

**SOURCES OF VARIATION FOR PERCENT NORMAL SPERMATOZOA IN *BOS INDICUS* TYPE BULLS: FIELD RESULTS**

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**SUMMARY**

Ejaculates from 120 bulls with single or repeated records were assessed for Percent Normal Spermatozoa (PNS). The data were used to identify the importance of genetic and environmental sources of variation using a restricted maximum likelihood algorithm and an animal model. Effects fitted included year-season combination, technician and age. Only year-season combination was found to be important. The heritability of the trait appears to be in the range 0.4-0.6. Permanent environment effects do not appear to be important in the expression of the trait for the animals studied.

While the heritabilities are not significant at the 5% level due to the small number of records and the unbalanced structure of families, there is a trend in the results to suggest that the trait is heritable and may be useful as a selection tool for improving bull fertility. The results highlight the difficulties arising from the attempted analysis of field results.

**Keywords:** Semen quality, bull, heritability, repeatability

**INTRODUCTION**

The identification of abnormal sperm cells in bull semen has been widely associated with impaired fertility (Rollinson 1951, Barth and Oko 1989;). Most of the recorded evidence suggests that the semen of bulls of normal fertility contains less than 20 percent abnormal spermatozoa (Barth and Oko 1989).

The spermiogram (analysis of individual sperm morphology) is a reflection of the health of the seminiferous epithelium at the time of the spermiogenesis of the cells being observed. It also reflects the health of the epididymis during epididymal transit of the spermatozoa and storage of the spermatozoa in the caudal epididymis (Barth and Oko 1989).

Previous estimates of heritability for reproductive traits in bulls have been low indicating a strong role of environment. Several workers have reported the heritability for fertility to be between 0.15 and 0.30 (Chandler *et al* 1985; Godfrey *et al* 1988). Davis (1992) reported very low heritability for individual records of semen quality, measured as the proportion of cows in calf to artificial insemination, although for repeated inseminations the heritability was increased to 0.29. Other work indicates that several semen traits may be more heritable than previously estimated offering hope that selection can improve the genetic potential for semen quality (Entwistle 1992). Chandler *et al* (1985) reported a heritability for percent primary abnormalities of 0.84, although the standard error on the estimate was large.

There is considerable evidence to suggest that season may influence semen characteristics. Seasonal variation in semen quality may be due to seasonal changes in nutrition and environmental temperature. Typically in Northern Australia there may be a decline in semen quality in bulls (particularly young bulls) grazing poor quality native pasture during the dry season. Non-adapted bulls may also experience a decline in semen quality during the hot summer months.

The objectives of this study were to investigate the genetic and environmental factors influencing the trait PNS in the semen of Droughtmaster bulls. The results are expected to assess whether PNS is a repeatable measurement and to provide an indication of the value of sire selection as a means of improving PNS.

#### **MATERIALS AND METHODS**

Semen records were obtained from Droughtmaster bulls bred on The University of Queensland veterinary science farm at Pinjarra Hills in South Eastern Queensland. Droughtmaster cattle are a composite breed derived from the *Bos indicus* (Brahman and Zebu) (50%) and Shorthorn (50%) breeds. The data were not part of a designed experiment but were field data collected as part of student training in reproduction. All bulls that were included were pedigreed.

Semen collection and evaluation were carried out at least once for each bull. Semen was collected using electro-ejaculation. For each bull PNS was determined by microscopic (bright light) examination of an eosin-negrosin stained semen smear (Barth and Oko 1989). Between 1981 and 1991 only the percent live-normal spermatozoa and the percent live spermatozoa were recorded. The majority of records were from this period. The definition of PNS used in the study for records dating from this period was the proportion of live spermatozoa that were morphologically normal expressed as a percentage of the proportion of spermatozoa that were live. Later records reported PNS as the proportion of both live and dead spermatozoa that were morphologically normal and so this statistic was used directly.

Variance components were estimated by Restricted Maximum Likelihood using a derivative-free algorithm (DFREML) fitting an animal model (Meyer 1993). A univariate analysis considered the effect of the genes of each animal (random), the year-season combination of the collection of the semen sample (fixed) and age in days fitted as a linear covariate. Three views of the available data were considered. For the first, Data set 1, all available data (215 records, 22 sires) were used in the analysis. Further analyses considered only the individual bulls' highest PNS performance (120 records, Data set 2), and only those bulls with repeated records (152 records from 57 tested bulls, Data set 3).

