PN-AAW-541

REPRODUCTION IN JAVANESE SHEEP: EVIDENCE FOR A GENE WITH LARGE EFFECT ON OVULATION RATE AND LITTER SIZE[1]

G. E. Bradford[2], J. F. Quirke[3], P. Sitorus[4], Ismeth Inounu[4]
Bess Tiesnamurti[4], F. L. Bell[2,5], I. C. Fletcher[4] and D. T. Torell[2,6]

University of California, Davis, CA 95616 and
Small Ruminant CRSP, Bogor, Indonesia

ABSTRACT

Three breeds of Javanese sheep are described briefly and data suggesting the segregation of a gene with large effect on ovulation rate and litter size are presented. The three breeds are Javanese Thin Tail (JTT), Javanese Fat Tail (JFT) and Semarang (SEM), the last possibly a substrain of JTT. All three breeds have mean mature ewe weights under 30 kg. Ovulation rate and litter size did not differ significantly among the three; all had litter sizes of up to 4 or 5 with a mean for mature ewes of approximately 2. Ovulation rate ranged from 1 to 5 and had an average within-breed repeatability of .8 within season and .65 between seasons. Within-breed repeatability of litter size was .35 ± .06. Prenatal survival in pregnant ewes with two, three and four or more ovulations averaged 93, 88 and 86% over two seasons. Dams that had at least one ovulation rate or litter size record ≥3 produced two groups of daughters in approximately equal numbers, one group with many records ≥3 and mean ovulation rate and litter size of 2.73 and 2.31, respectively, and one group with ovulation rates and litter sizes of 1 or 2 and corresponding means of 1.39 and 1.38. Dams with ovulation rate or litter size records of only 1 or 2 produced daughters in which over 90% had records of only 1 or 2. Estimated heritabilities for the mean of approximately three ovulation rate or litter size records from these daughter-dam comparisons exceeded .7. These results suggest segregation of a Booroola-type gene, one copy of which increases ovulation rate by about 1.3 and litter size by .9 to 1.0. Relationships between duration of estrus and ovulation rate, and between timing of release of luteinizing hormone and number of eggs shed, resemble the pattern in Booroola Merino more closely than that in Finnish Landrace or Romanov, supporting the hypothesis of a major gene.
(Key Words: Sheep, Major Genes, Ovulation, Embryo Mortality, Repeatability, Hormones.)

Introduction

The Javanese Thin Tail (JTT) breed of sheep, sometimes referred to as Priangan, has been reported to have high prolificacy (Mason, 1978, 1980; Obst et al., 1980; Fletcher et al., 1982). This characteristic of Indonesian sheep is not surprising, in view of the fact that typically these animals are kept in small flocks in close association with human households, characteristic of the conditions in which most, if not all, prolific breeds of sheep have been found.

In the early stages of a research program designed to characterize performance traits of Indonesian sheep, we confirmed the existence of highly prolific ewes in the JTT breed, but a notable aspect of this finding was an exceptional variability among ewes in litter size at birth. For example, the proportion of quadruplet births reached 10% or more in groups with a mean litter size of 1.9 to 2.0. There also appeared to be a high repeatability of litter size. These observations are characteristic of a population in which the Booroola gene for high ovulation rate is segregating (Piper and Bindon, 1982). We decided therefore to obtain information on ovulation rate and to examine the mode of inheritance of ovulation rate and litter size in Javanese sheep.

The animals used were a group of JTT ewes in an experimental flock maintained by the Indonesian Animal Production Research Institute (BPT) at the Cicadas Station near Bogor, West

[1]The authors express their appreciation to Rafael Sitomorang, Subandriyo and Rolf Jensen for their contributions to this project. Supported by the Small Ruminants Collaborative Res. Support Program, U.S. AID Grant DSAN XII 0049, the Irish Agr. Inst., and Coop. Ext. Serv., Univ. of California.

[2]Dept. of Anim. Sci., Univ. of California, Davis, CA 95616, USA. Address for reprint requests.

[3]The Ag. Inst. Grange, Dunsany, Co. Meath, Ireland.

[4]Balai Penelitian Ternak, P.O. Box 123, Bogor, Indonesia.

[5]Coop. Ext. Serv., Orland, CA 95963.

[6]7950 Saner Drive, Ukiah, CA 95482.

Received August 30, 1985.
Accepted February 4, 1986.

Java; the flock also contained Javanese Fat Tail (JFT) ewes and a small group of ewes from the Semarang area of Central Java (SEM), and these two groups were also included in the study. Preliminary summaries of portions of these data have been reported elsewhere (Bradford et al., 1983; Inounu et al., 1985; Sitorus et al., 1985). We report here the mean and repeatability for ovulation rate and litter size of the three groups of ewes, duration of estrus and some endocrine parameters, and discuss the mode of inheritance of the unusual prolificacy of the Javanese breeds suggested by the results.

Materials and Methods

The Cicadas Station is located 45 km northeast of Bogor, towards Jakarta, at an elevation of 65 m, latitude 6°S and longitude 106°E. Mean maximum and minimum temperatures at the Station are quite uniform throughout the year, with monthly averages ranging from 32 to 34.5 C and 21 to 22 C, respectively. Mean rainfall (1971 to 1984) averaged 350 to 400 mm per month from December through April; about 300 mm each in May, October and November; and 180 to 220 mm per month from June through September.

Sheep of the JTT breed, Garut strain, were purchased from the Garut region of West Java and brought to the Station in 1978 and 1980. At the time the first measures of ovulation were made in 1983, there were approximately 90 breeding-age JTT ewes in the flock, one-half representing purchased ewes and one-half representing younger ewes born in the flock in 1981 and 1982. In addition, there were 37 mature JFT ewes, purchased in April, 1981 and 13 of their daughters born in 1981 and 1982, and 18 Semarang ewes purchased in late 1981 and 4 of their daughters. The JTT and JFT ewes had all been at Cicadas from 1980 or 1981 on or from birth, while the Semarang ewes had been kept at a similar station in the region until December 1982.

