Swimming speed and fertilisation rates of ram sperm from high and low prolificacy populations

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ABSTRACT

A study in 1980 showed that rams from a high prolificacy flock (Waihora) were more successful than rams from a low prolificacy flock (Whatawhata fertility control) in fertilising ewes by copulation. In this study translational swimming speeds of ram sperm in caprogen at 39°C were measured by twin-beam laser velocimetry.

The mean swimming speed of the Waihora ram sperm was slower in both years (99 *v* 144 μm/s, $P < 0.01$, 1981; 111 *v* 144 μm/s, $P < 0.05$, 1982). Seven rams were common to both years; the between year ram repeatability in swimming speed was 0.92 ($P < 0.001$). Within year ram repeatabilities were 0.22 in 1981 and 0.45 in 1982.

Other semen traits (flash frequency, flash time, sperm concentration and motility score) did not differ between ram flocks in both years.

Keywords Rams; prolificacy; fertilisation rates; copulation; sperm; swimming speed; laser.

INTRODUCTION

Romney rams from the Waihora high prolificacy selection programme fertilised 80% of ewes by copulation while rams from a Whatawhata Romney flock unselected with respect to fertility fertilised 59% of similar ewes (Moore, 1981). The ewes were classified as fertilised if cleaved ova could be flushed from their oviducts 3 days after copulation.

Subsequently ejaculates from rams from these 2 flocks were collected and examined for several traits that might explain the above fertilisation differences, including the determination of swimming characteristics by twin beam laser velocimetry (Wilson and Harvey, 1983).

MATERIALS AND METHODS

Of the 10 Waihora and 10 Whatawhata rams assessed for fertilisation rate differences in 1980 (Moore, 1981), 7 Waihora rams and 5 Whatawhata rams were assessed for semen traits in 1981, and 4 and 5 respectively in 1982. An additional 5 rams of each flock were assessed in 1982. All rams were trained for semen collection by means of the artificial vagina. In 1981 ejaculates were collected from 12 rams on 9 days from 2 July to 17 July while in 1982, a sample of 8 rams out of a total of 19 were collected on 15 days from 25 May to 14 June. In 1981 a total of 6 to 9 ejaculates/ram were obtained and in 1982, 10 ejaculates/ram. There were 7 rams common to both years. Sperm concentration of the ejaculate was estimated from the absorbance measured at a dilution of 1 to 400 in a Spectronic 20. The semen was also scored undiluted under a low power microscope on the vigour of the wave motion. A score of 1 was given to a sample with minimal individual sperm forward motion, and a score of 3.3 to a sample with vigorous shoal movement, the intermediate scores being 1.3, 1.7, 2, 2.3, 2.7 and 3. Immediately after collection the ejaculates were diluted to a concentration of 80 million sperm/ml with 5% egg yolk caprogen (Shannon, 1964). Samples were jacketed in water at 32°C which then cooled slowly to ambient temperature. They were futher diluted to 8 million cells/ml in 0.5% caprogen and warmed to 39°C immediately before assay for swimming velocity.

In the assay technique (Wilson and Harvey, 1983), sperm cells are suspended in a cuvette and illuminated by 2 focussed parallel laser beams

separated by 60 μ m. As sperm swim they rotate and scatter light (flash) into 1 of 2 detectors. If a sperm happens to swim perpendicularly across both beams a cross-correlation technique is used to determine the time taken for the cell to travel between the beams (transit time). One of the walls of the cuvette is placed perpendicular to the parallel beams, and use is made of the pronounced tendency of sperm to swim along the wall of the assay cuvette (Harvey and Woolford, 1980)

Each assay was carried out over a period of 11 to 22 minutes allowing the accumulation of 1300 to 2600 transit-times. Each ejaculate was assayed twice within 9 hours from the time of collection during the 1981 experiment and 6 times during the 1982 experiment. Besides mean swimming speed, the velocimeter also produced estimates of the mean flash frequency and the mean flash time, i.e. the average duration of reflection from the flat surface of the sperm head.

Data were analysed within years using the maximum likelihood technique of Patterson and Thompson (1971). Ram means from these analyses were used for between-year repeatabilities.

RESULTS

While the Waihora ram semen was more concentrated than the Whatawhata in both years, the difference was significant only in 1982 (Table 1). Sperm from the Waihora rams swam significantly slower in both years. Flash time was significantly longer for the Waihora rams in 1982, but not in 1981. There were no significant differences in motility score or flash frequency.

Sperm concentration (log transformed to normalise the distribution) had high ram repeatabilities in both years (Table 2). Where there was a large flock effect, as in the 1982 concentration data, the removal of the flock effect reduced the repeatability. Motility score was repeatable in 1981 because there were rams present in this year that had consistently sluggish wave motion; in 1982 the observer was evidently not able to detect consistent differences among rams. Swimming speed was more repeatable in 1982 than in 1981. Flash frequency had a very low repeatability in both years. Flash time had a very low repeatability in 1981 but a significant repeatability in 1982.

