the Landeskriminalamt Hessen (HLKA). DNA extraction and amplification were performed as previously described (Wiegand et al. 1993). DNA typing was conducted according to the method published by Kimpton et al.(1994) using the automated fluorescent detection system ABI 310, 373 or 377 (ABI), or the ALF system (Pharmacia). The sequenced allelic ladder for the system ACTBP2 was kindly donated by Dr. A. Junge (Institute of legal medicine Bonn).

H. R. Schneider (✉)
Hessisches Landeskriminalamt, Hölderlinstrasse 5,
D-65187 Wiesbaden, Germany
Fax: +49 (611) 832 705

S. Rand
Institut für Rechtsmedizin,
Westfälische Wilhelms-Universität Münster,
Von-Esmarch-Strasse 62, D-48129 Münster, Germany

H. Schmitter
Bundeskriminalamt, Thaerstrasse 11,
D-65193 Wiesbaden, Germany

G. Weichhold
Institut für Rechtsmedizin der Universität München,
Frauenlobstrasse 7a, D-80337 München, Germany

Results and discussion

Among the STRs currently in use in forensic laboratories, the system ACTBP2 (SE33) is one of the most informative. It was recently reported that more than 100 alleles have been observed due to sequence variations (Rolf et al. 1997). But this high degree of polymorphism leads to considerable problems for the designation of alleles. Sequencing of the alleles is technically correct but is too time consuming and costly for routine practical application. The nomenclature proposed by Schmitter and Sonntag (1995) is unwieldy and does not accurately reflect the number of repeats as recommended by the ISFH (1994). The nomenclature of Kratzer et al. (1994) does not consider information on the allele sequences. The nomenclature proposed by Möller et al. (1995) while accurately reflecting the repeat structure, appears not to have found favour with the forensic community due to its relative complexity. It was therefore proposed by a satellite group of GEDNAP in-

Table 1 Proposed nomenclatures for ACTBP2 (*n.s.* not sequenced)

ACTBP2-Nomenclature					
ILM-Münster Möller et al. 1994/95	# bases	BKA Schmitter and Sonntag, 1995	ILM-Zürich Kratzer et al. 1994	EDNAP Gill et al. 1997b	Consensus
12	233	48	–1	# 233	12
13 (–2)	235	48.2	–1.2	#235	12.2
14 (–2)	239	49.2	1.2	#239	13.2
14	241	50	2	#241	14
15	245	51	3	#245	15
16	249	52	4	#249	16
17	253	53	5	#253	17
18	257	54	6	#257	18
19	261	55	7	#261	19
20	265	56	8	#265	20
21	269	57	9	#269	21
22	275	58.2	11	#275	22.2
23	279	59.2	12	#279	23.2
24	283	60.2	13	#283	24.2
25	287	61.2	14	#287	25.2
26	291	62.2	15	#291	26.2
27	295	63.2	16	#295	27.2
28	299	64.2	17	#299	28.2
29	303	65.2	18	#303	29.2
30	307	66.2	19	#307	30.2
31	n.s.	67.2	20	#311	31.2
32	n.s.	68.2	21	#315	32.2
34 (–4)	319	69.2	22	#319	33.2

Table 2 Results of ACTBP2 typing in two GEDNAP blind trials (*Uk* Unknown individual)

GEDNAP XII	Person A	Person B		Stain 1	Stain 2	Stain 3	Stain 4
Kind of stain	Blood	Blood		Mixed blood stain	Mixed blood stain	Mixed blood stain	Saliva
Mixing ratio	./.	./.		1:4 (A:B)	1:9 (A:Uk1)	1:9 (B:Uk2)	./.
Typing results	14, 17	21, 30.2		14, 17, 21, 30.2	14, 17, 21, 25.2	21, 23.2, 26.2, 30.2	23.2, 26.2
n (Labs)	17	17		17	17	17	17
GENAP XIII	Person A	Person B	Person C	Stain 1	Stain 2	Stain 3	Stain 4
Kind of stain	Blood	Blood	Blood	Mixed blood stain	Diluted blood stain	Seminal stain	Hair root telogenic phase
Mixing ratio	./.	./.		4:1 (A:C)	1:4 (C:Aqua)	./.	./.
Typing results	17, 28.2	28.2, 29.2	15, 27.2	15, 17, 27.2, 28.2	15, 27.2	23.2, 27.2	28.2, 29.2
n (Labs)	14	14	14	14	14	14	14