The JTT strain, as the name implies, is characterized by a relatively narrow tail, of medium length. Some animals show evidence of Fat Tail ancestry, in that the base of the tail is appreciably wider. Coat color is usually black, white, or a combination of the two; white with black circles around the eye and some black spots on the body is a common pattern. Brown spotting and the blackbelly pattern occur at low frequencies. Males are horned; rams of the Garut strain are frequently used for ram fighting, a popular spectater sport of the Garut region. The earless gene occurs at quite high frequency, especially in the Garut strain, and thus ear length phenotypes include normal, short ear and earless, corresponding to homozygotes for the normal ear allele, heterozygotes and homozygotes for the short ear allele, respectively. The animals typically are partially to completely covered with a mixture of wool and kemp, which in the warm, humid climate of the country tends to be severely cotted. The amount of kemp and presence of partially woolless animals suggest some hair sheep ancestry.

The JFT strain in its purest form is found on the island of Madura in the eastern part of Indonesia. It is also the predominant type in East Java (Mason, 1978), while a wide range of admixtures of JTT and JFT types exists in Central Java. The JFT animals in this flock had been purchased in Grati, East Java. They included apparently pure Fat Tail, with the large, heavy tails characteristic of this type, and individuals that appeared to have some Thin Tail admixture. All had the white color characteristic of the JFT breed. Javanese Fat Tail males are polled, and sheep of this strain show more evidence of hair sheep ancestry than the JTT breed, with some JFT animals being completely free of wool at maturity, although most have fleeces similar to those described for JTT sheep. Ear length is normal.

Sheep of the Semarang strain are white, with relatively thin tails and on average smaller stature than either of the other two strains. Males are horned and ear length is normal.

The sheep were housed at all times in raised houses with slatted floors, with fresh-cut forage brought to them daily. The principal forage fed was Napier grass grown on the 5 Ha Cicadas Station. This was supplemented with various species of local grasses trucked in during the dry season, usually June through September or October, when the forage growth on the Station was inadequate to meet needs of the flock. A concentrate supplement, consisting of 60% corn, 25% rice bran, 10% coconut meal, 2.5% bone meal, 1.5% salt and 1% limestone was fed in amounts ranging from 100 to 580 g per head per d, depending on production status of the animals and on forage supply and quality.

During the period from acquisition of the first animals to September 1, 1982, young ewes were mated as they reached approximately 19 kg, and all ewes were mated directly after weaning their lambs. In preparation for measurement of ovulation rate, mating was suspended on September 1, 1982, leaving all ewes open as of February, 1983. All ewes over 9 mo of age were treated with vaginal sponges containing 60 mg medroxyprogesterone acetate[2] on April 4, 1983. The sponges were removed on the morning of April 18 and the ewes were weighed on the following day. Ewes were checked twice daily for estrus with vasectomized rams from April 19. Ninety to 100% of each of the three groups came in estrus following sponge removal, with the largest number on d 3 in each group. Of the 146 which came in estrus, 1, 16, 45, 32, 3 and 3% were in estrus on d 1 through 6, respectively. Endoscopies to count corpora lutea (CL) were performed on all ewes, including those that had not shown estrus, on April 27, 28 or 29. Estrous checking was continued and ewes detected in estrus were hand-mated to one of 16 different JTT rams, at the next estrus. More than 80% of all matings occurred on May 7, 8 and 9. Most ewes received one service only, when first detected in estrus. All ewes were reexamined for ovulation rate on May 12. All endoscopies were performed by one person.

A subsample of 58 ewes, 24 JTT, 22 JFT and 12 Semarang, including ewes with 1, 2 and 3 or more ovulations at first estrus within each breed and balanced by age to the extent feasible, were selected for a study of duration of estrus and to characterize the pre-ovulatory LH discharge. Ewes were checked for estrus every 2 h beginning on May 4. As soon as a ewe was observed in estrus, blood samples were collected at 2-h intervals from a jugular vein into evacuated tubes containing EDTA. The time of cessation of estrus was established by continuing testing at 2-h intervals with the teaser rams until at least two tests confirmed that the ewe would not accept the ram. The duration of estrus was determined using the same protocol in 18 additional ewes. The blood samples were chilled in iced water; after centrifugation the plasma samples were stored at −20 C until assayed for luteinizing hormone (LH).

Plasma LH concentrations were determined in duplicate 200-μl aliquots in a single assay using a liquid phase double antibody radioimmunoassay (Avenell et al., 1985). The sensitivity of the assay was .05 ng/tube and intra- and inter-assay coefficients of variation for levels of 2.5 and 8.8 ng/ml were 13 and 11%, respectively. The results were expressed in terms of NIH-HL-15. The overall mean LH concentration in the two samples taken following termination of estrus was 2.6 ng/ml (SD = 2.4) and the pre ovulatory LH discharge was considered to have commenced when the LH concentration exceeded 10.0 ng/ml.

Blood samples also were taken from 109 and 92 ewes on the morning of d 10 and 11, respectively, of the cycle in which the ewes were mated, for progesterone assay. The objectives were to establish mean progesterone levels for these sheep and to determine if there were higher progesterone levels in the ewes with ovulation rates of 3 to 5. Plasma progesterone concentrations were determined using a conventional radioimmunoassay with extraction into hexane, a tritiated progesterone label (1, 2, 6, 7 - ^{3}H Progesterone, TRK413, Amersham) and a charcoal dextran separation step (Avenell et al., 1985). The sensitivity of the assay was .01 ng/ml and the within-assay coefficient of variation for a plasma pool containing 2.25 ng/ml progesterone was 7%.

Ewes were not checked further for estrus after the early May matings, so only those conceiving at the one cycle lambed. They were housed in small groups under close supervision at lambing time to ensure a valid count of lambs born to each ewe and hence as accurate an estimate as possible of prenatal survival.

Following weaning of the lambs, the same groups of ewes were again synchronized and mated at the synchronized estrus in the first week of February 1984. The ewes were divided into two groups 6 wk prior to the synchronized estrus and given two levels of concentrate supplement. Corpora lutea were again counted by endoscopic examination, this time by one of three persons not including the person who had done the examinations in 1983. Embryo survival by number of CL was calculated for each lamb crop. Where number of lambs born exceeded the number of CL recorded (3% of ewes in the first season, 11% in the second), the CL count was increased to equal litter size.

