The Effect of Superovulation of Javanese Thin-Tail Ewes Prior to Mating on Lamb Birth Weight and Preweaning Growth

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ABSTRACT : Forty-four Javanese thin-tail ewes were used to study the effect of superovulation prior to mating and of ration quality on lamb birth weight at parturition. Twenty-two ewes weaning at least one lamb were used to measure lamb preweaning growth. Prior to mating, the experimental ewes were injected twice with prostaglandin, with an 11 d interval between injections to synchronize the estrous cycle. At the last prostaglandin injection, 24 ewes were also injected with 700 IU of pregnant mare serum gonadotropin (PMSG) to stimulate superovulation and the remainder were injected with saline as control. During pregnancy and lactation, the experimental ewes were fed either on a low (12% CP and 65% TDN) or a high (15% CP and 75% TDN) quality ration. During lactation, the milk was collected twice a day and was refed to the lambs by bottle feeding immediately after collection. Superovulation or ration quality as a main factor did not significantly affect lamb birth weight. Litter size significantly affected lamb birth weight ($p<0.05$), and there was an interaction of superovulation and litter size. Nonsuperovulated ewes giving birth to multiple lambs had significantly lower average lamb birth weight (1.34 kg) as compared to those giving birth to a single lamb (1.97 kg) ($p<0.05$). However, superovulated ewes giving birth to multiple lambs had no significant difference in average lamb birth weight (1.68 kg) as compared to those giving birth to a single lamb (1.91 kg) ($p>0.05$). Superovulation of ewes prior to mating resulted in a significant improvement in lamb birth weight in the multiple litter size, without significant effect on average preweaning daily gain ($p=0.07$). Superovulation had a promising use in improving animal production through improvement of prenatal growth during pregnancy and milk production during lactation. (*Asian-Aus. J. Anim. Sci. 2000. Vol. 13, No. 3 : 292-299*)

Key Words : Superovulation, Lamb Birth Weight, Preweaning Growth, Weaning Weight

INTRODUCTION

Birth weight and postnatal life of many domestic animals are profoundly affected by prenatal growth conditions (Dziuk, 1992). Growth and development of the embryo during the embryonic stage of pregnancy are influenced by uterine factors (Denker, 1994), including biochemical changes before implantation initiated by estradiol, and by the processes initiated by progesterone post ovulation (Mulholland et al., 1994) and probably by other hormones and growth factors secreted by the corpus luteum during the luteal phase of the estrous cycle.

Growth of the fetus during the fetal stage of pregnancy is affected by placental growth and development (Schoknecht et al., 1991; Robinson et al., 1995), nutrient availability in the maternal, placental and the fetal circulations (Harding and Johnston, 1995; Hay, Jr., 1995) and the endocrine status of the mother and the fetus (Fowden, 1995).

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The endocrine status of pregnant animals prior to implantation is largely dictated by the corpus luteum and is represented by progesterone which plays a key role in initiating a cascade of uterine growth and secretions (Knight et al., 1977; Mulholland et al., 1994). Administration of progesterone in the early pregnancy in sheep and cattle increases fetal weight (Garrett et al., 1988; Kleemann et al., 1994). Ewes with higher mean serum progesterone during pregnancy give birth to heavier lambs at parturition (Manalu and Sumaryadi, 1998d).

Superovulation prior to mating in sheep dramatically increases maternal serum progesterone concentration which is associated with the increase in uterine and placental weight and individual fetal weight (Manalu et al., 1998). During the embryonic stage of pregnancy, the level of maternal progesterone highly correlates with uterine and fetal growth (Manalu, 1999).

Placental weight positively correlates with placental lactogen concentration and fetal weight (Schoknecht et al., 1991), and estrogen and progesterone concentration in the bovine (Rasby et al., 1990). Placental lactogen and insulin-like growth factor have a positive correlation with fetal size at the end of pregnancy in sheep (Gluckman and Barry, 1988). The objective of this present experiment was to study the effect of superovulation prior to mating on lamb birth weight and postnatal growth of the lamb bottle-fed twice a day with their ewes milk.

MATERIALS AND METHODS

Animals and environmental conditions

This experiment was conducted during the hot (25 to 32℃) and wet (70 to 80% relative humidity) season in the humid tropics of Indonesia. Experimental animals were Javanese thin-tail ewe lambs with live weights ranging from 12 to 15 kg, and from 1 to 1.5 years of age at breeding. The Javanese thin-tail sheep is a meat-type indigenous breed.

