TECHNICAL NOTE

Y. Katsumata,[1] M.D.; H. Ito,[1] B.S.; M. Aoki,[1] M.S.; H. Tsutsumi,[1] B.S.; K. Sato,[1] M.D.; M. Oya,[1] M.D.; O. Suzuki,[2] M.D.; and S. Yada,[1] M.D.

Phosphoglucomutase$_1$ and 6-Phosphogluconate Dehydrogenase Types in Human Skin and Adipose Tissue

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ABSTRACT: Attempts were made to detect phenotypes of the enzymes phosphoglucomutase$_1$ (PGM_1) and phosphogluconate dehydrogenase (PGD) in human skin and adipose tissues. Both enzymes could be typed using approximately 3 mg wet weight of tissue. Phenotypes could be distinguished after up to 15 days of aging for PGM_1 and ten days of aging for PGD. Analysis of isoenzymes is potentially useful for mediolegal identification of human skin and adipose tissue.

KEYWORDS: pathology and biology, phosphoglucomutase, phosphogluconate dehydrogenase, human skin, human adipose tissue, isoenzymes

Human skin or adipose tissue of victims of traffic accidents is often found adhering to motor vehicles. Identification of such tissues is important, especially in hit-and-run cases. Absorption-elution grouping of adipose tissue has been used for that purpose [*1,2*]. Polymorphism of phosphoglucomutase$_1$ (PGM_1), demonstrated in aqueous extract of human skin [*3*], could be used for the same purpose. Isoenzymes of PGM_1 and 6-phosphogluconate dehydrogenase (PGD) present in red cells are also known to occur in human semen [*4,5*], dental pulp [*6*], vaginal contents [*5,7*], and the hair bulb [*8-10*]. In the present study, attempts were made to detect phenotypes of PGM_1 and PGD in human skin and adipose tissue.

Materials and Methods

Human skin, subcutaneous adipose tissue, and cardiac blood were obtained from 55 cadavers at autopsy. Small specimens of skin were taken from the head and the abdomen

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[1]Senior lecturer, forensic chemist, forensic chemist, forensic chemist, graduate student, assistant professor, and professor, respectively, Department of Legal Medicine, Nagoya University School of Medicine, Nagoya 466, Japan.

[2]Assistant professor, Department of Legal Medicine, Hamamatsu University School of Medicine, Hamamatsu 431-31, Japan.

and of adipose tissue from the abdomen. Approximately 3 mm^3 (3 mg wet weight) of skin or adipose tissue was freeze-thawed, macerated for 30 min on a hollowed glass plate in 0.01 mL of 0.1*M* sodium phosphate buffer (pH 7.0) containing 1% Triton X-100, and then repeatedly compressed with a glass rod to lyse the cells as completely as possible.

PGM_1 and PGD typing was made essentially as described by Spencer et al [*3*] and by Fildes and Parr [*11*], respectively. Starch gels were prepared to 1-mm thickness using 10% Starch-Hydrolysed (Connaught, Toronto, Canada). Small strips of filter paper (Toyoroshi No. 2, Tokyo, Japan), 1 by 4 mm in size, were dipped into the lysates and then inserted into the starch gels. Electrophoresis was carried out at 4°C for 18 h at a constant voltage of 5 V/cm, followed by specific staining for PGM_1 or PGD. For comparison, hemolysates of the same subjects were simultaneously analyzed for isoenzyme types.

Results

The PGM_1 isoenzyme patterns of abdominal skin, scalp skin, and adipose tissue were clearly detected and were identical to those of hemolysates of the same individuals. Of these, 37 were Type 1, 16 Type 2-1, and 2 Type 2. Examples of the three phenotypes are shown in Fig. 1. It should be noted that the a and b bands are much more dominant than the c and d