In early June 1983, following completion of the first set of matings and ovulation rate measurements, several scores and measurements were recorded to provide a more complete description of the sheep in the experiment. A condition score on a 1-to-5 scale, with 1 thinnest and 5 fattest (Russell et al., 1969) was assigned

---

[2]Upjohn Co., Kalamazoo, MI.

independently to each ewe by three persons, with the average used as the score for the animal. Tail length, measured from the anus to the tip of the last coccygeal vertebra; and tail width and thickness measured at the widest part of the tail, were recorded. Measurements were made with a tape measure, ruler and calipers, and were recorded in centimeters. Wool cover and wool density were scored on a 1-to-9 scale, with increasing number indicating increasing area covered, and increasing average density of the existing wool cover whatever the area, respectively.

Statistical Analyses. Data were analyzed using least-squares procedures (Harvey, 1977). The models included breed and birth year where appropriate. The tables of results indicate factors included in the analysis of those data. Repeatability of ovulation rate was calculated by correlation between pairs of observations, within each breed and pooled across breeds. Repeatability of litter size was calculated from the ewe-within breed and within-ewe components of variance computed using the method of Turner and Young (1969), using all available records, on up to six lambings, for all ewes in the experiment.

Results and Discussion

Tail measurements and wool and condition scores are summarized in table 1. The model used included breed and year of birth (as a measure of ewe age). Body weight was included as a covariate; its inclusion removed most of the effects of ewe age for some but not all variables.

These data suggest that the Semarang type is much more similar to the Thin Tail than to the Fat Tail breed, with the only significant difference between SEM and JTT in the six traits being a slightly shorter tail. The JFT ewes, on the other hand, had longer, wider, thicker tails, as expected, less wool cover, a less dense fleece, and a higher condition score. Based on both body weights and tail measurements, it appears the JFT ewes in this study were of a different strain or in poorer condition (or both) compared with those described by George (1982).

The reason for the significant ewe age effect on tail length, independent of body weight, is not clear, but it may indicate that tail length continues to increase with age, whether or not the ewe gains weight. The effect of age on wool cover score, independent of weight, is consistent with the observation that older ewes have less wool.

Litter size recorded in the flock prior to the start of the present experiment is summarized in table 2, as an indication of the prolificacy level of the three breeds. The data illustrate the variability of litter size of the JTT group, particularly as they mature. Data on the other two breeds are limited, but the JFT data suggest a prolificacy approaching that of the JTT, when ewe age is taken into account. This is consistent with the results reported by Obst et al. (1980). The older group of original JTT ewes contained a few very prolific individuals, including a ewe that produced 25 lambs (litters of 3, 3, 6, 4, 5 and 4) in 4 yr. It is possible that sampling variation, as well as age, is involved in the difference between this group and the other age groups, although Mason (1980) also reported much higher mean litter size for mature than for young ewes of this breed. It is of interest to note that even in this group, with a mean litter size of 2.47, single births are the most frequent class.

Observed means for body weight at sponge removal and ovulation rate at first and second estrus are presented in table 3 by ewe breed and age. The data were analyzed within breed and combined, using models including birth year only and birth year plus body weight as a covariate, and including breed for the combined analysis. The effect of body weight on ovulation rate, with birth year in the model, was not significant in five of the six breed × estrus period groups. Birth year effects on body weight and ovulation rate were significant, within breeds and overall. Breeds differed significantly in body weight, with JTT heavier than the other two, but the three breed groups did not differ significantly in ovulation rate at either estrus.

There are several points of interest in these results. First is the wide variability in ovulation rate in all three breeds, with up to four CL even in the small sample of Semarang ewes. A second item is the large age effect; numbers of very young ewes were small, but the pattern of low mean ovulation rate for 1-yr-old ewes was similar in all three breeds. A third point of interest is the high ovulation rate of the mature JTT ewes, consistent with the high litter size recorded earlier for these ewes (table 2).

Ovulation rate and body weight data for the February 1984 mating season are presented in table 4. The mean ovulation rate at the synchronized estrus in 1984 was similar to that at

TABLE 1. LEAST-SQUARES MEANS ADJUSTED FOR BODY WEIGHT, FOR TAIL MEASUREMENTS, WOOL AND CONDITION SCORES FOR EWES OF JAVANESE THIN TAIL (JTT), JAVANESE FAT TAIL (JFT) AND SEMARANG (SEM) BREEDS OF SHEEP

Item	No.	Tail			Wool scores[a]		Condition score[b]
		Length, cm	Width, cm	Thickness, cm	Cover	Density	
JTT	79	19.3	5.6	2.7	6.7	7.3	2.0
JFT	44	22.9	10.2	5.1	5.9	6.6	2.3
SEM	19	17.5	5.5	3.1	6.5	6.9	2.1
Differences[c]							
JTT - JFT		−3.64**	−4.63**	−2.37**	.81**	.73**	−.32**
JTT - SEM		1.79**	.06	−.34	.22	.37	−.06
Regression on body wt (b ± SE)		.26 ± .053**	.16 ± .030**	.09 ± .015**	−.01 ± .027	.04 ± .022	−.07 ± .008**
Effect of birth yr		**	NS[d]	NS	**	NS	NS
Residual variance		6.90	2.26	.56	1.75	1.20	.16

[a]Subjective scores, 1-to-9 scale; area covered and density increasing with increasing score.

[b]1-to-5 scale, condition increasing with score.

[c]Significance determined by Bonferroni t.

[d]NS = nonsignificant (P > .05).

**P < .01.

the corresponding estrus in 1983. The two levels of supplementation had no effect on body weight and only a small effect on ovulation rate (Inounu et al., 1985).

An interesting feature of the ovulation rate data is the exceptionally high repeatability compared with that in other breeds (e.g., Hanrahan and Quirke, 1985). The results are summarized in table 5.

Correlations between the two measures of ovulation rate at consecutive cycles in the first season were ≥.80 for each of the three breeds.

