

Practical Value of PGM 1 Subtyping in Blood- and Semen Stains and Hair Roots

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Introduction

There is no doubt about the value of PGM 1 typing of bloodstains. The first investigations using starch gel electrophoresis were done by Culliford (1967), Heidel (1968) and Brinkmann (1969). The amount of stain needed was extremely large (between 10 and 50 mg of dried blood). With the introduction of agarose gel electrophoresis (Monn 1968; Culliford 1971; Grunbaum 1974) and cellulose acetate electrophoresis (Zajac and Sprague 1975) the quantity of stain could be reduced considerably. Isoelectric focusing allowed subtyping of PGM 1 of fresh blood and stains with a significantly higher value of information than before (Bark et al. 1976). Especially Berg et al. (1979, 1981), Schwed and Hein (1983), Rittner et al. (1981), Oya et al. (1984) and Lawton and Kerr (1984) pointed out that additional or conversional bands occur with old blood samples respectively artificially prepared bloodstains as well as with cadaver tissue, thus leading to a false zymograms interpretation in some cases. Such conversional bands were not reported on in the few field studies published so far (blood- and seminal stains: Budowle et al. 1986; seminal stains: Oya et al. 1982). Therefore further examinations with practical cases seemed to be desirable.

Material and Method

The results of 370 blood- and 197 seminal stains as well as 40 hair roots in PGM 1 subtyping were interpreted. They were obtained from 81 different forensic cases from the Erlangen-Nürnberg Institute of Forensic Medicine which in their majority meant rape (46) and homicide (29).

The preparation of the samples was carried out depending on the carrier material. Whereas we did not pretreat bloodstains on cloth, cotton wool, and paper, we transferred blood- and seminal stains on other carriers (e.g. glass, metal, and others) to cotton tissue after having them partially dissolved. Bloodstains on cotton cloth and the like having a size of 0,1 to 1 cm² were cut into small strips or fibrillated and then incubated with 1-2 drops of dithiothreitol solution (8 mg dithiothreitol + 1 ml aqua bidest) in a filter sampler (Porex Medical, no. FS 410) at room temperature for 2 hours. The sampler was put into an Eppendorf tube and centrifuged at 2000 g. The eluate at the bottom of the tube was placed onto the gel using an applicator strip (Serva, no. 42989) in a distance of 1 cm from the anode. Focusing was done in commercial gels with immobilised pH gradients (LKB Immobiline Dry Plate, pH 5,6-6,6). Gel's rehydration was carried out according to the manufacturer's recommendation. Focusing at 5000 V, 4 mA, and 12 W; prefocusing 30 min, focusing 3 h. Enzyme specific agarose overlay staining as described by Spielmann and Kühnl (1982).

Results

Unambiguous results could be obtained with 58 % of the bloodstains, 51 % of the seminal stains, and 5 % of the hair roots; doubtful results were found with 6 % of the bloodstains, 12 % of the seminal stains, and 3 % of the hair roots. Due to small sample size or advanced inactivation of the enzymes, clearly readable zymograms could not be obtained with 36 % of the bloodstains, 37 % of the seminal stains, and 92 % of the hair roots. Seminal stains sometimes showed a mixture of the victim's and the accused's phenotype. 6 bloodstains showed additional bands being either in the region of the 1- or the 2+ band. These stains simultaneously exhibited misleading results in the ABO-system (acquired B).

Discussion

Concerning the bloodstains the percentage of unambiguous results in the present study is in good agreement with those of other investigations (Budowle et al. 1986). With seminal stains Budowle et al. (1986), however, found considerably less usable results (16 %), whereas Oya et al. (1984) obtained positive zymograms in 40 %. It is remarkable that Budowle et al. (1986) and Oya et al. (1984) in their examinations could not find one single false result. Berg et al. (1979, 1981), Schwerd and Hein (1983), Rittner et al. (1981) and Oya et al. (1984) had observed additional bands in the 1- and 2+ region with 1+ phenotypes, additional bands in the 2+ region with 1+ 2- phenotypes, and a disappearing of the 1- band with 1+ 1- phenotypes with artificially made bloodstains, respectively blood samples stored for months as well as with cadaver tissue.

