STRATEGIES IN FORENSIC STAIN ANALYSIS

Hermann Schmitter

Bundeskriminalamt, Wiesbaden, Germany

Introduction

With the projects of creating national DNA databases, the basis of development in forensic science has changed. Masses of samples from individuals have to be analyzed in a short time. Therefore, research units dealing with forensic DNA analysis as well as companies providing reagents have concentrated their interests in creating analytical systems that enable laboratories to produce DNA profiles rapidly and with high throughput. Thus, choices of loci to be integrated in a database profile as well as primer designs were mainly influenced by their suitability in polyplex approaches.

In this development the demands of stain analysis in daily forensic casework played only a marginal role. The aim of this paper is to address these items.

With respect to both the special situation of evidential stains and the circumstances of a given case the conditions are quite different in casework compared to databasing work. Various strategies have to be developed in order to answer the questions of any given case. Even costs and effectiveness have to be considered. The stain analyst has to react to two main conditions he is confronted with in each single case.

Stain situation

Stains collected in connection to crimes always are very inhomogeneous with respect to both quantity and quality.

Due to the high sensitivity of PCR, the quantity of stain material is more or less negligible. Once minute traces have been found, results will be obtained if other conditions are good. On the other hand minute stains raise the question of their relationship to the case. Very critically, the circumstances of collection and shipping have to be considered. The theoretical possibility of sample transfer has to be scrutinized. This problem is very well known in the area of forensic fiber analysis. Preceding any analysis it must be cleared up that the stain in question really is related to the given case and not created by any secondary transfer.

In most cases where problems arise, bad quality of the stain material is the reason.

- Due to environmental conditions before collection but sometimes also by incorrect sampling and shipping, biological materials can be heavily degraded before reaching the laboratory. Thus, degradation is a problem the analyst often has to deal with. To get as much information as possible in degraded stains it can be advantageous to apply loci with small fragments. For some loci we have designed primers very close to the repeat sequences in order to enable analysis even in very degraded materials.

- Not only in rape cases, stains can contain mixtures of cells from more than one person. Here various strategies for the analysis can be chosen. In our experience when working with complicated mixtures where one component dominates the other one, results obtained by monoplexing are more reliable. A special situation is given when a large amount of female cells and only a few male cells are mixed. Here the application of Y-chromosome markers can be a helpful tool.

- In daily casework reality, stains are dirty. Various contaminants may influence the reactions. These problems can in many cases but not always be overcome by choosing special extraction methods and purification steps. Even monoplex approaches can be superior to polyplexes due to a higher tolerance window.

Case situation

In order to do the work with high efficiency and the lowest cost possible, the circumstances of any given case also have to be considered before analyzing the evidence materials. Some examples may show this.

- When there are a limited number of possible donors in a given case the preferred strategy is to profile these individuals. Stains then are screened using monoplex analysis at that locus where the individuals in question differ from each other.

- A frequent example of multiple stains occur in an ashtray full with cigarette butts where the analyst is asked whether a special individual has left some of them in the ashtray. Situations like that often occur in cases of bank robbery when the perpetrators used a stolen car and left it at any place to escape with another one. Often these are typical case where the perpetrators are unknown.

  In cases like that, screening the System SE33 (ACTBP2) is most effective due to the high discrimination power of that locus.

- In many cases the offenders are unknown. In severe crimes of high public interest, investigation may decide on mass screening of great numbers of persons. Even in situations like that, monoplex applications can be superior to polyplex approaches in terms of efficiency and costs as explained by the following example:

A "mass screening" case

In 1998 in a region of the state of Niedersachsen an eleven year old girl had been raped and murdered. The analyses described in the following mainly had been performed in the state laboratory of Niedersachsen in Hannover. Later the state laboratories of Berlin and Sachsen-Anhalt supported the work.

The semen stain collected from the victim was analyzed and it turned out that it matched a semen stain collected two years earlier in another case where in the same area another a young girl had been raped, but fortunately not killed. Based upon these results as well as other circumstances, investigators had the strong suspicion that the perpetrator lived in that area. They even had some indication of the age of the suspect. Therefore, investigators decided to ask all men in the age range between 25 and 45 years living in that region to voluntarily give saliva samples. At least 16,000 individuals fall into this category creating tremendous amount of analyses to be done within a short time for there had been a high pressure due to the public interest in the case.

The laboratory chose the locus D1S80 for screening for two reasons:

- First, the semen stain exhibited a relatively rare phenotype at that locus occurring at about 2.4% in the German population.

- Secondly, from isoelectro-focusing time the laboratory still had a number of electrophoresis equipment to run a lot of samples (at least 200 per day) by horizontal polyacrylamide gel electrophoresis followed by silver staining. Only those samples matching the D1S80 phenotype had to be analyzed at further loci, all STR loci including SE33.

Following this strategy the laboratory was able to type about 8000 samples within two months after the screening had been started. 4000 samples had been analyzed in the other state laboratories mentioned above. After about 12000 samples had been typed, a profile matching the stain profile was found.

The donor of the sample, a 30 year old man, confessed to both crimes. During the trial he even confessed to a third case where a young girl was missing. He also had killed her. Within his confession he showed the place where he had hidden the body of that girl.

Conclusion

The analyst dealing with stains of blood and of other materials of human origin cannot restrict on a few standard methods. There never will be a universal tool applicable to all demands of forensic casework. Something like a toolbox is needed including polyplex as well as monoplex approaches. It even has to contain loci not represented in the given DNA databases e.g. Y-chromosome marker systems. Thus, providers of polyplex kits also should provide the primer pairs of the loci included in the kits to ensure the analyst that he is using the same primer sequences to look at a given locus when deciding on monoplexes.

I hope companies will consider this when developing new sets of reagents. The efficiency of DNA databases essentially depends on optimum stain analysis.