

A study of puberty in Romney ewe lambs by oestrus detection, laparoscopy and progesterone assay

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ABSTRACT

Attainment of puberty in Romney ewe lambs was studied at Wye Sheep Research Unit of the Imperial College (University of London) in the U.K. 77% of females exposed to rams during the breeding season reached puberty. Ovulation effectively accompanied first behavioural oestrus. The evidence of silent ovulation before pubertal oestrus and after the last oestrus of the breeding season was established.

Key words: Ewe lamb-Puberty-Ovulation-Laparoscopy-Progesterone.

RESUME

Une étude sur l'avènement de la puberté chez les agnelles Romney a eu lieu à l'unité de recherche des ovins de l'Imperial College (Université de Londres) à Wye au Royaume uni. 77% des agnelles ont atteint la puberté au cours de la saison de reproduction. L'oestrus pubertal était effectivement suivi des ovulations. L'évidence des ovulations avant l'oestrus pubertal et après le dernier oestrus de la saison de reproduction est confirmée.

Mots clés: Agnelle-Puberté-Ovulation-Laparoscopie-Progesterone.

INTRODUCTION

Puberty is difficult to define because of the complexity of the mechanisms of growth, development, occurrence of puberty and maturity. Therefore each author determines this important stage of life largely according to the discipline of interest. The general expression to define puberty is the age at which an animal becomes capable of reproducing sexually. Dyrmondsson (1972) gave a more precise definition of puberty as the time at which reproduction first becomes possible, whereas sexual maturity is not yet reached. For Hafez and Hafez (2000), puberty is basically the result of a gradual adjustment between increasing gonadotropic activity and the ability of the gonads to simultaneously assume steroidogenesis and gametogenesis. It is well established that the intensity of behavioural oestrus is lower in ewe lambs than in yearlings and adult ewes (Keane, 1974; Dyrmondsson, 1978). The apparent oestral behaviour may not be followed by ovulation (Edey et al., 1977). The peripheral progesterone concentration in pubertal ewe lambs had been studied by many authors (Edey et al., 1978; Hare and Bryant, 1982; Boly et al., 1992; Boly et al., 2000). This paper reports on the results of the use of laparoscopy together with the measuring of progesterone concentration in pubertal Romney ewe Lambs.

MATERIALS AND METHODS

-Animals

This study was carried out at the sheep research unit of Imperial College (University of London) at Wye in Kent (UK).

Eighty-one female lambs born in spring 2000 were split into 3 groups when they were 4 months old approximately. The group A (n = 31) was designated for puberal studies. The groups B and C (n = 25 each one) were to be exposed to fertile rams.

All the ewe lambs were weighed and body condition was score on 7 September 2000 and every subsequent 2 weeks until March the following year.

-Oestrus detection

At the beginning of the breeding season (October) a teaser ram equipped with a raddle harness was introduced to each of the 3 groups.

Raddle crayon colours were changed every two weeks in order to distinguish between marks of different

oestrous cycles. The teaser ram was finally removed from group A on 9 March, 21 days after the last observed oestrus.

All the animals were checked every day for raddle mark recording of puberal behaviour. Teaser rams were replaced by fertile ones in groups B and C on 2 November. The mating period lasted 2 weeks, the rams being removed on 30 November when the observations on these animals ended.

The teaser ram was finally removed from group A on 9 March 21 days after the observed last oestrus.

-Blood sampling and progesterone immunoassay

Blood samples were taken twice weekly in group A from 20 October to 4 December and thereafter only ewe lambs that had shown puberal oestrus were blood sampled twice a week until about 17 days after the day of teaser raddle mark. Blood samples were centrifuged and the plasma pipetted off and stored frozen (at -20°C) till analysis. Immunoassay for the measurement of plasma progesterone concentration was carried out using the kit system described in Ovucheck (Cambridge Life Sciences progesterone EIA kit, Instruction Leaflet, 1987).