Prior to the experiment, the ewes had been raised in a semigrazing system i.e., they grazed in the field during the day and were housed at night, without concentrate supplementation. During the experiment, the ewes were maintained in experimental pens and fed with a complete mixed ration provided as pellet. The experimental ewes were adapted to the experimental conditions and rations for 2 months prior to commencement of treatment.

Experimental design and protocol

Eighty ewes were used at the commencement of the experiment and at the time of breeding. Forty ewes were fed at a low quality ration (12% CP and 65% TDN) and the others (forty ewes) were fed at a high quality ration (15% CP and 75% TDN) during the adaptation period and throughout the experiment. The composition and a chemical analysis of the experimental rations are presented in table 1.

Table 1. Composition and chemical analysis of the experimental rations

Constituent	Low quality ration (12% CP & 65% TDN)	High quality ration (15% CP & 75% TDN)
Dry elephant grass, %	49.21	19.67
Corn, %	27.29	35.07
Ricebran, %	12.27	10.01
Soybean meal, %	5.13	14.44
Coconut meal, %	5.62	19.95
Bone meal, %	0.15	0.19
Fish meal, %	0.15	0.49
Premix, %	0.18	0.18
Total	100.00	100.00
Crude protein, %	12.12	15.20
Crude fiber, %	15.91	11.88
Calculated TDN, %	65.00	75.00
Ether extract, %	5.51	5.80
Nitrogen-free extract, %	43.75	43.71
Ash, %	9.48	9.50
Calcium, %	0.88	1.02
Phosphorus, %	0.61	0.73
Gross energy, MJ/kg	17.41	16.13

At the end of adaptation period, to synchronize the estrous cycle, all ewes were injected twice with 7.5 mg PGF_2 (i.m), the injections being 11 days apart. Forty of the experimental ewes (equal proportion of low and high quality rations) were injected with 700 IU PMSG (Folligon, Intervet, North Holland) at the time of the last prostaglandin injection. The remainder were injected with saline as a control (nonsuperovulated ewes). Two days after the last prostaglandin injections, and at the onset of estrous cycle, the experimental ewes were mated as a group in their respective group (with 1 ram for 3 ewes). The experimental ewes were maintained in their respective feeding group throughout pregnancy. In the week of predicted parturition the experimental ewes were maintained in individual pens.

Live weight was determined at breeding. At parturition, the number of lambs born and their weights were determined.

During gestation, the experimental ewes were fed twice daily. Feed intake was restricted in early pregnancy; during the first 7 weeks the ewes were fed at maintenance level (average consumption of 0.4 kg/d), and feed on offer was increased gradually until week 15 of pregnancy when average consumption was 0.56 kg/d. From week 15 to parturition, the ewes were fed *ad libitum* and average intake was 1.0 kg/d. During lactation, the experimental ewes were fed twice a day and the ewes had free access to feed and water.

During the first week postpartum, milk was not collected, and the ewes were allowed to nurse their respective lambs. One week after parturition, milk was collected twice daily for 84 days postpartum by hand milking with a prior injection of 3 IU oxytocin (i.m.), and the collected milk from each ewe was fed to her lamb immediately after milking. Feeding of the collected milk to the lambs in this particular experiment was decided only once after each milking. Unconsumed milk in a single bottle feeding was not refed to maintain the freshness of the milk. Any unconsumed milk after each milking was discarded or fed to orphaned lambs in the flock. Actually, the amount of milk produced by the ewe in each milking was not all consumed by the lamb at once in a single bottle feeding. The amount of milk consumed by the lamb was measured. Lamb live weight was determined weekly during 84 days postpartum.

Statistical analyses

The lamb birth weight data were analyzed in a randomized design with a $2\times2\times2$ factorial arrangement (unequal n). The first factor was ration quality (low and high quality ration). The second factor was superovulation (nonsuperovulation and superovulation). The third factor was litter size (single and multiple).