TABLE 2. LITTER SIZE OF THREE BREEDS OF JAVANESE SHEEP (1979 to 1982)

	Litter sequence													
	1st			2nd				3rd and later						
Item / Litter size:	1	2	3	1	2	3	4	1	2	3	4	5	6
Javanese Thin Tail													
No. litters	26	15	5	17	15	6	5	12	9	7	8	1	1
Mean		1.54			1.98					2.47			
Javanese Fat Tail													
No. litters	16	13	2	7	5	3							
Mean		1.55			1.73								
Semarang													
No. litters	12	3											
Mean		1.20											

TABLE 3. OBSERVED MEANS FOR BODY WEIGHT AND OVULATION RATE BY AGE OF EWE FOR THREE BREEDS OF JAVANESE SHEEP (1983)

			No. CL 1st cycle						No. CL 2nd cycle					
Birth Year	No. ewes	Ewe wt, kg	1	2	3	4	5	Mean	1	2	3	4	5	Mean
Javanese Thin Tail														
≤79	15	29.6	4	2	4	4	1	2.73	3	2	5	3	2	2.93
80	28	25.6	15	9	4			1.61	11	12	2	3		1.89
81	25	21.1	14	8	3			1.56	11	9	2	3		1.88
82	7	17.1	5	1	1			1.43	5	1	1			1.43
Total	75	24.1	38	20	12	4	1	1.80	30	24	10	9	2	2.05
Error mean square		14.6						.79						1.10
Javanese Fat Tail														
80	32	24.9	11	11	8	2		2.03	9	11	8	3	1	2.25
81	8	19.9	6	1	1			1.38	6	1	1			1.39
82	4	17.1	4					1.00	4					1.00
Total	44	23.2	21	12	9	2		1.82	19	12	9	3	1	1.98
Error mean square[a]		16.7						.77						.97
Semarang														
80	18	22.2	8	7	2	1		1.78	5	8	4	1		2.06
82	3	16.3	3					1.00	2	1				1.33
Total	21	21.3	11	7	2	1		1.67	7	9	4	1		1.95
Error mean square[a]		13.1						.69						.67

[a]From analyses including birth year in the model.

TABLE 4. OBSERVED MEANS FOR BODY WEIGHT AND OVULATION RATE BY AGE OF EWE FOR THREE BREEDS OF JAVANESE SHEEP (1984)

Birth year	No. ewes	Ewe weight, kg	No. ewes with CL count 1	2	3	4	5	Mean no. CL
Javanese Thin Tail								
≤80	48	27.0	21	19	6	2		1.77
81	32	23.1	17	12	3			1.61
82	13	21.0	4	7	1	1		1.92
Javanese Fat Tail								
≤80	33	26.8	11	14	7		1	1.97
81	11	21.1	10	1				1.09
82	4	24.4	4					1.00
Semarang								
80	16	23.5	8	6	2			1.63
82	3	19.1	3					1.00

Inclusion of birth year in the model used for analysis of the data reduced these only slightly, and the pooled average, considering birth year, was .80. Including body weight as a covariate reduced this only marginally, to .79. This high repeatability is also clearly evident from the joint distributions for the two cycles (table 6); breeds are combined in this table, because of the similarity of the repeatability estimates. As may be noted from this table, 95 of the 140 ewes had identical ovulation rates at the two cycles, and of the 31 ewes that had three or more CL at first estrus, 28 had three or more at second estrus. Repeatability is high for all ovulation rates, including one and two as well as the higher multiples. If ewes with a CL count ≥3 are excluded, the repeatability within the first season was .64 and between the first and second seasons was .43 and .48, based on first and second ovulation rates in season 1, respectively. As indicated by the repeatabilities in table 5, the correlation was not quite as high between seasons, possibly in part because of the two nutritional treatments described earlier. However, of the 40 ewes measured in the second season that had had at least one ovulation rate record ≥3 in the first season, 21 were ≥3, 17 were ≥2 and only two had records 1 in the second season. Only three ewes that had not had a record ≥3 in the first season had three in the second season.

Repeatability of litter size, estimated from a within-breed analysis of 376 records of 126

TABLE 5. REPEATABILITY OF OVULATION RATE IN THREE BREEDS OF JAVANESE SHEEP WITHIN AND BETWEEN SEASONS

Breed	Observations correlated[a] 1 · 2 No.	r	1 · 3 No.	r	2 · 3 No.	r
JTT	75	.81	71	.52	71	.59
JFT	43	.88	43	.79	43	.80
SEM	21	.80	19	.89	19	.77
All[b]	140	.80	133	.63	133	.66

[a]1 and 2, observations at consecutive cycles in season 1, 3, season 2.

[b]Estimate from analysis with breed and birth year in the model.

TABLE 6. JOINT DISTRIBUTIONS OF OVULATION RATE AT FIRST AND SECOND CYCLES (MAY 1983 THREE BREEDS COMBINED)

No. CL 1st cycle	No. CL 2nd cycle 1	2	3	4	5
1	53	14	3		
2	3	28	7	1	
3		3	10	9	1
4			3	3	1
5					1

ewes, was .35 ± .06. The estimate from the first two records of these ewes was .32 ± .08, from analysis of variance, and .35 by simple correlation. These values may be contrasted with estimates ranging from .03 to .24 reported by Hanrahan and Quirke (1985) for a number of ewe breeds in Ireland, and an average within-group value of .14 ± .07 for several crossbred groups (Dzakuma et al., 1982).

The relationship between prior litter size and ovulation rate in the first reason was very high. Of 19 ewes with two or more lambings that had previously given birth to triplets or more, 18 had three or more ovulations in one or both cycles. The ewe that had had 25 lambs in six lambings was the only one, out of 140, with CL counts of 5 in both cycles in 1983. Of 33 ewes that had previously given birth to singles or twins only, in two to five lambings, 27 had an ovulation rate ≤2 in both cycles. This close relationship between prior litter size and current ovulation rate contrasts with correlations of .05 to .13 found in three breeds by Hanrahan and Quirke (1985). These results indicate a high repeatability of ovulation rate between seasons in the Javanese sheep, and also suggest a high prenatal survival rate.

Fertility, prenatal survival rate and litter size for the 1983 and 1984 seasons are summarized in table 7. Overall fertility of mated ewes in the first season, following mating at one estrus only and with a single service in most cases, was 84%. One of the 16 rams used was apparently sterile, and exclusion of mates of this ram gives a mean fertility estimate of 89%. Matings in the second season were at the estrus immediately following sponge withdrawal, and fertility was somewhat lower, 72% overall at the one cycle. An additional 19% lambed to mating at a subsequent cycle, resulting in an overall lambing rate of 91%. Thus, in general, these sheep exhibit good fertility.

