ESTIMATION OF THE OPTIMUM INTERVAL BETWEEN INSEMINATION AND OVULATION IN THE RABBIT BY DOUBLE INSEMINATION

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(Received 28th October 1968)

Rabbit spermatozoa do not attain fertilizing ability until several hours after deposition in the female genital tract but, equally, they do have a finite life. Rabbit spermatozoa aged in vitro for 24 hr fertilized only 5% of eggs in competition with equal numbers of fresh spermatozoa (Roche, Dziuk & Lodge, 1968). When does were double-mated, the first buck sired 86% of offspring when the interval between the first mating and ovulation was 10 hr or less (Dziuk, 1965). This showed that spermatozoa that have spent 10 hr in the female genital tract have an advantage in fertilizing ability compared to those which spent less time there.

The following experiment was conducted to determine whether or not the first male would maintain this advantage as the interval between the first insemination and ovulation was increased beyond 10 hr.

Semen was collected from New Zealand White (W) and Dutch Belted (B) bucks with an artificial vagina. Genetic homozygosity was established by periodic non-experimental inseminations. Each W female was inseminated within 45 min of collection with $20 \times 10^6$ motile spermatozoa from either a B or W male in 0.3 ml of 2.9% sodium citrate in distilled water. Three hours later, the doe was re-inseminated with spermatozoa from the male of opposite colour. The interval between the first insemination and ovulation was 19, 16, 13 or 10 hr. Ovulation was induced by an intravenous injection of 50 i.u. of human chorionic gonadotrophin at the appropriate time. It was assumed that insemination does not by itself induce ovulation (Hammond & Asdell, 1926). The paternity of the offspring was established by frequent examinations of young following delivery from the fifty-eight pregnant does.

A summary of the results is shown in Table 1. The first buck had a distinct advantage (sired 73% of offspring) over the second, when mated 10 hr before ovulation. As the interval between the first buck and ovulation increased from 13 to 16 hr and finally to 19 hr, the proportion of offspring from the first buck was reduced from 54% to 39% and finally to 18%. Litter size varied considerably among does but was not different among groups. There were twenty-nine litters from does inseminated first with spermatozoa from a W buck and twenty-nine litters when a B buck was mated first. The B and W bucks were

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mated first with almost equal frequency in each of the four intervals before ovulation. There were 247 W and 168 B offspring. The first buck sired 209, and the second sired 206 offspring.

The observation that the first buck has an advantage and sired a larger proportion of the offspring (73%) when mated 10 hr before ovulation substantiates the previous observation using a similar régime (Dziuk, 1965). As the interval from insemination to ovulation increased, the proportion of offspring from the first buck decreased to 18%, indicating that intervals greater than 10 to 13 hr are disadvantageous. On the basis of these two observations, it appears that rabbit spermatozoa fertilize eggs more readily when they have spent 10 to 13 hr in the female reproductive tract than when they have spent either more or less time there. The optimum time for insemination would thus appear to be 10 to 13 hr before ovulation. The rabbit is an induced ovulator, ovulation occurring 10 to 12 hr after mating. From an evolutionary stand-

### Table 1

<table>
<thead>
<tr>
<th>Interval between insemination and ovulation</th>
<th>No. of litters</th>
<th>No. of offspring</th>
<th>Offspring from first male</th>
</tr>
</thead>
<tbody>
<tr>
<td>First insemination (hr)</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>19</td>
<td>9</td>
<td>51</td>
<td>9</td>
</tr>
<tr>
<td>16</td>
<td>16</td>
<td>117</td>
<td>46</td>
</tr>
<tr>
<td>13</td>
<td>17</td>
<td>138</td>
<td>74</td>
</tr>
<tr>
<td>10</td>
<td>16</td>
<td>109</td>
<td>80</td>
</tr>
<tr>
<td>Total</td>
<td>58</td>
<td>415</td>
<td>209</td>
</tr>
</tbody>
</table>

point, those females ovulating 10 to 13 hr after mating would appear to have a selective advantage because spermatozoa were best able to fertilize eggs after this interval. Equally, bucks whose spermatozoa reached optimum fertilizing ability 10 to 13 hr after mating would have an advantage over others if the mating-to-ovulation interval were genetically inflexible.

The spermatozoa from W bucks apparently have some advantage over B spermatozoa because they fertilized a greater proportion of eggs than B spermatozoa in reciprocal double matings. This innate advantage of one male over another has been noted previously by Beatty (1960).

### REFERENCES