-Laparoscopic examination

Laparoscopy is a surgical opening of the abdomen to examine the enclosed organs in case of diseases or research operations. Laparoscopy is an alternative procedure for similar purposes. Laparoscopy involves very minor surgery avoiding serious cuts and trauma for the animals (Haresign, 1979). It basically consists of a source of light, a laparoscope with optic fibre for light transmission, a trocar and cannula, a pair of forceps, a probe and Mitchel clips (Kelly, 1979). Its main use in the sheep industry is to examine the reproductive tract of the ewe (Roberts, 1968; Walker et al., 1984) with various objectives: to record the ovulation rates (Quirke, 1978), for ova recovery and embryo transfer (McKelvey and Robinson, 1985; McKelvey et al., 1986), uterine insemination (Killen and Caffery, 1982; Evans, 1984). It was used in this study in order to examine the activity of the ovaries of the ewe lambs at puberty.

Every animal which exhibited puberal oestrus underwent laparoscopy within 5 days following the teaser marking day. Laparoscopy was performed again on 1 April on the 10 ewe lambs marked last in the season.

Table 1: Comparison of puberal and non-puberal ewe lambs.

TRAITS	Puberal (50)	Non-puberal (31)
Dam age (years)	3.91±0.6	3.19±0.2
Birth type	2.09±0.0	1.87±0.0
Birth weight (kg)	4.52±0.1	5.09±0.1*
Weaning weight (kg)	23.77±0.5	23.29±0.8
Growth rate (g/day)		
- from birth to weaning	228.8±5.1	217.2±8.7
- from weaning to joining	205.4±4.9	107.7±5***
- from birth to joining	193.5±3.7	176.0±4.5**
Joining live weight (kg)	41.59±0.6	38.04±1.0**
Body condition score	3.21±0.0	3.03±0.1

* P < 0.05 ** P < 0.01 *** P < 0.001

RESULTS

-Puberty onset proportion during the breeding season

Out of 81 ewe lambs of the 3 groups put to rams 63 (77.8%) reached puberty (first oestrus) during the breeding season over the period from 8 October to 17 February.

The mean date of birth of the animals that exhibited puberal oestrus was 8 April (s.e. 1.2 days). Mean age at first oestrus was 220.5 ± 1.9 days (range 191-246 days) and the mean joining weight in October was 41.6 ± 0.6 kg (range 30-52 kg). The group of ewe lambs that showed oestrus and that of their mates which had not reached puberty by 28 November were compared in certain parameters (Table 1). There were no significant differences between the 2 groups in terms of dam age, birth type, weaning weight, growth rate from birth to weaning and body condition score. However, ewe lambs exhibiting oestrus from 20 October to 28 November were born earlier (P < 0.05) in the season. They were born as multiples and were lighter (P < 0.01) at birth. They did not differ from non-puberal lambs up to weaning in July but grew much faster (P < 0.001) from then to joining in October. Obviously overall growth rates from birth to joining became higher (P < 0.01) and this resulted in a heavier body weight (P < 0.01) at joining in October.

-Frequency of puberal Oestrus throughout the breeding season

The following results on puberty were based on the 31 ewe lambs of group A which were monitored from 8 October to 1 April. Only 17 of them had shown puberal oestrus by 28 November but by 9 March only 2 animals had not reached puberty. The first puberal oestrus of the season was observed on 20 October and the last oestrus was noticed on 17 February. The frequency of puberal oestrus during the season is shown in Figure 1.

A high proportion (65.5%) of puberal ewe lambs showed their first oestrus in November and 83.9% had

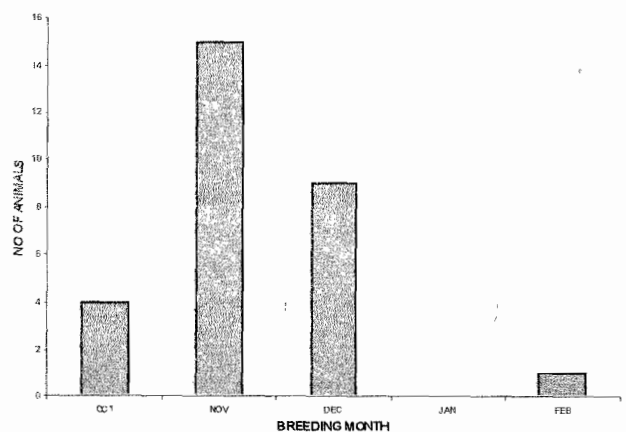


Figure 1: Frequency of puberal oestrus during the breeding season

Table 2: Ovarian examination at puberty by laparoscopy.