Repeated records were used to assess variance due to permanent environment by fitting a second animal effect in the model. Solutions were obtained for fixed effects and covariates as a by-product of the univariate analysis.

**RESULTS**

215 records with pedigree information were available. Data were recorded from routine semen collection records from June 1981 through to October 1996. The bulls were sons of 22 sires with 1 to 21 sons per sire. Bull age at collection ranged from 512 to 1776 days with an average age of 804 days ( $\pm 225.5$  days). The total number of individuals with records included in the analysis was 120. Most collections occurred in autumn and winter (during the teaching period).

The average value for PNS was  $64\% \pm 23\%$ . 124 of the 215 collections registered values for PNS of greater than the 70% threshold indicating normal fertility (Barth and Oko 1989). Of the 120 individual bulls' highest PNS collections, 87 were above the recommended threshold. The heritability for PNS appeared to fall within the vicinity of 0.4-0.6 for the population studied when models were optimised for log likelihood, however the nature of the data meant that the standard errors for the estimates were quite large (Table 1).

The large reduction in log likelihood for models fitting permanent environment as a second random effect suggested that the effect of permanent environment was not important in the observed population. The observed repeatability at convergence for PNS was 0.63-0.67 for the models and data fitting repeated records. The log likelihood when fitting all effects for data set 1 (all records) was 91% lower than for data set 2 (highest PNS record, log likelihood = -326.34) and the log likelihood for data set 3 (repeated records) was 26% lower than that for set 2. This may have been caused by the biased sample of bulls for which records were taken on multiple occasions.

**Table 1. Estimates of genetic parameters for Percent Normal Spermatozoa (PNS)**

	Data set	Number of records (No. bulls)	Heritability		Permanent environment effect		Phenotypic variance
			$h^2$	s.e.	$c^2$	s.e	
Model fitting age,	1	215 (120)	0.40	0.22	0.23	0.40	515.13 % <sup>2</sup>
year-season and	2	120 (120)	0.56	0.37			283.81 % <sup>2</sup>
technician	3	152 (57)	0.48	F*	0.19	F	517.06 % <sup>2</sup>
Model fitting	1	215 (120)	0.40	0.32	0.24	0.57	516.34 % <sup>2</sup>
year-season and	2	120 (120)	0.55	0.37			280.85 % <sup>2</sup>
technician	3	152 (57)	0.47	F	0.20	F	514.37 % <sup>2</sup>

\* F denotes failure to approximate sampling variances

Fitting age to the model decreased the log likelihood for all data sets. The largest positive influence on the log likelihood was the year-season combination. Fitting year-season increased the log likelihood by 19%, 24%, and 24% for the respective data sets (1,2 and 3). Because technician effects were confounded with some year-season combinations and the inclusion of technician as a fixed effect did not elevate the log likelihood by more than 2%, the effect was not considered to be important.

## DISCUSSION

The log likelihood was maximised when no records other than the highest PNS record for each bull was used in the analysis and when year-season and technician were included in the analysis. The highest PNS record for the sire was thought to best represent the genetic potential of the animal because the bulls tested a number of times were usually animals with an initial test performance below the threshold value for PNS. A test result may have been low for one of many reasons including the stage of maturity of the bull, the occurrence of systemic illness, and localised inflammation of the reproductive tract.

The more usual semen quality characteristics, volume and motility of the semen sample, were not considered in the analysis because they were thought to be biased by the method of collection of the sample (electro-ejaculation in this case) and the skill of the operators (student veterinarians). PNS may have been influenced by the skill or biases of the technician assessing the trait (trained laboratory personnel in this case) however in this study the effect of technician was shown not to be important.

While the heritabilities are not significant at the 5% level due to the small number of records and the unbalanced structure of families, there is a trend in the results to suggest that the trait is heritable and may be useful as a selection tool for improving bull fertility. The analysis does not support that permanent environment plays an important role in the expression of PNS although this may have been affected by the nature of the sample.

The results of the analysis highlight the difficulties associated with the analysis of field data, particularly when the trait is measurable in a single sex. Despite having 15 years of well maintained records with pedigree information available, it was difficult to draw any meaningful conclusions from the data.

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