Prenatal survival rate, measured as number of lambs born/number of corpora lutea, in pregnant ewes, was exceptionally high, with survival rates in ewes with two, three and four or five CL of 96, 87 and 84% in the first season and 91, 89 and 88% in the second season. Although the numbers of ewes are not large, the high survival rates are consistent across strain, age, season and ovulation rate. There seems little doubt that these sheep are above the general average embryo survival rates as given, for example, by the equation of Hanrahan (1982), which yields estimates of 85, 75, 66 and 57% for survival of two to five ova, respectively. Among those who have examined prenatal survival for ewes with up to four or more corpora lutea, Ricordeau et al. (1976, 1982) have reported similarly high rates for Romanov and Romanov cross ewes. Meyer (1985) reported above-average values for Booroola cross ewes and G. E. Bradford and Lahlou-Kassi (unpublished) have found high values for the D'Man breed. On the other hand, survival for twin ovulations in several non-prolific breeds, e.g., Berrichon (Ricordeau et al., 1976), Romney (Meyer, 1985) and Targhee (Bradford, et al., 1986) are equal to or lower than the estimate of 85%, while the Border Leicester × Romney cross was significantly higher than contemporary Romneys in this respect (Meyer, 1985). Collectively, the data available suggest that prolific breeds and crosses, with the possible exception of the Finnish Landrace, have higher prenatal survival rates than the average of the breeds providing the data for Hanrahan's (1982) summary. The different breed estimates cited here are, of course, confounded with environmental differences; however, there is nothing obvious in the nutritional or environmental conditions for the sheep in the present study, or for the D'Man sheep in Morocco (G. E. Bradford and Lahlou-Kassi, unpublished) that would lead one to expect superior prenatal survival for these breeds. If there is a real difference in prenatal survival in favor of prolific breeds, a possible explanation is that there has been greater opportunity for natural selection for prenatal survival in breeds with high ovulation rate.

Gestation period for the ewes that lambed to first service in season 1 is summarized in table 8. The overall observed mean of 148.7 d is similar to that of many breeds, and not shorter than typical of the species, as is the case for at least two prolific breeds, Finnish Landrace and Romanov. The difference among the three groups was significant, with the JTT breed having a longer gestation than the other two breeds by about 1.5 d. The effect of litter size was not significant. There was essentially no difference among litter sizes one to three; gestation period for litters of four and five was shorter, as has been observed previously (Bradford et al., 1974), but there were not enough of these litters to establish statistical significance. In general, these sheep appear typical of the species in terms of gestation period, including a low coefficient of variation, 1.5% in this case.

TABLE 7. FERTILITY AND EMBRYO SURVIVAL IN RELATION TO NUMBER OF CORPORA LUTEA

		Number of corpora lutea														
		1		2			3			4,5			All		Mean litter size
Breed	Age, yr	No.[a]	NL[b]	No.	NL	ES[c]	No.	NL	ES	No.	NL	ES	No.	% L[d]	
May 1983[e]															
JTT	≥2	13	10	14	11	100	7	2	100	7	7	77	41	73	2.03
JTT	1,2	14	14	11	10	95	3	3	67	3	2	75	32	91	1.55
JFT	≥2	8	8	11	9	100	8	6	83	4	3	100	31	84	2.08
JFT	1,2	10	10	1	1	100	1	1	100				12	100	1.25
SEM	All	7	6	9	8	88	4	3	100	1	1	100	21	86	1.88
Total		52	48	46	39	96	23	15	87	15	13	84	137	84	
February 1984[e]															
JTT	All	38	22	39	32	89	12	9	89	3	2	88	92	71	1.69
JFT	All	25	20	15	13	96	8	4	83				48	77	1.49
SEM	All	7	5	6	4	88	3	2	100				16	69	1.64
Total		70	47	60	49	91	23	15	89	3	2	88	156	72	

[a]No. ewes mated that had that CL count.

[b]No. ewes lambed.

[c]Percent embryo survival in pregnant ewes.

[d]Percent lambed.

[e]Season.

TABLE 8. EFFECTS OF BREED AND LITTER SIZE ON GESTATION PERIOD (1983)

Breed	No.	Gestation period [a], d	Litter size	No.	Gestation period [b], d
JTT	56	148.5 ± .58[c]	1	52	148.4 ± .33
JFT	38	146.9 ± .57	2	42	148.8 ± .37
SEM	18	147.0 ± .72	3	12	148.6 ± .65
			4	5	146.7 ± 1.01
			5	1	144.6 ± 2.27

[a]Breed effect (P<.01).

[b]Litter size effect (P = .14).

[c]Least-squares mean ± SE.

Inheritance of Ovulation Rate and Litter Size. As indicated in the Introduction, some characteristics of the early data on these sheep raised a question about the possible segregation of a Booroola-type gene (Davis et al., 1982; Piper and Bindon, 1982, 1985). The high repeatabilities for ovulation rate provide support for this hypothesis. The fact that exceptional ewes, in terms of repeated high ovulation rate and litter size, were found in each of three breeds, among which there has no doubt been some opportunity for gene exchange, is more compatible with a "major" gene explanation than with the more usual situation for these traits of low heritability and additivity in crosses.

To provide further evidence relevant to this question, we compared the performance of daughters with that of their dams. The results are presented in table 9. The 81 daughters in this table include 68 JTT and 13 JFT ewes; the pattern of inheritance appeared the same in the two breeds, and they were combined. Taking the criterion of one CL count or litter size ≥3 as diagnostic of carrier ewes (Davis et al., 1982), dams were segregated into two classes. As expected from the high repeatability of ovulation rate and high prenatal survival in these sheep documented earlier, the mean of all observations for ovulation rate and litter size of the high ("≥3") group averaged around 3.0, while the corresponding values for the low ("1, 2") group were about 1.5. Excluding the record, either corpora lutea count or litter size, which first identified a ewe as "high" changed the results very little.