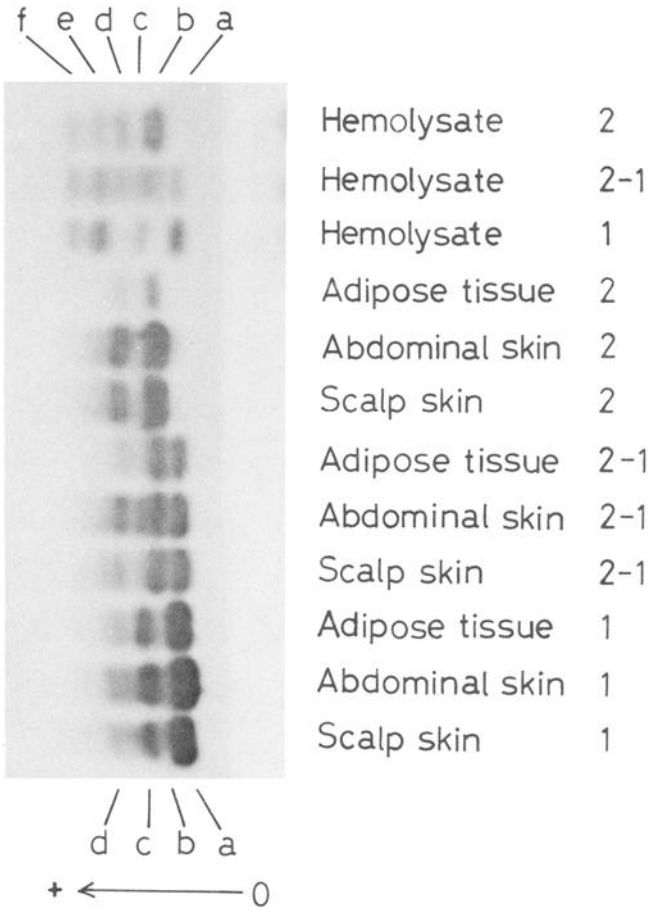

FIG. 1—*PGM_1 patterns in skin, adipose tissue, and red cell hemolysates.*

bands and that there appear to be no PGM_2 bands in the skin and adipose tissue. The PGD isoenzyme patterns of the skin and adipose tissue were also identical to those of hemolysates of the same individuals. Of these, 44 were Type A, 10 Type AC, and 1 Type C. Examples of the three phenotypes are shown in Fig. 2. In Type C, the hemolysate showed a minor band in addition to the main band, whereas the skin and the adipose tissue showed only a main band. The distributions of the phenotypes of PGM_1 and PGD are comparable to those of blood samples previously examined in the Japanese population [*12*]. The skin of both head and abdomen regions showed similar concentrations of PGM_1 and PGD.

The PGM_1 isoenzyme patterns of both skin and adipose tissue from ten individuals could be detected after up to 15 days of aging at room temperature. Of these, six were Type 1, three Type 2-1, and one Type 2. The PGD isoenzyme patterns of both skin and adipose tissue from ten individuals could be distinguished after up to ten days of aging. Of these, eight were Type A and two Type AC.

Discussion

The present results clearly show that PGM_1 and PGD phenotypes are detectable in human skin and adipose tissue. The concentrations of PGM_1 and PGD in abdominal skin were similar to those in scalp skin; therefore, the enzymes seem to be derived mainly from the skin tissue itself, and the contribution, if any, of the roots of scalp hairs, shown to exhibit PGM_1 and PGD activity [*8–10*], is very small. The presence of PGM_1 in skin tissue is not surprising, since the characteristic PGM_1 pattern has been shown to be present in fibroblasts from primary human skin cultures [*13*].

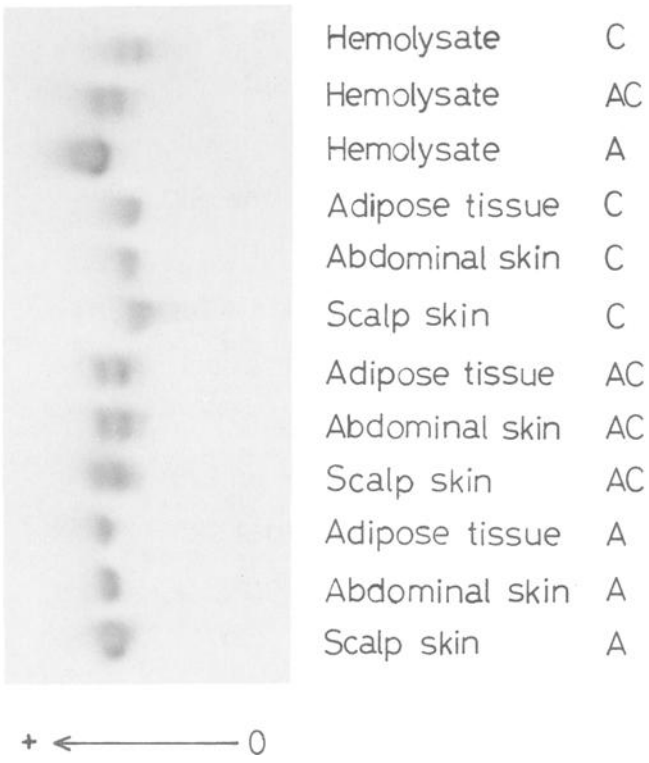

FIG. 2—*PGD patterns in skin, adipose tissue, and red cell hemolysates.*

The concentrations of PGM_1 and PGD were so high in the skin and adipose tissue that only a small amount (3 mg wet weight) of the tissues was required for detecting enzyme types. Although the skin is rich in collagen fibers and the adipose tissue is rich in fat droplets, both tissues showed high concentrations of PGM_1 and PGD. Therefore, the cells present in both tissues seem to contain large amounts of PGM_1 and PGD. These results are consistent with those of Ishimoto and Kuwata [*14*], who showed that tissue homogenates of most human organs such as liver, heart, and lung showed strong PGM_1 activity.

The results indicate that skin or adipose tissue can be identified using isoenzymes that are genetically determined. Studies using other isoenzymes are now in progress.

Summary

Isoenzyme typing of phosphoglucomutase$_1$ (PGM_1) and 6-phosphogluconate dehydrogenase (PGD) can be made with human skin and adipose tissue. About 3 mg wet weight of the tissues is required for typing of PGM_1 and PGD, and phenotypes are detectable after up to 15 days of aging for PGM_1 and 10 days of aging for PGD. Analysis of isoenzymes may find application in mediolegal identification of human skin and adipose tissue in certain cases of traffic accidents.

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Address requests for reprints or additional information to
Y. Katsumata, M.D.
Department of Legal Medicine
Nagoya University School of Medicine
Nagoya 466, Japan