THE ESS LOCI

Expansion of the European Standard Set of DNA Database Loci—the Current Situation

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Only the inclusion of robust and powerful mini STR loci into the European Standard Set will ensure that difficult casework samples with low amounts of degraded DNA can be fully typed.

INTRODUCTION

National DNA databases have become one of the most efficient tools to provide intelligence about unknown perpetrators in criminal investigations. At present, almost six million DNA profiles from both suspects and convicted offenders are stored in European databases, and more than one million person-to-stain and stain-to-stain hits have been obtained (1). The databases have various organisational structures depending on the national legislative background in each country (2). These regulate inclusion criteria for offender profiles, storage of DNA profiles and reference samples, and record deletion. In addition, each country has selected a defined number of STR loci that must be typed routinely for each sample to be accepted for inclusion in the database. The selection of STR loci was based on experimental data from collaborative exercises [e.g., from the European DNA Profiling (EDNAP) Group], on small multiplex typing kits originally designed by The Forensic Science Service (Birmingham, UK), on commercially available products from the major reagent suppliers, and finally on recommendations by the Interpol DNA working party in 1998 (3). One year later, the DNA working group of the European Network of Forensic Science Institutes (ENFSI) decided on a European Standard Set (ESS), which includes seven loci: TH01, vWA, FGA, D21S11, D3S1358, D8S1179 and D18S51. These loci have been confirmed by a resolution of the European Council in 2001 (4) and now form the core of all national DNA databases in Europe.

Due to the overwhelming success of DNA databases, a political process was initiated by a number of European countries to establish a legal basis for exchanging DNA database profiles between countries in criminal investigations. This led to the Treaty of Prüm, which was signed in 2005 with the purpose of stepping up cross-border cooperation, particularly in combatting terrorism, cross-border crime and illegal migration (5). Subsequently, the ENFSI DNA working group has established recommendations for DNA database management, including criteria for including and deleting DNA profiles, matching rules, and handling of partial profiles. Furthermore, the occurrence of adventitious matches between DNA profiles that have no case-related connection has been addressed in detail (6). When massive exchanges of DNA profiles are undertaken following the implementation of the Treaty of Prüm, the seven ESS loci will not be sufficient because the chance of adventitious matches will no longer be negligible. In addition, each DNA database contains a significant portion of partial profiles with an even higher probability to match randomly.

ENFSI AND EDNAP MEET TO DISCUSS THE EUROPEAN STANDARD SET

The ENFSI and EDNAP groups met in Glasgow in 2005 and discussed extension of the ESS and recommendations for additional European STR systems. Since the ESS loci are typically part of larger multiplexes with 10–15 loci, which are already used in forensic laboratories throughout Europe, it would have been straightforward to choose among these loci. However, at the same meeting, the results of a collaborative exercise carried out by the EDNAP group to examine typing of heavily degraded DNA samples were presented (7). This exercise addresses the fact that many casework samples include only minimal amounts of DNA or DNA that is degraded due to environmental

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exposure. Furthermore different typing approaches, including standard and short amplicon STR systems as well as single nucleotide polymorphism (SNP) typing, were compared. The results clearly demonstrated that both mini STRs and SNPs with amplicon sizes of 150 bp or less were more successful than the currently used STRs with amplicon sizes of up to 400 bp. As a consequence, the following objectives for an extended ESS were defined: i) improve the discrimination power; ii) improve the sensitivity of testing so that smaller amounts of DNA are detected, and iii) improve robustness or the quality of the result (8). Shortly before the meeting, a set of newly discovered mini STR loci that had good potential in fulfilling these objectives was published (9).

