FORENSIC APPLICATIONS OF HUMAN BODY FLUID ESTERASES

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ABSTRACT

Esterases of human semen and vaginal secretion show different electrophoretic patterns when studied using polyacrylamide gel with a thin strip of starch gel at the origin. Vaginal esterase occurs as a single cathodal zone, while semen esterase is manifested as a number of cathodal bands and hence the method can be used to distinguish semen and vaginal secretion in sexual assault cases.

Under these conditions, pseudocholinesterase (PCE) from blood plasma is detected as anodal zones and hence a mixture of blood and vaginal secretion can be demonstrated, indicating, but not proving that a bloodstain is of menstrual origin.

Experiences with the technique in the examination of a range of vaginal swabs is presented.

INTRODUCTION

Multiple forms of esterases have been found in a variety of human tissues, most of which are non-specific, showing overlapping substrate preferences and varying inhibition characteristics. Some of these esterases have, however, been identified by the use of selected substrates and inhibitors, of which esterase D (EsD) and carbonic anhydrase (CA) are regularly exploited in forensic blood grouping. Pseudocholinesterase (PCE) also has a polymorphic variant which is demonstrated by electrophoresis, but its value as a forensic blood typing marker is low.

The isozymes of these non-specific esterases have been reported as varying from tissue to tissue (Coates et al, 1975) and since the physiological production of seminal plasma (Mann, 1964) and vaginal fluid (Raffi et al, 1977) is different, it appeared that a comparison of the esterase isozymes from semen and vaginal fluid may enable each to be distinguished.

METHOD

Semen and vaginal fluid esterases were examined by electrophoresis in a 6.5% polyacrylamide gel with a thin strip of 7% starch gel at the origin. The gel buffer was 0.015M succinic acid - TRIS, PH4.8 and the bridge buffer was 0.016M citric acid - sodium hydroxide PH4.8.

Electrophoresis was carried out at 5V/cm for 16 hours.

The esterase isozymes were located by the method of Stern and Lewis, 1962.

RESULTS AND DISCUSSION

After electrophoresis, the semen resolved into six zones of cathodal esterase activity (fig. 1), with zones 1, 2 and 3 the strongest. Vaginal esterase occurred as a single zone with a mobility between that of zones 1 and 2 of semen esterases. Under these conditions, blood plasma cholinesterase (PCE) occurred as strongly staining zones anodal to the origin.

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When electrophoresis was carried out in polyacrylamide gels of concentrations above 7%, the vaginal esterase was sometimes seen as two zones giving a possible, though unlikely, confusion with semen esterases. Hence, a concentration of 6.5% acrylamide was used.

A further attempt at distinguishing the esterases of semen and vaginal fluid was made by investigating their substrate specificities. A number of α-naphthol salts and 4-methylunbelliferyl salts were used to locate the esterase isozymes.

The results (fig. 2) showed that both semen and vaginal esterases show strong activity with acetate salts only, indicating that these are non-specific acetyl esterases.

Vaginal swabs

Vaginal swabs were donated by females aged between 20 years and 45 years, taken at known times after intercourse and examined for semen esterase and vaginal esterase activity.

Of the thirty-five swabs taken a minimum of seven days after intercourse, twenty-four showed vaginal esterase activity and the others either showed no esterase activity (four swabs) or were scored as inconclusive (seven swabs). No swab gave an isozyme pattern similar to that produced by semen.

Of the swabs taken within four days post intercourse, semen esterase was regularly detected up to about eight hours post intercourse. Swabs taken after eleven hours post intercourse normally showed no semen esterase activity (fig. 3)

Menstrual blood

Under these conditions, the presence of vaginal esterase and PCE from blood plasma is indicative, though not proof of menstrual blood (fig. 1)

CONCLUSIONS

The results so far in this investigation show that using the electrophoretic conditions described,

1) semen and vaginal fluid esterases can be distinguished.

2) On a vaginal swab, the presence of azoospermic semen can be strongly indicated up to eight hours after intercourse. (Although such esterase isozymes may be present in other tissues, these are unlikely to be found on vaginal swabs).

3) Vaginal fluid esterases cannot be reliably detected in the presence of semen.

4) The presence of vaginal esterases and PCE from blood plasma gives an indication, though not proof, of menstrual blood.

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Fig. 1. Semen, vaginal secretion and blood esterases after polyacrylamide gel electrophoresis.

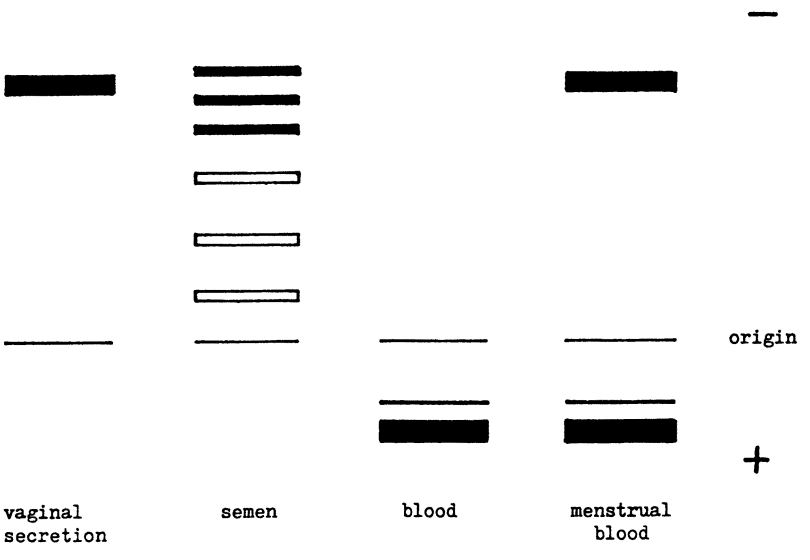

Fig.2. Reaction of semen and vaginal esterases with a number of substrates.

	Semen	Vaginal secretion
α-naphthyl acetate	++++	++++
propionate	+	+
butyrate	-	-
stearate	-	-
valerate	-	-
4-methylumbelliferyl-		
acetate	+	++
propionate	-	-
butyrate	-	-

Fig. 3. Semen esterase activity on vaginal swabs taken at known times after intercourse.