

DEMONSTRATION OF LDH-X FOR IDENTIFICATION OF TESTICULAR TISSUES

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INTRODUCTION

The sperm-specific lactate dehydrogenase isoenzyme (LDH-X) appearing in electrophoresis between LDH-3 and LDH-4 is characteristic of human mature testis and spermatozoa.^{1,2)} Our previous studies have shown that detection of LDH-X by starch gel electrophoresis is quite suitable for forensic demonstration of semen from vaginal swabs.³⁻⁸⁾ This communication reports the successful application of the LDH isoenzyme method for determining the origin of a specimen adherent to the muffler of a car in a traffic accident and the results of some supplementary experiments.

CASE REPORT

A 30-year old man was run over by a car on a road at night and was found dead on the scene. There were severe lacerations around the left inguinal region and the perineal region. The scrotum was also lacerated, and the left testis was severely crushed (Fig. 1). Many abrasions and contusions were seen on the face, the chest and the left thigh. A medicolegal autopsy was made the following day, and the cause of death was diagnosed as brain contusions associated with basal skull fractures. One week after the accident a suspected car was found, and a piece of black, dried and dirty specimen with hairs weighing 0.5 g was collected from the muffler of the car. Histologic examination of the specimen revealed some sperm-like bodies in tubular structures. However, it was rather difficult to determine its origin microscopically because of the postmortal changes. The blood group of the specimen was determined by an absorption-elution technique as O, which was identical with the blood group of the victim.

Demonstration of LDH-X was carried out two weeks after collection of the specimen. Using an Ultra-Turrax homogenizer (Janke & Kunkel A. G., Staufen in Breisgau, Federal Republic of Germany), 0.1 g of the specimen was homogenized in 1.0 ml of physiological saline at 4°C. The homogenates were centrifuged at 3000 rpm for 15 min, and the supernatants were used for electrophoresis. Small filter papers (Toyoroshi No. 2, Tokyo, Japan) measuring 3 x 8 mm were dipped into the extracts and inserted into the gel. Starch gel electrophoresis and LDH staining was performed according to the method previously described.⁴⁾

Fig. 2 shows the electrophoretic pattern of LDH isoenzymes in normal fresh human testes (A) and in the specimen collected from the suspected car (B). As can be seen, LDH was successfully demonstrated from the specimen in spite of storage of three weeks and severe contamination. As a result, the specimen proved to be of testicular origin, which strongly indicates that the victim had been hit and run by the suspected car.



Fig. 1 Severe lacerations around the left inguinal region and the perineal region.

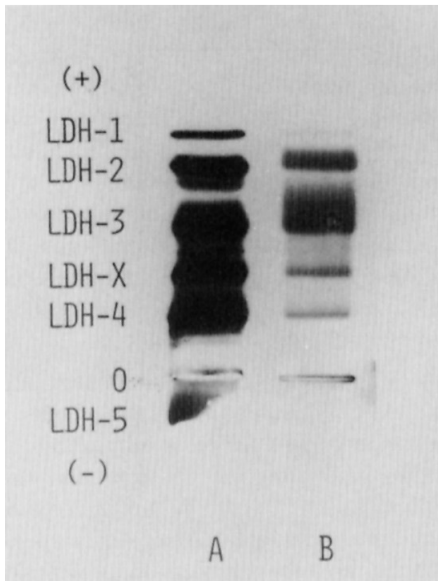


Fig. 2 Starch gel electrophoretic pattern of LDH isoenzymes in normal fresh human testes (A) and in the specimen collected at an actual traffic accident (B).

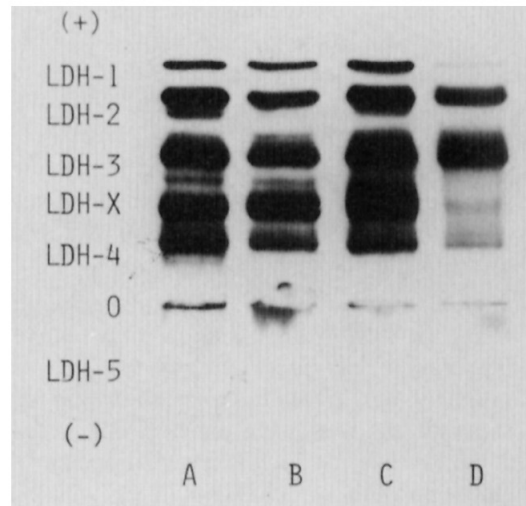


Fig. 3. Starch gel electrophoretic pattern of LDH isoenzymes in normal fresh human testes (A), and in human testes stored at 4°C (B), 20°C (C) and 37°C (D) for one week.

EXPERIMENTAL PART

Human testes were obtained from five male cadavers who were medicolegally autopsied within 24 hours after death. The testicular tissues weighing 0.1 g were stored at 4°C, 20°C and 37°C for varying periods from one week to six months and subjected to the demonstration of LDH isoenzymes as described above.

The results are summarized in Table 1. Fig. 3 shows the electrophoretic pattern of LDH isoenzymes in normal fresh human testes (A), and in the human testes stored at 4°C (B), 20°C (C) and 37°C (D) for one week. LDH-X was demonstrated at 4°C up to five months at 20°C up to five weeks and 37°C up to two weeks of storage. Therefore, detection of LDH-X is of practical use as another tool in identifying testicular tissues, especially in mass catastrophes such as traffic accidents and explosions. The principle herein reported may also be applicable to distinguishing the sex of cadavers in such cases.

Table 1. Detection of LDH-X on human testes stored for varying periods

Case No.	Age of cadaver	Temperature (°C)	Periods of storage											
			1W	2W	3W	4W	5W	6W	7W	3M	4M	5M	6M	
1	32	4	+	+	+	+	+	+	+	+	+	+	+	-
		20	+	+	+	+	+	-						
		37	+	+	-									
2	48	4	+	+	+	+	+	+	+	+	+	+	+	-
		20	+	+	+	+	+	+	-					
		37	+	+	+	-								
3	50	4	+	+	+	+	+	+	+	+	+	+	+	-
		20	+	+	+	+	+	-						
		37	+	+	-									
4	21	4	+	+	+	+	+	+	+	+	+	+	+	-
		20	+	+	+	+	+	+	-					
		37	+	+	-									
5	58	4	+	+	+	+	+	+	+	+	+	+	+	-
		20	+	+	+	+	+	+	-					
		37	+	+	-									

W : week, M : month

SUMMARY

The sperm-specific lactate dehydrogenase isoenzyme (LDH-X) was demonstrated in human testicular tissues stored for several weeks by starch gel electrophoresis. This method was successfully applied to determining the origin of a specimen adherent to the muffler of a car in a traffic accident.

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