THE RECOMMENDED ESS LOCI

A decision was adopted to recommend two sets of mini STRs of three loci each as candidates for new database loci (8,10). In group I, three loci: D10S1248, D22S1045, and D2S441 (9), with amplicon sizes of 70–125 bp, were selected with first priority, and in group II, the established loci: D12S391, D1S1656, and TPOX, with amplicon sizes of up to 180 bp, were selected. For implementation into laboratory practice, we hoped that reagent manufacturers would include at least the very short group I loci into existing multiplexes within a reasonable time period. Alternatively, all the new group I and II loci could be combined into a separate multiplex for use as an adjunct to existing reagents (10). Of course, an extensive validation and testing period is required using experimental reagents before a final recommendation on the new loci can be expected. Keep in mind that, even after this scientific recommendation, the final political decision must be made by the European Council. In the case of the original seven ESS loci, this took place two years after an agreement among the scientists was obtained (4). Therefore, the EDNAP and ENFSI groups have offered to closely collaborate with commercial manufacturers to speed up the validation process (8,10).

It was not expected at the time of the Glasgow meeting that more than three years would pass without having at least one multiplex containing the new loci commercially available, while the Prüm process on international data exchange made significant steps forward. Experimental comparisons between the German and Austrian databases were carried out. These were based on the seven ESS loci only because the additional Austrian loci D2S1338, D16S539 and D19S433 were not part of the German database, and the additional German locus SE33 (ACTBP2) was not part of the Austrian database. Thus a large number of presumably adventitious matches was encountered, of course, due in part to the presence of partial profiles, which have an even lower discrimination power. Similar observations were made following comparisons between the German and Dutch databases (11).

Following these and other observations, an agreement was reached at the 2008 ENFSI meeting in Prague to recommend using all information currently available from STR typing by adding these results to the respective national database. Many laboratories are currently analyzing more loci than required, and these results were not transmitted previously to the national DNA database. In principle, this strategy was already devised in the first recommendation by Gill *et al.* about the new loci: "Given that substantial national DNA databases have already been constructed using divergent multiplexes, it is unrealistic to suggest that laboratories can change by abandoning loci in favour of new ones. Rather, it is proposed that new core loci are decided and then laboratories expand their systems while retaining their existing set of STRs" (8). Nevertheless, the forensic community is hopeful that new typing kits with the recommended loci will be available soon from commercial manufacturers so that validation work can proceed as rapidly as possible. Only the inclusion of robust and powerful mini STR loci into the ESS will ensure that difficult casework samples with low amounts of degraded DNA can be fully typed to avoid unacceptable rates of adventitious matches due to increasing numbers of partial profiles in the databases.

REFERENCES

1. ENFSI DNA Working Group: ENFSI survey on DNA databases in Europe, Sept. 2008, www.enfsi.eu/get_doc.php?uid=227
2. Schneider, P.M. and Martin, P.D. (2001) Criminal DNA databases: The European situation. *Forensic Sci. Int.* **119**, 232–8.
3. Martin, P.D., Schmitter, H. and Schneider, P.M. (2001) A brief history of the formation of DNA databases in forensic science within Europe. *Forensic Sci. Int.* **119**, 225–31.
4. European Council: Resolution of 25 June 2001 on the exchange of DNA analysis results,
5. Council of the European Union: Treaty of Prüm, 27 May 2005, http://register.consilium.europa.eu/pdf/en/05/st10/st10900.en05.pdf
6. ENFSI DNA Working Group: DNA database management review and recommendations, April 2008, http://www.enfsi.eu/get_doc.php?uid=226
7. Dixon, L.A. *et al.* (2006) Analysis of artificially degraded DNA using STRs and SNPs — results of a collaborative European (EDNAP) exercise. *Forensic Sci Int.* **164**, 33–44.
8. Gill, P. *et al.* (2006) The evolution of DNA databases — recommendations for new European STR loci. *Forensic Sci. Int.* **156**, 242–4.
9. Coble, M.D. and Butler, J.M. (2005) Characterization of new mini-STR loci to aid analysis of degraded DNA. *J. Forensic Sci.* **50**, 43–53.
10. Gill, P. *et al.* (2006) New multiplexes for Europe — amendments and clarification of strategic development. *Forensic Sci. Int.* **163**, 155–7.
11. Van der Beek, C. Exchange of DNA profiles by the Treaty of Prüm. Presented at DNA Data Exchange in Europe conference, June 5–6, 2008, http://www.dna-conference.eu/ppt/Van%20der%20Beek.pdf