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SE33 (HumACTBP2): native gel electrophoresis versus denaturing capillary electrophoresis, and population data

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Abstract

The aim of the study was to compare results obtained by electrophoresis under denaturing conditions with results of native horizontal gel electrophoresis and to investigate additional population data on the STR locus SE33 (HumACTBP2) in an Austrian Caucasoid population sample. The results of the comparative study showed that a conversion of results between both typing strategies is not feasible for the entire size range of this locus.
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1. Introduction

The STR locus SE33 (also named HumACTBP2) is located in the 5′ flanking region of the human beta-actin related pseudogene H-beta-Ac-psi-2. Due to its highly polymorphic allele distribution and the large number of sequence variations [1], it is a complex and a very informative system. The aim of our study was to determine whether a direct conversion between results obtained by electrophoresis under denaturing conditions and results obtained by native horizontal gel electrophoresis is possible. Furthermore, additional population data on SE33 were also to be investigated in our local population.

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2. Materials and methods

The study encompassed 135 unrelated Austrian Caucasoid individuals from which DNA was extracted from blood or buccal swabs using the salting out procedure, Chelex® 100 extraction or Dynabeads®DNA DIRECT™ system II (Dynal). Typing under native conditions was performed with our former standard method as described by Schwartz et al. [2]. To obtain results under denaturing conditions the commercially available kit genRES® Kit$^{6\text{-FAM}}$ STR System HumACTBP2 (Serac) was applied for PCR according to the manufacturer's instructions. The amplification products were analyzed using POP-4™ on the ABI Prism 310 capillary electrophoresis instrument (Applied Biosystems). Sequencing was carried out using the DNA Sequencing Kit BigDye™ Terminator Cycle Sequencing Ready Reaction (Applied Biosystems).

3. Results and discussion

In our population sample (n=135) a total of 30 different alleles were found when working under denaturing conditions. Allele frequencies and further statistical data are shown in Table 1. In chi-square analysis no deviations from Hardy–Weinberg expectations could be detected (p=0.330). In the smaller allelic size range (≤allele 19=≤261 bp) each allele of the same size typed under denaturing conditions could be unambiguously assigned to a distinct allele typed by native horizontal gel electrophoresis. In the larger allelic size range, a direct association between results obtained by the two methods could no longer be achieved. We assume that the high sequence polymorphism found in alleles

Table 1
SE33: allele frequencies and further statistical data obtained by denaturing electrophoresis (n=135)

Allele designation	Allele frequency	Allele designation	Allele frequency
12	0.007	22.2	0.019
12.2	0.004	23.2	0.033
13	0.007	24.2	0.044
13.2	0.004	25.2	0.052
14	0.026	26.2	0.037
15	0.037	27.2	0.093
16	0.052	28.2	0.041
17	0.074	29.2	0.067
18	0.070	30.2	0.088
19	0.070	31.2	0.037
20	0.052	32.2	0.015
20.2	0.011	34	0.004
21	0.022	34.2	0.007
21.2	0.015	35.2	0.004
22	0.004	36.2	0.004
Rate of heterozygosity: 0.932		Matching probability: 0.013	
Power of exclusion: 0.861		Power of discrimination: 0.987	
Polymorphism information content: 0.95		Typical paternity index: 8.44	

larger than 267 bp [1] could be an explanation for this phenomenon. Therefore, we sequenced five alleles that were typed as the same allele under denaturing conditions and as three different alleles under native conditions. The results confirmed our hypothesis that the alleles were all the same length but showed three different sequence variations. An allele with a novel sequence variant was also discovered which was designated allele 20 under denaturing conditions (265 bp) and showed a T at position 21 of the 5′ flanking region, whereas alleles of the same size known until now show an A at this position.

The results of our study showed that typing results obtained under denaturing and non-denaturing conditions are not comparable for all alleles at this locus. The conversion of results between both strategies is not feasible for the entire size range. This is of great importance regarding the interlaboratory reproducibility and exchange of data and therefore needs to be considered.

References

[1] B. Rolf, M. Schürenkamp, A. Junge, B. Brinkmann, Sequence polymorphism at the tetranucleotide repeat at the human beta-actin related pseudogene H-beta-Ac-psi-2 (ACTBP2) locus, Int. J. Leg. Med. 110 (1997) 69–72.

[2] D.W.M. Schwartz, E.M. Jungl, O.R. Krenek, W.R. Mayr, Typing for STR-loci by electrophoresis on rehydratable polyacrylamide gels: phenotype and allele frequencies of SE33 and TC11 in an Austrian population sample, in: Adv. Forensic Haemogenet. vol. 5, Springer, Berlin, 1993, pp. 581–583.