BRIEF COMMUNICATION

EFFECT OF EJACULATION FREQUENCY AND BREED
ON SEMEN CHARACTERISTICS AND SPERM OUTPUT
OF RABBITS*

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Summary. Six Dutch-type and nine New Zealand White rabbits were each ejaculated at frequencies of 1×12 hr, 1×24 hr and 2×48 hr. Ejaculate volume and sperm concentration were lower ($P < 0.01$) at 1×12 hr than at either of the other two frequencies. Daily sperm output did not differ significantly among the three frequencies. Dutch-type rabbits had a significantly lower daily sperm output than New Zealand Whites.

Gregoire, Bratton & Foote (1958) found that increasing the ejaculation frequency of rabbits from once weekly to once daily resulted in decreases of 50% in sperm concentration and 40% in ejaculate volume. Weekly sperm output, however, was almost 400% greater at the higher frequency. Desjardins, Kirton & Hafs (1965) compared the sperm output of twelve rabbits each collected at four frequencies. Weekly sperm output ($\times 10^{-6}$) averaged 273 with one ejaculate weekly, 519 with one ejaculate collected daily except Sundays and 619 when two successive ejaculates were collected each Monday, Wednesday and Friday.

The present study was designed to determine an ejaculation frequency which would maximize sperm output and to study breed differences in semen characteristics. Three groups, each consisting of two Dutch-type and three New Zealand White rabbits over 1 year old, were used in a change-over design. Body weight averaged 2.40 kg for Dutch-type and 3.88 kg for New Zealand White rabbits. Ejaculation frequencies tested with each rabbit were: one ejaculate each 12 hr (1×12 hr), one ejaculate each 24 hr (1×24 hr) and two ejaculates about 15 min apart each 48 hr (2×48 hr). Each ejaculation was preceded by one false mount. The male was allowed to mount the teaser doe, make a series of pelvic thrusts and then allowed or forced to dismount. Collection of the ejaculate by artificial vagina normally followed during the next mount. Each ejaculation frequency was imposed for 36 days; the 6-day adjustment period was followed by a 30-day experimental period.

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After ejaculation, the collection tube was separated from the artificial vagina and the gel plug was removed if present. Ejaculate volume was read directly from the calibrated collection tube. Paired ejaculates for rabbits collected 1×12 hr or 2×48 hr were pooled before determination of sperm concentration by haemocytometer counts. Since some semen remains in the artificial vagina, residual spermatozoa were flushed with about 15 ml distilled water from the latex liner into a small flask immediately after collection. Artificial vagina flushings were pooled for each rabbit on a 48-hr basis.

There were no statistically significant differences between the characteristics of ejaculates collected at 1×24 hr and 2×48 hr except for the percentage of ejaculates containing gel (Table 1). At 2×48 hr, 70% of the first ejaculates

TABLE 1

EFFECT OF EJACULATION FREQUENCY AND BREED ON CHARACTERISTICS OF RABBIT SEMEN

Characteristic	1×12 hr	1×24 hr	2×48 hr	Dutch-type	New Zealand White	Mean, weighted by rabbits
Volume/ejaculate (ml)	0.31**	0.41	0.42	0.34	0.41	0.38
Total volume/48 hr (ml)	1.24**	0.83	0.84	0.83	1.06	0.97
Sperm concentration (10^6 /ml)	152**	243	260	198	232	219
Daily sperm output (dso) (10^6)	103	110	114	88	123*	109
dso in flushings (%)	12	10	10	12	10	11
Ejaculates containing gel (%)	65	62	35**	54	54	54

* Mean is significantly ($P < 0.05$) different from that for the other breed.

** Mean differs ($P < 0.01$) from the means for the other two ejaculation frequencies.

contained a gel plug, but only 5% of the second ejaculates contained gel. In six of 225 pairs of ejaculates both the first and second ejaculates contained gel. First ejaculates (0.45 ml) were larger ($P < 0.01$) than second ejaculates (0.38 ml) during 2×48 hr. Collection at 1×12 hr reduced ($P < 0.01$) ejaculate volume and sperm concentration as compared to either 1×24 hr or 2×48 hr. Morning and evening ejaculates during 1×12 hr averaged 0.31 and 0.31 ml and 69% and 63% of these ejaculates contained gel.

Daily sperm output (dso) was not significantly affected by ejaculation frequency. Including spermatozoa flushed from the artificial vagina, dso averaged 109×10^6 cells and values for individual rabbits ranged from 48 to 174×10^6 cells. About 11% of the dso was recovered in artificial vagina flushings which agrees with the value reported by Bredderman, Foote & Yassen (1964) for a Walton-style artificial vagina. Excluding spermatozoa recovered in the flushings, the fifteen rabbits ejaculated an average of 679×10^6 spermatozoa/week. This value is about 10% greater than the weekly sperm output of 619×10^6 cells (without flushings) reported by Desjardins *et al.* (1965) for rabbits collected twice successively each Monday, Wednesday and Friday. However, these workers did not use false mounting for sexual preparation and found that second ejaculates

contained more spermatozoa than first ejaculates. Possibly for these reasons, they obtained fewer spermatozoa (519×10^6 /week) with collections daily except Sunday. In the present study, sperm counts were not made on individual successive ejaculates, but the volume of first ejaculates exceeded that of second ejaculates.

Dutch-type rabbits tended to have smaller and less concentrated ejaculates than New Zealand Whites, but the differences did not approach statistical significance. However, DSO for the Dutch-type rabbits was significantly lower ($P < 0.05$). Mean DSO for the six Dutch-type rabbits of 88×10^6 cells was probably lowered about 8% by the value of 48×10^6 cells for one rabbit subsequently found to have only one active testis. If the value for this rabbit is doubled, the Dutch-type rabbits would have averaged 96×10^6 spermatozoa/day. This value agrees with that of about 101×10^6 spermatozoa/day for four Dutch-type rabbits collected once daily (Gregoire *et al.*, 1958).

The number of spermatozoa recovered in the flushings was correlated with DSO ($r = 0.47$). The percentage of the DSO recovered in the flushings was correlated with ejaculate volume in the collection tube ($r = -0.44$) but not with sperm concentration ($r = 0.11$).

This study demonstrates that for most experiments where it is desirable to maximize DSO an ejaculation frequency of 2×48 hr can be used. This ejaculation frequency should be practical for most rabbits and, except for very critical in-vivo kinetic studies, should reveal the most information with the least amount of work.

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