We could show by an additional electrophoretic separation of stored blood samples in agarose gel the 2+ band of 1+ phenotypes as degradation artefact.

Finally it should be pointed out that these additional bands may even pose difficulties to persons experienced in PGM¹ subtyping. In our material such additional bands, however, occurred in less than 2 % of the examined bloodstains; therefore PGM 1 subtyping of stains can be recommended for routine cases as a reliable method.

References

- Bark JE, Harris MJ, Firth M (1976) Typing of the Common Phosphoglucomutase Variants Using Isoelectric Focusing - A New Interpretation of the Phosphoglucomutase System. *J Forensic Sci Soc* 16: 115-120
- Berg S, Ladiges ML, Ladiges O (1981) Der Einfluß von Blutproben- und Spurenalterung auf das PGM 1- und Gc-Subtypenmuster. *Z Rechtsmed* 87: 85-94
- Berg S, Ladiges O, Ladiges ML (1979) Über die Nachweisbarkeit der Tf c- und PGM 1-Subtypen in gealterten Blutproben. *Arch Kriminol* 164: 101-106
- Brinkmann B (1969) Bestimmung der Phosphoglucomutasetypen aus Blutproben. *Dtsch Z Ges Gerichtl Med* 66: 31-34
- Budowle B, Murch RS, Davidson LC, Gambel AM, Kearny JJ (1986) Subtyping Phosphoglucomutase 1 in Semen Stains and Bloodstains: A Report on the Method. *J Forensic Sci Soc* 31: 1341-1348

- Culliford BJ (1971) The Examination and Typing of Bloodstains in the Crime Laboratory. Law Enforcement Assistance Administration, Washington, D.C.
- Culliford BJ (1967) The Determination of Phosphoglucomutase (PGM) Types in Bloodstains. *J Forensic Sci Soc* 7: 131-133
- Grunbaum BW (1974) A Micro Procedure for Fast Typing of the Genetic Variants of Phosphoglucomutase. *J Forensic Sci Soc* 14: 151-157
- Heidel G (1968) Die spurenkundliche Bedeutung der Typen der sauren Erythrocyten-phosphatase. *Dtsch Z Ges Gerichtl Med* 63: 37-43
- Lawton ME, Kerr LR (1984) Phosphoglucomutase Types in Blood and Hair Roots Taken from Post-Transfusion Subjects. *J Forens Sci* 29: 445-448
- Monn E (1968) A New Red Cell Phosphoglucomutase Phenotype in Man. *Acta Genet* 18: 123-127
- Oya M, Kido A, Komatsu N, Shibata R (1984) Die Anwendung der Histoelktrofokussierung zur Bestimmung der PGM 1-Subtypen an menschlichen Körpergeweben. *Z Rechtsmed* 92: 225-230
- Oya M, Tröger HD, Tutsch-Bauer E (1982) PGM 1-Fokussierung von Sperma aus Scheidenabstrichen. *Beitr Gerichtl Med* 40: 233-236
- Rittner C, Fohlmeister M, Fittkau-Tombergs M (1981) Die PGM 1-Subtypen in der forensischen Spurenkunde. *Krim Forens Wiss* 44: 27-30
- Schwerd W, Hein M (1983) Lagerungsbedingte Änderungen bei der Isofokussierung von Merkmalen der Phosphoglucomutase (PGM 1) in Blutproben und -spuren. *Fortschritte der Rechtsmedizin*, Springer, Berlin-Heidelberg-New York
- Spielmann W, Kühnl P (1982) Blutgruppenkunde. Thieme, Stuttgart-New York
- Zajac PL, Sprague AE (1975) Typing of Phosphoglucomutase (PGM) Variants in Dried Bloodstains by the Grunbaum Method of Cellulose Acetate Electrophoresis. *J Forensic Sci Soc* 15: 69-74