TYPE OF OVULATION	N° OF ANIMALS	PERCENTAGE
SINGLE OVULATION	19	70.37
• Right ovary	11	40.74
• Left ovary	8	29.63
DOUBLE OVULATION	6	22.22
• 1 CL on each ovary	3	11.11
• 2 CL on right ovary	2	7.40
• 2 CL on left ovary	1	3.70
TRIPLE OVULATION	2	7.40
• 2 CL on right ovary	1	3.70
• 2 CL on left ovary	1	3.70
TOTAL	27	100.0

^a One CL on opposite ovary

reached puberty by 21 December, the shortest day of the year. The mean October weight of all ewe lambs attaining puberty during this breeding season was 41.0 ± 1 kg (range 30-52 kg). October live weights of the 2 ewe lambs that failed to show oestrus were 38, and 40 kg and increased to 50 and 58 kg, respectively, by 2 March.

-Ovarian activity

Twenty-seven of the 29 puberal ewe lambs marked by the teaser rams underwent laparoscopy within 5.2 ± 0.5 days post-puberal oestrus. There was clear evidence that ovulation effectively accompanied first behavioural oestrus in all the examined animals. Results of laparoscopy showing the distribution of corpora lutea (CL) on ovaries of those 27 ewe lambs are presented in Table 2. Mean ovulation rate was 1.37. There were more CL on right ovaries (26 vs. 16) but the difference failed to reach statistical significance.

Plasma progesterone pattern showed that 19 (65.5%) out of 29 animals that reached puberty had silent ovulation(s) preceding the first exhibited oestrus, as indicated by at least one rise of progesterone concentration to 2.0 ng/ml plasma. The low frequency of blood sampling may not have shown the maximum progesterone concentrations reached. However, mean

peak concentration of progesterone in the luteal phase following these assumed silent ovulations was 5.2 ± 0.6 ng/ml. It was not certain whether they were single or multiple silent ovulations, nor was it easy to determine progesterone rise duration. There was no noticeable rise in plasma progesterone concentration in 4 of the 23 puberal animals before exhibition of their first behavioural oestrus. These probably ovulated for the first time only at their puberal oestrus. Progesterone level remained at a low basal level ($0.25 \text{ ng/ml} < x < 1.80 \text{ ng/ml}$). Moreover, one of the ewe lambs did not show any rise above the basal level for about one month following the puberal oestrus despite the evidence of the presence of three CL at laparoscopy.

The 2 ewe lambs not reaching puberty during the breeding season also showed some rise in plasma progesterone concentration over the period from 20 October to 4 December. The first one had an elevation up to 2.4 ng/ml on 30 October when live weight was 38.5 kg. The second lamb had a rise up to 3.3 ng/ml on 24 November while weighing about 40 kg.

Ovarian examinations using laparoscopy were made on the 10 latest oestrous ewe lambs in the breeding season on 1 April within 58.8 ± 2.5 days after their last oestrous behaviour as indicated by raddle marks, 23 days after teaser ram removal (Table 3). One ewe lamb