The performance of daughters of these two groups of ewes provides strong support for the hypothesis of segregation of a gene with large effect on prolificacy. Differences between daughter means in table 9 are 39 and 37% of the difference between dam means, for ovulation rate and litter size, respectively. Because the two groups of dams were mated to the same sires, this yields estimates of heritabilities of about .75 for the mean of approximately three records for each trait, clearly much higher than the usual estimates for these traits. Equally pertinent to the question is the fact that the high group of dams produced two distinct groups of daughters, in about equal numbers. The first of these, identified in the table as high on the same basis as their dams (one record ≥3), had a mean performance nearly as high as that of their dams, particularly considering their younger average age. The other group, identified solely on the basis of having no record ≥2, were virtually identical in mean performance to the group containing all daughters of low dams. This suggests that the high dams were heterozygous for the postulated gene, and that their mates did not carry it.

The difference between the two groups of daughters of high dams (table 9) provides a preliminary estimate of the effect of one copy of the postulated gene, i.e., about 1.3 on ovulation rate and .9 on litter size. The difference between the high and low dam groups is somewhat greater, but in good agreement considering that the high dams were slightly older on average and had more mature records than the low dams, and that the high dams may have included at least one homozygote. The difference between the high and low daughter groups of the low dams is also in good agreement, considering the small number of high daughters. Davis et al. (1982) reported a difference in ovulation rate between carriers and non-carriers of the

TABLE 9. OVULATION RATE AND LITTER SIZE OF DAUGHTERS CLASSIFIED BY DAM'S PERFORMANCE

Performance of	High dams (1 or more records ≥3)						Low dams (All records = 1 or 2)					
	Ovulation rate			Litter size			Ovulation rate			Litter size		
	No. ewes	No. records	Mean	No. ewes	No. records	Mean	No. ewes	No. records	Mean	No. ewes	No. records	Mean
All dams[a]	14 (14)[c]	37 (35)	3.14 (3.11)	23 (20)	74 (53)	2.74 (2.53)	27	76	1.45	37	103	1.48
All daughters	41	99	2.04	39	95	1.85	43	114	1.38	42	86	1.38
High daughters[b]	19 (17)	48 (36)	2.73 (2.61)	18 (17)	48 (41)	2.31 (2.15)	3 (2)	7 (5)	2.71 (2.60)	2 (2)	6 (5)	2.83 (2.80)
Low daughters[b]	22	51	1.39	21	47	1.38	40	107	1.29	40	80	1.28

[a]Counting each dam once only, regardless of no. of daughters.

[b]Classified as high or low on the same basis as dams.

[c]Values in parentheses exclude the record that first identified the individual as high.

Booroola gene of 1.24 (2.78 vs 1.54), rather similar to the preliminary estimate from these data.

The implication that the sires used in this flock did not carry the gene merits some comment. Given a frequency of the gene of the order .2, estimated from the dams represented in table 9, one would expect some males to carry the gene. Unfortunately, only five sires had sufficient known daughters to permit a preliminary estimate of their genotype under this hypothesis. Data on daughters of these males are summarized in table 10. The performance of 55 of the 56 daughters of these five rams is consistent with the hypothesis that the latter were all homozygous normal (+ +). The exceptional dam had no ovulation rate data, but had lambed three times without producing more than twins. Although less than one-half of the ewes classified as high on the basis of all ovulation rate and litter size information would have been identified as carriers on the basis of their first litter, all "high" ewes had produced triplets or higher multiples by their third lambing. All four litters of the daughter in question contained three or more lambs, so she is definitely classified as carrying the gene. The dam may, of course, have been a carrier but never produced more than two lambs, due to prenatal loss. There is also the possibility of pedigree error, due to mis-mothering or recording error, although such errors are apparently infrequent in this flock.

TABLE 10. DISTRIBUTION OF HIGH[a] AND LOW[a] DAUGHTERS OF RAMS WITH FIVE OR MORE DAUGHTERS, ACCORDING TO CLASSIFICATION OF DAM

	High dams		Low dams	
	No. daughters		No. daughters	
Sire No.	High	Low	High	Low
100	3	1	0	4
185	4	6	0	5
186	7	6	1[b]	5
300	3	3	0	3
400	1	1	0	3
Total	18	17	1	20

[a]High: one or more records ≥3; Low: all records = 1 or 2.

[b]Dam had three lambings: two singles, one set twins.

The other two high daughters from low dams (table 9) were from dams with three and five ovulation or litter size records each. One daughter was sired by a ram of unknown parentage with only three daughters, the other two being a high daughter from a high dam, and a low daughter from a low dam. The second was one of two daughters (the other a low daughter from a low dam) of a ram whose dam was the ewe with 25 lambs in six lambings. It seems possible that both these rams were heterozygous for the postulated gene, but the evidence suggests that none of the other sires of daughters born in the flock was carrying the gene. This indicates the possibility of selection against carrier males. In view of the heavy emphasis on fighting rams in the Garut strain of Thin Tail sheep, selection of the larger singles seems quite plausible. If single-born animals are favored by breeders of these sheep in their selection of sires, but survival of at least triplets is such that the more prolific ewes wean more lambs, for which there is evidence from village flocks (Inounu et al., 1982), this would represent a model for maintaining the postulated gene at intermediate frequency in populations of these sheep.

The high repeatability of ovulation rate for ewes producing only one or two ovulations, mentioned earlier, is not explained by the hypothesis of a Booroola-type gene producing ovulation rates ≥3 in carriers. A high repeatability is not characteristic of non-carrier segregates from Booroola populations (Owens and Davis, 1985). Examination of daughter-dam pairs within this "1, 2" population did not reveal any clear pattern, as did the analysis of daughters of high and low dams (table 9). It appears that more data will be required to determine the basis of the high repeatability of ovulation rates of 1 and 2 in this population.

Endocrine Studies. The release of LH prior to ovulation was defined in terms of the following variables: 1) interval between the onset of estrus and the beginning of the LH discharge, 2) duration of the LH release, 3) maximum LH concentration observed and 4) the integral of the concentration of LH in excess of 10 ng/ml over the duration of the discharge. The least-squares breed means for these variables, together with the duration of estrus, cycle length and plasma progesterone concentrations on d 10 and 11 after the onset of estrus are given in table 11 for the ewes involved in these aspects of the study.