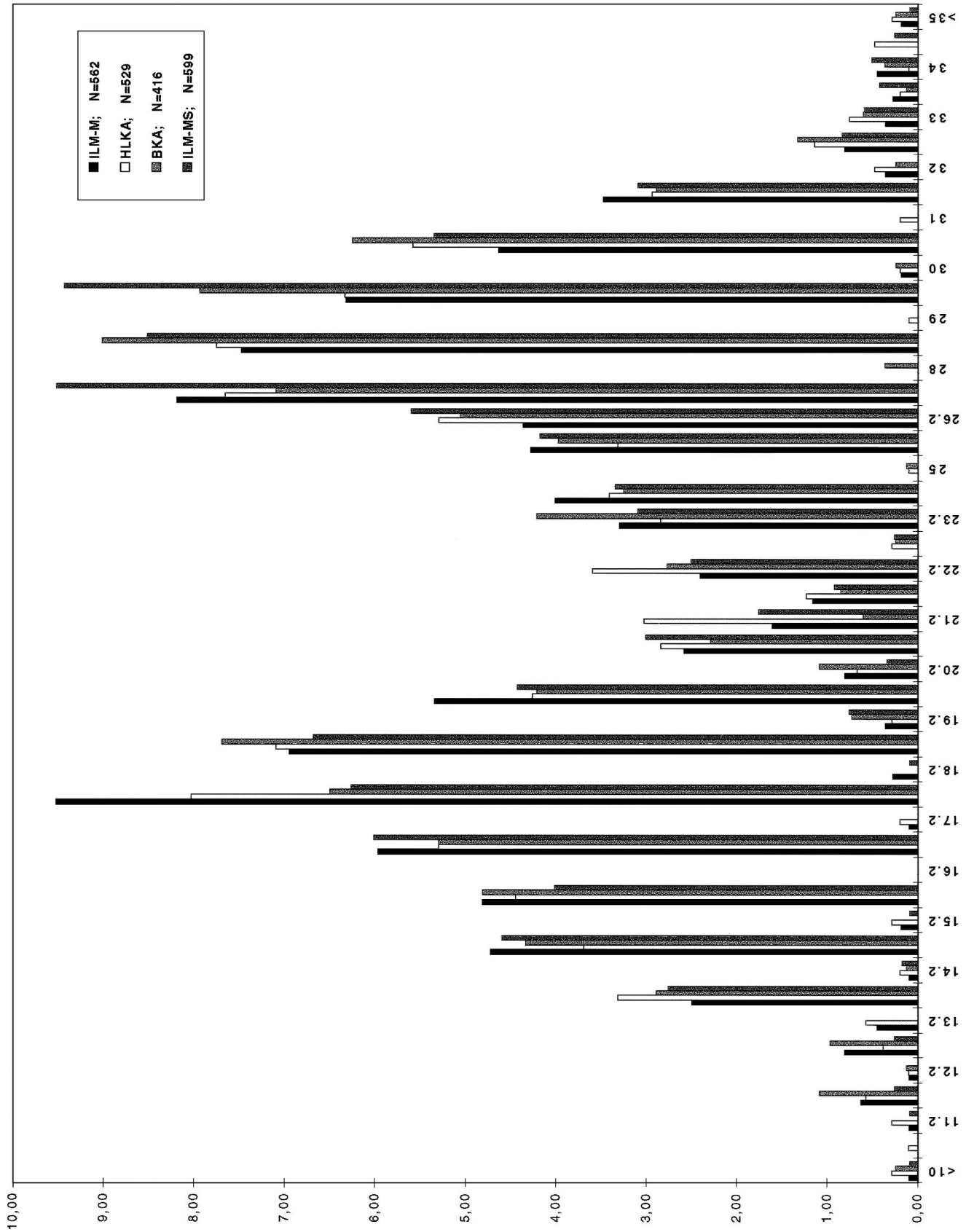

Fig. 1 Allele frequencies for ACTBP2 from four population studies in Germany

volving more than 20 laboratories that a consensus nomenclature should be based on the defined and sequenced ladder developed by Möller et al. (1995). For simplicity this nomenclature is operationally based on the number of repeats within the defined variable region and is applicable only for typing carried out under denaturing conditions. The alleles included in the ladder are regularly spaced at 4 bp intervals from alleles 7–21 corresponding to the number of repeat units. The larger alleles contain a hexamer motif AAAAAG and are designated by a suffix (.2) to indicate the presence of an incomplete repeat unit. The position of this hexamer is variable and some have transitions resulting in a AGAAAG motif. This does not affect the electrophoretic mobility under denaturing conditions but causes considerable problems in native gel systems. Some alleles larger than 317 bp (i.e. 33.2) have been found which contain a second hexamer motif (Rolf et al. 1997) and these are designated by a whole number (e.g. 33) because the two incomplete motifs can be considered as one complete unit. Alleles which fall between ladder fragments should be sequenced but these are rare and can be assigned temporarily to the next adjacent allele.

This system of nomenclature is as simple as possible and is considered to offer the most practical solution because it complies with international recommendations as far as possible and will be acceptable to the forensic community. Each laboratory can still use a local nomenclature internally to avoid problems with established data bases, but this system should be used for interlaboratory exchange of data. For comparative purposes a list of previously published systems of nomenclature is given in Table 1.

To demonstrate the robustness of the consensus nomenclature a group of 17 laboratories participated in a collaborative exercise within the biannual GEDNAP blind trial. Despite the variety of protocols employed, all laboratories obtained the same results and all alleles were correctly typed (N = 530) using the proposed nomenclature and a sequenced standard allelic ladder (Table 2).

A comparison of the four population samples from Germany (Fig. 1) demonstrates that the use of the consensus nomenclature leads to inter- and intralaboratory reproducibility with regards to typing and frequencies. However it must be pointed out that the use of the standard sequenced ladder and separation under denaturing conditions are essential for comparative purposes.

This system has been adopted by the members of GEDNAP for exchange of data and is now routinely included in blind trials.

