AN ARBORACEOUS CRYSTAL PATTERN IN AIR-DRIED SEMINAL FLUID

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(Received 26th February 1969)

The characteristic palm-leaf-like crystals in dried human cervical mucus, first observed by Papanicolaou (1933), occur abundantly around the day of ovulation, but are uncommon or even completely missing during other parts of the menstrual cycle (Rydbeg, 1948). An analogous variation of the crystallization pattern of cervical mucus is also seen during the menstrual cycle of the rhesus monkey (David & Mastroianni, 1968) as well as during the oestrous cycle of the cow (Garm & Skjerven, 1952). In women, the crystals are composed of sodium chloride, their forms being determined by a proteinaceous matrix (Rydbeg, 1948).

As early as 1921, similar crystals were observed in semen from cattle, horses, and rabbits (Yamane, 1921). The existence of a male sexual cycle corresponding to the oestrous cycle of female animals is strongly indicated by a number of investigations carried out during the last decade which have been reviewed by Kihlström (1966). The following study was undertaken to see whether a similar cyclic variation occurs in the palm-leaf-like crystals of seminal fluid. The relationship between the crystal pattern and the concentrations of proteins and some inorganic ions in the seminal fluid was also studied.

Semen was collected by artificial vagina from twelve rabbits of different breeds and twenty-one bulls of Swedish Red Breed. Human semen was obtained from two healthy men by masturbation. The ejaculates were freed from spermatozoa by centrifuging at 975 g for 10 min. Small drops of seminal fluid were placed on well-cleaned micro-slides and dried in air at room temperature. Palm-leaf-like crystals were seen in about 50% of the ejaculates from bulls and men, and in the overwhelming majority of the samples from rabbits.

In air-dried preparations of seminal fluid from rabbits, two distinct types of crystals were observed, the one called type R (right-angled crystals; Pl. 1, Fig. 1), the other type A (acute-angled crystals; Pl. 1, Fig. 2). Crystals having a transitional form intermediate between those of type R and A were also seen.

The rabbits were used for a continuous study of crystals in dried seminal fluid from ejaculates collected once a day for at least 80 successive days. The presence or absence of a cyclic variation in the observed proportions of the two types of crystals was studied by applying a serial $\chi^2$-test as described earlier (Kihlström & Fjellström, 1969). Statistically significant cyclic variations were
found in all but two rabbits. The mean duration of the cycles was 6·0 days, 
87% of the cycles having a duration between 5 and 7 days. This cyclic variation 
closely resembles that found earlier in other manifestations of male sexual 
functions in rabbits (Kihlström, 1966).

In the seminal fluid of nine rabbits, protein-nitrogen concentrations (Lowry, 
Rosebrough, Farr & Randall, 1951) and chloride ion concentrations (Kulhánek 
& Fíšer, 1966) were estimated.

Six of the same rabbits, together with three different rabbits, were used for 
determinations of sodium (Gilbert, Hawes, & Beckman, 1950; Smit, Alkemade 
& Verschure, 1951) and potassium ions (Gilbert et al., 1950; Smit et al., 1951) 
in the seminal fluid. The data thus obtained were studied in relation to the 
type of crystals of the same ejaculates.

The results are given in Table 1. Ejaculates containing only crystals of type 
R had low concentrations of protein-nitrogen and high concentrations of

### Table 1

<table>
<thead>
<tr>
<th></th>
<th>Crystals of type R only</th>
<th>Crystals of type A only</th>
<th>No crystals</th>
<th>Comparison by means of analysis of variance</th>
</tr>
</thead>
<tbody>
<tr>
<td>Protein-nitrogen</td>
<td>2·43±0·316 (n = 163)</td>
<td>2·71±0·346 (n = 135)</td>
<td>3·17±0·273 (n = 160)</td>
<td>v^2 = 19·97, P &lt; 0·0005</td>
</tr>
<tr>
<td>Cl^-</td>
<td>3·62±0·005 (n = 123)</td>
<td>3·48±0·006 (n = 112)</td>
<td>3·05±0·005 (n = 155)</td>
<td>v^2 = 44·06, P &lt; 0·0005</td>
</tr>
<tr>
<td>Na^+</td>
<td>1·88±0·010 (n = 201)</td>
<td>1·62±0·077 (n = 151)</td>
<td>1·39±0·107 (n = 40)</td>
<td>v^2 = 41·46, P &lt; 0·0005</td>
</tr>
<tr>
<td>K^+</td>
<td>0·96±0·048 (n = 201)</td>
<td>0·93±0·050 (n = 151)</td>
<td>1·05±0·082 (n = 40)</td>
<td>v^2 = 1·73, P &lt; 0·05</td>
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</tbody>
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sodium and chloride ions. In a few samples, no crystals at all were observed. 
In these cases, the concentrations of protein-nitrogen were relatively high and 
those of Cl^- and Na^+ were low. Crystals of type A occurred in ejaculates having 
moderate concentrations of protein-nitrogen, sodium ions and chloride ions. 
No relationships between the concentrations of K^+ and the type of crystals 
could be observed.

On the basis of these results, it is suggested that, in accordance with the 
conditions in the cervical secretion (Rydberg, 1948), the crystals are composed 
of sodium chloride, their shape being determined by a proteinaceous matrix. 
A full account of these results will appear elsewhere.

This work was supported by grants from the Swedish Natural Science 
Research Council and from the Faculty of Mathematics and Science of the 
University of Uppsala.
Fig. 1. Crystals of type R. × 1630.
Fig. 2. Crystals of type A. × 1630.

(Facing p. 376)
REFERENCES