The duration of estrus was similar for the three breeds and there was no evidence for any association between this trait and the number of ova shed; the regression of duration of estrus on ovulation rate was .7 ± .8 h/CL ($P>.36$). This contrasts with the results of studies with prolific breeds such as the Finnish Landrace and Romanov that have demonstrated a positive and significant relationship between the duration of estrus and ovulation rate or litter size (Land, 1970; Thimonier and Pelletier, 1971; Hanrahan and Quirke, 1975). However, the result is consistent with the finding that Booroola Merino ewes have a duration of estrus that is similar to control Merinos (Bindon et al., 1982). Differences among breeds in estrous cycle length were significant, with JTT ewes having cycles of longer mean duration than either of the other two breeds.

The interval between the onset of estrus and beginning of the pre-ovulatory LH discharge was similar for all three breeds; classification of the ewes according to the number of eggs shed revealed that in the case of the JTT and Semarang breeds, this interval increased only marginally over the range one, two and three or more eggs shed. Analyses in which ovulation rate was used as a covariate, however, revealed a significant linear relationship between the two variables. The pooled, within-breed correlation coefficient for the interval between the onset of estrus and beginning of the LH discharge with ovulation rate was .35, and the corresponding regression coefficient was 1.5 ± .72 h per egg shed ($P<.05$). This regression coefficient is considerably smaller than might be expected on the basis of the results obtained in studies (reviewed by Bindon, 1984) in which the interval between the onset of oestrus and beginning of the LH discharge was measured in Finnish Landrace, Romanov and prolific Ile de France ewes and compared with non-prolific controls. This interval, however, has been shown to be similar in Booroola and control Merinos (Bindon et al., 1984), and the present results indicate that the Javanese sheep are intermediate between the Booroola and other breeds in this respect.

The duration of the LH release and the total LH discharge were similar for the three breeds. Breed differences in the maximum LH concentration observed were significant. Analyses utilizing ovulation rate as a covariate provided no evidence for any significant association between this variable and either the duration of

TABLE 11. LEAST-SQUARES MEANS FOR CYCLE LENGTH, DURATION OF ESTRUS, CHARACTERISTICS OF THE PRE-OVULATORY LH DISCHARGE AND PLASMA PROGESTERONE CONCENTRATIONS

Item	Breed			Error SD
	JTT	JFT	SEM	
Cycle length, d	16.6 (68)[a]	16.3 (45)	16.0 (20)	.8
Duration of estrus, h	33.1 (28)	34.2 (30)	35.7 (18)	7.1
Characteristics of the pre-ovulatory LH discharge				
No. of ewes	24	22	12	
Interval to discharge, h				
Ewes with one ovulation	8.3	4.7	7.5	
Ewes with two ovulations	7.3	8.0	7.8	
Ewes with three or more	9.4	9.7	10.3	
All ewes	8.4	7.5	8.5	4.9
Duration of release, h	11.1	13.3	12.2	2.9
Maximum LH concentration observed, ng/ml	29.0	23.9	27.0	5.2
Total LH discharged, ng/ml	121.1	126.6	120.0	32.4
Plasma progesterone, ng/ml				
D 10 of the estrous cycle	1.20 (53)	1.42 (37)	1.50 (19)	.42
D 11 of the estrous cycle	1.14 (34)	1.27 (29)	1.42 (29)	.47

[a]Number in parentheses represents the number of observations comprising the mean.

the LH release, maximum LH concentration observed, or total LH released; this is in agreement with the results of studies with a wide range of ewe breeds (Bindon, 1984).

Breed differences in plasma progesterone concentration on d 10 of the cycle were significant, with JTT ewes having lower levels than either the JFT or Semarang breeds. Differences among the breeds in progesterone concentration on d 11 of the cycle, however, were not significant. The number of corpora lutea in the ovaries was a significant source of variation in plasma progesterone concentration on both occasions. The mean (± SE) concentrations on d 10 for ewes with one to five corpora lutea were 1.2 ± .43 (N = 44), 1.4 ± .48 (N = 37), 1.4 ± .42 (N = 18), 1.4 ± .26 (N = 7) and 1.9 ± .28 (N = 3), respectively. These values are within the range reported for other ewe breeds (Quirke et al., 1979).

General Discussion

Confirmation of the existence of the postulated gene would represent apparently the fourth case of a gene of this nature described recently. There is the now well-documented Booroola gene (Davis et al., 1982; Piper and Bindon, 1982, 1985; Bindon, 1984); a gene with substantial effect on litter size in Icelandic sheep (Jonmundsson and Adalsteinsson, 1985); the present example; and the Cambridge breed (Hanrahan and Owen, 1985; J. P. Hanrahan, personal communication). Family data on ovulation rate in the Cambridge breed are limited as yet, but the mean, range and repeatability of ovulation rate for the breed clearly represent a pattern different from that in most breeds. The apparent existence of major genes for ovulation rate in several different populations of sheep suggest that this may be a trait affected by genes more subject to mutation than had previously been suspected. The comparative genetics and physiology of these mutations are obviously areas of considerable scientific interest.

With regard to breeding practice, the existence of the postulated gene in Indonesian sheep should provide a relatively rapid means of producing two distinct strains, in one of which ewes would give birth to predominantly singles and twins, at all ages, and in the other to a high proportion of triplets, at least at maturity. Producers could then choose the strain that gave the best returns for their management system. Management systems favoring each of these types appear to exist in Indonesia, and probably in other tropical countries as well.

Literature Cited

Avenell, J. A., Y. Saepudin and I. C. Fletcher. 1985. Concentrations of LH, oestradiol 17β and progesterone in the peripheral plasma of swamp buffalo (Bubalis bubalis) cows around the time of oestrus. J. Reprod. Fertil. 74:419.

Bindon, B. M. 1984. Reproductive biology of the Booroola Merino sheep. Australian J. Biol. Sci. 37:163.

Bindon, B. M., L. R. Piper and R. Evans. 1982. Reproductive biology of the Booroola Merino. In: L. R. Piper, B. M. Bindon and R. D. Nethery (Ed.) The Booroola Merino. Proceedings of a Workshop. p 21. CSIRO, Melbourne, Australia.

Bindon, B. M., L. R. Piper and J. Thimonier. 1984. Preovulatory LH characteristics and time of ovulation in the prolific Booroola Merino ewe. J. Reprod. Fertil. 71:519.

