EFFECTS OF $\alpha$-CHLORHYDRIN UPON THE FERTILITY OF SPERMATOZOA OF THE CAUDA EPIDIDYMIS OF THE RAT

M. A. TURNER

Unit of Reproductive Biology, University of Liverpool

(Received 26th May 1970, revised 7th September 1970)

Male rats receiving daily injections of $\alpha$-chlorhydrin (U-5897) at a dose of 7 to 8 mg/kg become infertile (Ericsson, 1969; Ericsson & Baker, 1970). Fertility is lost within 4 to 5 days of the beginning of treatment and returns within 1 week of withdrawal of the drug. It has been proposed that $\alpha$-chlorhydrin acts preferentially on the epididymis rather than the testis, and that it particularly affects the head of the epididymis, possibly through interference with its normal blood supply (Ericsson, 1969). This, in turn, might hinder maturation of the contained spermatozoa. On the other hand, large doses of the drug are known to give rise to impaired motility in mature spermatozoa (Coppola, 1969; Samojlik & Chang, 1970) and changes in epididymal blood flow incurred by use of the drug have been shown to be most apparent in the lower regions of the duct (Turner, unpublished). It seemed likely, therefore, that the original speculations about the actions of the drug might be incorrect, but, first, it was necessary to determine whether the infertility resulting from use of the drug was due to an effect on the spermatozoa in the tail of the epididymis. The present paper reports on a small fertility trial designed to test out this possibility in rats.

Twenty mature male rats of the Sprague Dawley strain were divided into four groups. Group 1 consisted of untreated control animals, Group 2 of animals receiving daily subcutaneous injections of $\alpha$-chlorhydrin (8 mg/kg) and Group 3 of rats that had a ligature placed around the lower half of the body of the epididymis. Group 4 consisted of rats with ligated epididymides, which received daily injections of $\alpha$-chlorhydrin (8 mg/kg). The ligatures were placed in such a manner as to avoid any major blood vessel.

Presence of the fertilizing spermatozoon and cleavage of ova recovered from females mated in oestrus, were taken as criteria of male fertility. Rats in Group 1 were permitted to mate randomly five times, rats in Group 2 were divided into two sub-groups and allowed to mate on alternate days. Rats in Group 3 and 4 were permitted to mate on the 4th post-operative day. In these two groups, mating, if unsuccessful, was repeated on the 5th day. The success of a mating was judged by the presence of spermatozoa in the vagina the following morning. Ova were flushed from the uterine tubes 36 hr after mating and were examined under phase contrast.

The term 'fertilization rate' is used here to indicate the percentage of...
fertilized ova because recovery of eggs was almost 100%. On this basis, the fertilization rate in animals from Group 1 was 90.8%. Rats of Group 2 were infertile by the 3rd day and this was preceded by a significant decline in fertility (Table 1). Ligation of the epididymis had no effect upon the fertility of rats during this time, and since no fertilized ova were recovered from females paired with Group 4 males (Table 2), α-chlorhydrin must have been acting on mature spermatozoa from the ductus deferens and the tail of the epididymis.

### Table 1

**Fertilizing capacity of males treated with α-chlorhydrin**

<table>
<thead>
<tr>
<th>Males</th>
<th>No. of females*</th>
<th>Total eggs recovered</th>
<th>Total eggs fertilized</th>
<th>Mean fertilization rate</th>
</tr>
</thead>
<tbody>
<tr>
<td>Group 1 Controls</td>
<td>5</td>
<td>49</td>
<td>45</td>
<td>90.8</td>
</tr>
<tr>
<td>Group 2 Hr after first dose of U-5897</td>
<td>24, 36, 48, 60</td>
<td>30, 30, 37, 51</td>
<td>29, 20, 16, 0</td>
<td>97.2, 68.8, 40†</td>
</tr>
</tbody>
</table>

* The number of males/group was the same as the number of females.

† *P* < 0.01, significantly different from the control.

### Table 2

**Fertilizing capacity of males with ligated epididymides**

<table>
<thead>
<tr>
<th>Males</th>
<th>No. of females*</th>
<th>Total eggs recovered</th>
<th>Total eggs fertilized</th>
<th>Mean fertilization rate</th>
</tr>
</thead>
<tbody>
<tr>
<td>Group 1 Controls</td>
<td>5</td>
<td>49</td>
<td>45</td>
<td>90.8</td>
</tr>
<tr>
<td>Group 3 Epididymis ligated</td>
<td>3</td>
<td>33</td>
<td>31</td>
<td>94.4</td>
</tr>
<tr>
<td>Group 4 Epididymis ligated and treated with U-5897</td>
<td>3</td>
<td>26</td>
<td>0</td>
<td>0</td>
</tr>
</tbody>
</table>

* The number of males/group was the same as the number of females.

A low dose of α-chlorhydrin (7 mg/kg/day) has been shown to produce no morphological changes in the epididymis, or in the contained spermatozoa, that can be seen with the light microscope, but larger doses of the drug are known to produce a swelling of the head of the epididymis (Gunn, Gould & Anderson, 1969; Ericsson & Baker, 1970). This swelling is in the duct itself and suggests the presence of an abnormal quantity of fluid. An extension of the present work has shown that transport mechanisms in the epididymis are not seriously affected by the drug since, in treated animals, the passage of Indian ink through the duct was unimpaired. It seems that, in addition to its effect on
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the motility and fertilizing capacity of spermatozoa in the tail of the epididymis, α-chlorohydrin interferes with fluid resorption in the duct (Gunn et al., 1969).

Coppola (1969) believed that α-chlorohydrin might interfere with phospholipid synthesis in the epididymis and Samojlik & Chang (1970) suggested that the oxidative-reductive enzyme system of the spermatozoa might be inhibited. Gunn et al. (1969) suggested that this compound might interfere with absorption of fluid in the duct and thus affect the maturation of the spermatozoa. It has been shown, however, that the blood flow in the epididymis is disturbed during treatment with the drug (Turner, in preparation) and this might be significant, particularly in relation to the swelling of the duct following heavy dosage.

While the precise mechanism of action of α-chlorohydrin still remains unclear, it does seem that it might be valuable as a physiological tool in the study of epididymal function.

This work was supported by grants from the Medical Research Council and the Ford Foundation. My thanks are due to Dr T. D. Glover, for his help and criticism.

REFERENCES