The repeatability of semen traits between years calculated from the 7 rams measured in both years was zero for motility and flash time and high for sperm concentration and swimming speed (Table 3). When flock effects were removed, repeatabilities of swimming speed and flash frequency were almost significant.

There was a negative correlation between

TABLE 1 Flock differences in semen traits.

	1981			1982		
	Waihora	Whatawhata	SED	Waihora	Whatawhata	SED
Number of rams	7	5		9	10	
Sperm concentration (millions/ml)	3548	3174	548 NS	3563	2864	225 *
Motility score	2.79	2.42	0.33 NS	2.36	2.48	0.13 NS
Swimming speed (μm/s)	99	144	13 **	111	144	14 *
Flash frequency (flashes/s)	33.1	33.4	1.61 NS	35.3	36.2	0.60 NS
Flash time (ms)	4.48	4.61	0.20 NS	4.41	4.09	0.11 *

TABLE 2 Within year ram repeatabilities ± SE of semen traits.

	1981		1982	
	Flock effect not removed	Flock effect removed	Flock effect not removed	Flock effect removed
Log_e sperm concentration	0.60 ± 0.12	0.61 ± 0.12	0.62 ± 0.10	0.48 ± 0.11
Motility score	0.54 ± 0.13	0.53 ± 0.13	0.11 ± 0.08	0.11 ± 0.09
Swimming speed	0.22 ± 0.12	0.06 ± 0.09	0.45 ± 0.11	0.38 ± 0.11
Flash frequency	0.08 ± 0.11	0.10 ± 0.12	0.00 ± 0.06	0.00 ± 0.06
Flash time	0.01 ± 0.09	0.02 ± 0.10	0.32 ± 0.11	0.22 ± 0.10

frequency and time of flash in both years and between swimming speed and flash time and swimming speed and concentration in 1982 (Table 4). Flash time and concentration were positively correlated in 1982.

Table 5 shows that swimming speed had a significant negative correlation with fertilisation rate in 1982 when flock effects were not removed. It is consistent in sign with the non-significant correlation in 1981. Flash time had a negative correlation with fertilisation rate in 1981, but a positive correlation in 1982.

DISCUSSION

Of the traits measured, only sperm concentration and swimming speed had acceptable repeatabilities within both years. In neither 1981 nor 1982 was sperm concentration related to fertilisation rate, whereas 1982 swimming speed had a significant negative correlation with fertilisation rate. The flock with the greater ability to fertilise ewes had markedly slower sperm swimming speed.

What rationale can there be for an inverse relationship between sperm swimming speed and fertility? It is tempting to suggest that fast swimming sperm may use up energy sources more quickly than slow swimming sperm, so that slow swimming sperm survive for a longer period in the female reproductive tract and increase the chances of fertilisation. K.L. Macmillan (unpublished) has quantified the survival times of bull sperm in caprogen at 37, 38 and 39.5°C. There was a decrease of 24 h in survival time/degree Celsius increase in the presence of catalase and a 15 h decrease without catalase. Part of this decrease in survival time may be due to increased swimming speed as M.C. Wilson (unpublished) has shown that bull sperm swimming speed in caprogen increased by 14 μm/s/degree Celsius from 35 to 40°C.

We are not aware of other studies relating objectively measured sperm swimming speed and fertility in sheep. Milligan *et al.* (1980) using multiple exposure photography showed that a group of fertile men had higher mean sperm velocities than a group of infertile men. However Cohen *et al.* (1982) found that human sperm samples swimming with higher than average velocity had a lower success rate in fertilising hamster ova than sperm samples from the same group swimming with average velocity.

TABLE 3 Between-year ram repeatabilities of semen traits.

	Flock effect not removed	Flock effect removed
Log_e sperm concentration	0.70 ± 0.21 *	0.38 ± 0.37
Motility score	0	0
Swimming speed	0.92 ± 0.16 ***	0.60 ± 0.28 †
Flash frequency	0.57 ± 0.27 †	0.59 ± 0.28 †
Flash time	0	0

† $P<0.10$

TABLE 4 Correlations between semen traits — 1981 below diagonal (10 df); 1982 above diagonal (17 df).

	Log_e sperm concentration	Motility score	Swimming speed	Flash frequency	Flash time
Log_e sperm concentration	-	-0.06	-0.49*	-0.44†	0.52*
Motility score	0.49	-	-0.07	0.28	-0.38
Swimming speed	-0.46	-0.47	-	0.24	-0.71***
Flash frequency	-0.19	0.20	0.36	-	-0.44†
Flash time	-0.02	-0.41	-0.12	-0.93***	-

† $P<0.10$

TABLE 5 Correlations of fertilisation rate with semen characteristics.

	1981		1982	
	Flock effect not removed	Flock effect removed	Flock effect not removed	Flock effect removed
Log_e sperm concentration	0.26	0.17	0.33	-0.23
Motility score	0.41	0.29	-0.31	-0.01
Swimming speed	-0.42	0.02	-0.69*	-0.55
Flash frequency	0.39	0.54†	0.12	0.70
Flash time	-0.68*	-0.70†	0.63†	0.43
Degrees of freedom	10	9	7	6

† $P<0.10$

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