Bradford, G. E., J. F. Quirke and T. R. Famula. 1986. Fertility, embryo survival and litter size in lines of Targhee sheep selected for weaning weight or litter size. J. Anim. Sci. 62:895.

Bradford, G. E., J. F. Quirke, P. Sitorus, I. Inounu, B. Tiesnamurti, F. L. Bell and D. T. Torell. 1983. Genetic basis of prolificacy in Javanese sheep. Working paper No. 25, Small Ruminant CRSP, BPT, Bogor, Indonesia.

Bradford, G. E., St. C. S. Taylor, J. F. Quirke and R. Hart. 1974. An egg transfer study of litter size, birth weight and lamb survival. Anim. Prod. 18:249.

Davis, G. H., G. W. Montgomery, A. J. Allison, R. W. Kelly and R. W. Bray. 1982. Segregation of a major gene influencing fecundity in progeny of Booroola sheep. New Zealand J. Agr. Res. 25:525.

Dzakuma, J. M., J. V. Whiteman and R. W. McNew. 1982. Repeatability of lambing rate. J. Anim. Sci. 54:540.

Fletcher, I. C., I. Chaniago and J. M. Obst. 1982. A comparison of the reproductive performance of Javanese Thin Tail and Border Leicester × Merino ewes in Indonesia. Proc. Australian Soc. Anim. Prod. 14:455.

George, J. M. 1982. The mating potential of Indonesian sheep. Anim. Reprod. Sci. 4:251.

Hanrahan, J. P. 1982. Selection for increased ovulation rate, litter size and embryo survival. Proc. 2nd World Congr. Genetics Applied to Livestock Production V:294.

Hanrahan, J. P. and J. B. Owen. 1985. Variation and repeatability of ovulation rate in Cambridge ewes. Paper No. 38, Proc. British Soc. Anim. Prod.

Hanrahan, J. P. and J. F. Quirke. 1975. Repeatability of the duration of oestrus and ovulation rate of sheep. J. Reprod. Fertil. 45:29.

Hanrahan, J. P. and J. F. Quirke. 1985. Contribution of variation in ovulation rate and embryo survival to within breed variation in litter size. In: R. B. Land and D. W. Robinson (Ed.) Genetics of Reproduction in Sheep. p 193. Butterworths, London.

Harvey, W. R. 1977. User's guide for LSML 76, mixed model least-squares and maximum likelihood computer program. Ohio State Univ., Columbus.

Inounu, I., P. Sitorus, B. Tiesnamurti, I. C. Fletcher and G. E. Bradford. 1985. Reproductive performance of Indonesian sheep on different planes of nutrition. Proc. 3rd Annu. AAAP Congr. (Seoul) 1:414.

Inounu, I., N. Thomas and P. Sitorus. 1982. Lambing characteristics of Javanese Thin Tail sheep. Report, SR-CRSP BPT, Bogor.

Jonmundsson, J. V. and S. Adalsteinsson. 1985. Single genes for fecundity in Icelandic sheep. In: R. B. Land and D. W. Robinson (Ed.) Genetics of Reproduction in Sheep. p 159. Butterworths, London.

Land, R. B. 1970. A relationship between the duration of oestrus, ovulation rate and litter size of sheep. J. Reprod. Fertil. 23:49.

Mason, I. L. 1978. Sheep in Java. World Anim. Rev. 27:17.

Mason, I. L. 1980. Prolific tropical sheep. FAO Anim. Prod. Health Paper No. 17, Rome.

Meyer, H. H. 1985. Breed differences in ovulation rate and uterine efficiency and their contribution to fecundity. In: R. B. Land and D. W. Robinson (Ed.) Genetics of Reproduction in Sheep. p 185. Butterworths, London.

Obst, J. M., T. Boyer and Y. Chaniago. 1980. Reproductive performance of Indonesian sheep and goats. Proc. Australian Soc. Anim. Prod. 13:321.

Owens, J. L. and A. H. Davis. 1985. Repeatability of ovulation rate in Booroola Merino and Merino ewes. New Zealand Ministry of Agr. Fish. Agr. Res. Div. Ann. Rep. 1983-84. Wellington. p 258.

Piper, L. R. and B. M. Bindon. 1982. Genetic segregation for fecundity in Booroola Merino sheep. Proc. World Congr. Sheep and Beef Cattle Breeding 1:395.

Piper, L. R. and B. M. Bindon. 1985. The single gene inheritance of the high litter size of the Booroola Merino. In: R. B. Land and D. W. Robinson (Ed.) Genetics of Reproduction in Sheep. p 115. Butterworths, London.

Quirke, J. F., J. P. Hanrahan and J. P. Gosling. 1979. Plasma progesterone levels throughout the oestrous cycle and release of LH at oestrus in sheep with different ovulation rates. J. Reprod. Fertil. 55:37.

Ricordeau, G., J. Razungles, F. Eychenne and E. Tchamitchian. 1976. Performances de reproduction des brebis Berrichonnes du Cher, Romanov et croisees. II. Composantes de la prolificite. Ann. Genet. Sel. Anim. 8:25.

Ricordeau, G., J. Razungles and D. Lajous. 1982. Heritability of ovulation rate and level of embryonic losses in Romanov breed. Proc. 2nd World Congr. Genetics Applied to Livestock Production VII:591.

Russell, A. J. F., J. M. Doney and R. G. Gunn. 1969. Subjective assessment of body fat in live sheep. J. Agr. Sci. (Camb.) 72:45.

Sitorus, P., Subandriyo and I. Inounu. 1985. A study of some aspects of reproduction of Javanese Thin Tailed and Javanese Fat Tailed sheep. Proc. 3rd Annu. AAAP Congr. (Seoul) 1:435.

Thimonier, J. and J. Pelletier. 1971. Difference genetique dans la decharge ovulante (LH) chez les brebis de race Ile de France; relations avec le nombre d'ovulations. Ann. Biol. Anim. Biochem. Biophys. 11:559.

Turner, N. H. and S. S. Y. Young. 1969. Quantitative Genetics in Sheep Breeding. pp 77–93. Cornell Univ. Press, Ithaca, NY.