References

1. Anderson J, Martin P, Carracedo A, Dobosz M, Eriksen B, Johnsson V, Kimpton C, Kloosterman A, Konialis C, Kratzer A, Phillips P, Mevåg B, Pfitzinger H, Rand S, Rosen B, Schmitter H, Schneider P, Vide M (1996) Report on the third EDNAP collaborative STR exercise. Forensic Sci Int 78 : 83–93
2. Barber MD, McKeown BJ, Parkin BH (1996) Structural variation in the alleles of a short tandem repeat system at the human alpha fibrinogen locus. Int J Legal Med 108 : 180–185
3. DNA recommendations-1994 report concerning further recommendations of the ISFH regarding PCR-based polymorphisms in STR (short tandem repeat) systems. Int J Legal Med 107 : 159–160
4. Gill P, Kimpton C, D'Aloja E, Andersen JF, Bär W, Brinkmann B, Holgersson S, Johnsson V, Kloosterman AD, Lareu MV, Nellermann L, Pfitzinger H, Phillips CP, Schmitter H, Schneider PM, Sternersen M (1994) Report of the European DNA profiling group towards standardisation of short tandem repeat (STR) loci. Forensic Sci Int 65 : 51–59
5. Gill P, D'Aloja E, Anderson J, Dupuy B, Jangblut M, Johnsson V, Kloosterman AD, Kratzer A, Lareu MV, Meldegaard M, Phillips C, Pfitzinger H, Rand S, Sabatier M, Scheithauer R, Schmitter H, Schneider P, Vide MC (1997a) Report of the European DNA profiling group (EDNAP): an investigation of the complex STR loci D21S11 and HUMFIBRA (FGA). Forensic Sci Int 86 : 25–33
6. Gill P, Brinkmann B, D'Aloja E, Anderson J, Bär W, Carracedo A, Dupuy B, Eriksen B, Jangblad M, Johnsson V, Kloosterman AD, Lincoln P, Morling N, Rand S, Sabatier M, Scheithauer R, Schneider P, Vide MC (1997b) Considerations from the European DNA profiling group (EDNAP) concerning STR nomenclature. Forensic Sci Int (in press)
7. Kimpton C, Fisher D, Watson S, Adams M, Urquhart A, Lygo J, Gill P (1994) Evaluation of an automated DNA profiling system employing multiplex amplification of four tetrameric STR loci. Int J Legal Med 106 : 302-311
8. Kimpton C, Gill P, D'Aloja E, Andersen JF, Bär W, Holgersson S, Jacobsen S, Johnsson V, Kloosterman AD, Lareu MV, Nellermann L, Pfitzinger H, Phillips CP, Rand S, Schmitter H, Schneider PM, Sternersen M, Vide MC (1995) Report on the second EDNAP collaborative STR exercise. Forensic Sci Int 71 : 137–152
9. Kratzer A, Gränacher A, Jamnicki M, Bär W (1994) Swiss population data for 3 STR-sysems (SE33, HumTH01, D21S11), HLADQa und D1S80. In: Bär W, Fiori A, Rossi U (eds) Advances in forensic haemogenetics 5. Springer, Berlin Heidelberg New York, pp 515–517
10. Möller A, Brinkmann B (1994) Locus ACTBP2(SE33). Sequencing data reveals considerable polymorphism. Int J Legal Med 106 : 262–267
11. Möller A, Schürenkamp M, Brinkmann B (1995) Evaluation of an ACTBP2 ladder composed of 26 sequenced alleles. Int J Legal Med 108 : 75–78
12. Moos M, Gallwitz D (1983) Structure of two human β-actin-related processed genes one of which is located next to a simple repetitive sequence. EMBO 2 : 757–761
13. Polymeropoulos MH, Rath DS, Xiao H, Merril CR (1992) Tetranucleotide repeat polymorphism at the human beta-actin related pseudogene H-beta-Ac-psi-2 (ACTBP2). Nucleic Acids Res 20 : 1432
14. Rolf B, Schürenkamp M, Junge A, Brinkmann B (1997) Sequence polymorphism at the tetranucleotide repeat of the human beta-actin related pseudogene H-beta-Ac-psi-2 (ACTBP2) locus. Int J Legal Med 110 : 69–72
15. Schmitter H, Sonntag M-L (1995) Proposal for general rules for the denominations of alleles of STR-loci based on the electrophoretic mobilities of their fragments. Klin Lab 41 : 173–176
16. Urquhart A, Kimpton CP, Gill P (1993) Sequence variability of the tetranucleotide repeat of the human beta-actin related pseudogene H-beta-Ac-psi-2 (ACTBP2) locus. Hum Genet 92 : 637–638
17. Wiegand P, Budowle B, Rand S, Brinkmann B (1993) Forensic validation of the STR systems SE33 and TC11. Int J Legal Med 105 : 315–320
18. Wiegand P, Ambach E, Augustin C, Bratzke H, Cremer U, Edelmann J, Eriksen B, Germann U, Haas H, Henke L, Holtz J, Keil W, Kreike J, Nagy M, Prinz M, Rand S, Rothämel T, Scheithauer R, Schneider HR, Schürenkamp M, Teifel-Greding J, Bär W (1995) GEDNAP IV and V. The 4th and 5th stain blind trials using DNA technology. Int J Legal Med 108 : 79–84