Table 3: Ovarian examination at the end of the breeding season

ANIMAL IDENTITY	NO. OF DAYS AFTER LAST OBSERVED OESTRUS	NO. OF CL ON OVARY		REMARKS
		Right	Left	
1	60	2	0	Bright red
2	58	0	0	No recent activity
3	66	0	0	Recent activity
4	60	0	2	
5	71	1	0	Recent activity on left Very fat/not seen
6	51	-	-	No recent activity
7	58	0	0	No recent activity
8	43	0	0	No recent activity
9	65	1	0	
10	58	0	0	Recent activity

could not be examined comfortably due to considerable fat depot around the ovaries. Three (30%) only out of these 10 animals did not show any evidence of ovarian activity after last oestrus. Therefore their breeding season ended as revealed by their loss of interest in the teaser ram. Two others did not ovulate but the morphological aspect of their ovaries clearly showed signs of recent activity, i.e. there had been some silent ovulation(s) after the last exhibited oestrus of the season. Likewise, the remaining 4 ewe lambs still cycling by 1 April as evidenced by presence of CL on one ovary, obviously experienced silent ovulation after their exhibited oestrus.

DISCUSSION

A percentage of 77.8% of the animals reached puberty. Mean September weight of puberal animals was 41.0 ± 0.6 kg in the range of 30 to 52 kg. The mean age at puberty was 225 days. The mean age and body weight at puberty found in this study are similar to the findings obtained with Galway, Finnish Landrace and crossbred ewe lambs by Quirke (1979). The results of Hafez (1953) with Romney ewe lambs stressing a minimum threshold weight as an important factor timing ovulation are in support of this study (30 kg). The heaviest ewe lamb was 52 kg. Some ewe lambs exhib-

iting their puberal oestrus late in the season (10) were so old that the mean age at puberty as a whole was increased to 225 days. The lambs that reached puberty were significantly different from those that did not in respect to several variables (Table 1). The first puberal oestrus was detected on 20 October and 83.8% of the lambs reached puberty before 21 December. Most of them exhibited their puberal oestrus in November (Figure 3). A total of 93% of the puberal ewe lambs recorded according to the teaser ram marks were examined through laparoscopy. All of these (27) had effectively ovulated as indicated by the presence of CL on their ovaries. This was not in agreement with the results of previous studies (Edey et al., 1977) which showed that 6 of the puberal ewe lambs did not ovulate following their first behavioural oestrus. The ovulation rate of 1.37 in this study was similar to that of Galway cross ewe lambs under natural conditions in Ireland observed by Quirke (1978). Low fertility in ewe lambs had been noticed in other studies (Hassan et al., 1994). Investigations on peripheral progesterone concentrations in 23 puberal ewe lambs showed that 65.5% of these had silent ovulations preceding the first exhibited puberal oestrus. These results are in agreement with several earlier findings (Edey et al., 1978; Foote et al., 1970; Foster and Karsch, 1971; Foster and Ryan, 1979;

Hare and Bryan, 1982). The fact that 4 animals ovulated for the first time following their puberal oestrus is in agreement with the reports of Edey et al. (1978). The 2 ewe lambs which remained unmarked throughout the breeding season were not examined through laparoscopy. However, the study of the progesterone concentrations showed that both ovulated before the end of November. Such a case had been reported by Hare and Bryant (1982). It was possible that the ram failed to detect oestrus in these 2 lambs when they were in heat. They could also have been mounted without the marks being detected.

The respective mean dates of first and last oestrus of 9 November and 21 January are close to those reported by several authors (Dyrmundsson and Lees, 1972; Hare and Bryant, 1982; Keane, 1976). The proportion of 0.4 experiencing silent ovulation after the last oestrus of the breeding season is close to 0.53 reported in Rambouillet ewe lambs by Foote et al (1970).

The analysis of the variation in the date of puberal oestrus showed that joining live weight and age at puberty significantly affected the date of first behavioral oestrus which is in agreement with other studies (Dyrmundsson, 1973).

CONCLUSION

It appears that a high percentage (77%) of Romney ewe lambs born to mature dams early in the lambing season (April) attain puberty in late October - early November with heavier body weight and at a lower age. There is a clear evidence of oestrous activity in Romney females at that stage of life.

The techniques of laparoscopy and progesterone assay helped to conciliate the external behaviour at the attainment of puberty and the real activity of the internal reproductive organs. These techniques can be used in many research domains especially in genetic programmes as to check on ovulation rate and furthermore, the number of foetuses carried in preselected females.

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