Table 2. Mean (±SE) lamb birth weight of lambs from the control and superovulated ewes fed at a low or high quality ration

Variables	Ration quality				Level of significance		
	Low		High				
	Control	Super-ovulation	Control	Super-ovulation	Super-ovulation	Ration quality	Inter-action
Ewe weight at breeding (kg)	12.63±0.68	15.13±1.16	13.39±0.55	13.63±0.62	ns	ns	ns
Lamb birth weight (kg)							
Single	1.78±0.12[a]	1.83±0.19[a]	2.17±0.29[a]	1.94±0.12[a]	ns	ns	ns
Multiple[1]							
Total litter weight	2.67±0.29	4.20±0.27	2.70±1.20	3.11±0.23	*	ns	ns
Average birth weight	1.33±0.15[b]	1.74±0.20[a]	1.35±0.60[b]	1.64±0.11[a]	*	ns	ns

[1] Includes both twin and triplet litters.

[a,b] Different superscripts in the same column (between birth weight of singleton and average weight of multiple) refer to significant effect of interaction of superovulation and litter size ($p<0.05$).

* $p<0.05$; ** $p<0.01$; ns=nonsignificant.

The preweaning to weaning data were analyzed in a randomized design with a 2×2 factorial arrangement (unequal n), since the number of twin litter size was not enough in each cell. For the twin ewes, the weaning weight was average of both lambs, and used in the data analysis. The first factor was ration quality (low and high quality ration). The second factor was superovulation (nonsuperovulation and superovulation). The effects of main factors and their respective interactions were tested using GLM procedure (SAS, 1985).

RESULTS

At parturition, of 40 nonsuperovulated ewes only 20 ewes (9 low quality [5 single and 4 twin bearing], and 11 high quality ration [9 single and 2 twin bearing]) were pregnant and gave birth during the week of predicted parturition. Of 40 superovulated ewes only 24 ewes (9 low quality [5 single and 4 twin bearing], and 15 high quality ration [9 single and 6 twin bearing]) were pregnant and gave birth during the week of predicted parturition. The remainder of the ewes were nonpregnant. Of the superovulated ewes fed on the low quality ration, 4 gave birth to multiple lambs, 2 sets of twins, and 2 sets of triplets.

Of forty-four ewes giving birth, 14 ewes were slaughtered at parturition to measure mammary gland growth and development. Therefore, 30 ewes remained for the study of lactation, with only 22 ewes (5, 4, 6, 7 nonsuperovulated and superovulated ewes fed the low quality ration, nonsuperovulated and superovulated ewes fed the high quality ration, respectively) succeeded in weaning at least one lamb at day 84 postpartum.

The 22 ewes were used in the evaluation of lamb preweaning growth. Of 5 ewes in the nonsuperovulated ewes fed the a low quality ration, 3 ewes gave birth to and weaned a single lamb; 2 ewes gave birth to twins, but only 1 ewe succeeded in weaning two lambs, the other lost one of her lambs during the first week of lactation. Of four ewes in the superovulated ewes fed a low quality ration, 3 ewes gave birth to and weaned a single lamb, and 1 ewe gave birth to twins, but weaned a single lamb at weaning. Of six ewes in the nonsuperovulated ewes fed a high quality ration, 5 ewes gave birth to and weaned a single lamb, and 1 ewe gave birth to and weaned twin lambs. Of seven ewes in the superovulated ewes fed a high quality ration, 4 ewes gave birth to and weaned a single lamb, and 3 ewes gave birth to twin, but only 1 ewe weaned twin lambs. The others weaned a single lamb. Therefore, the number of lambs surviving to weaning per ewe was generally a singleton, except one twin in the superovulated ewes fed a low quality ration, nonsuper- ovulated ewes fed a high quality ration, and super- ovulated ewes fed a high quality ration, respectively. Live weights of twin lambs were averaged for each ewe and treated as a single litter size. The effect of litter size on lamb preweaning growth and weaning weight could not be tested. Sexual difference in the surviving lambs to weaning could not be tested, since most of the surviving lambs were male.

Live weight of the experimental ewes at breeding was not significantly different among treatments (table 2). Therefore, any difference in lamb birth weight was not associated with the difference in the ewe live weight.

In general, superovulation or ration quality did not significantly influence lamb birth weight, regardless of litter size. Multiple lambs had significantly lower birth weight than a singleton, regardless of superovulation and ration quality (1.93 and 1.55 kg in the single and multiple lambs, respectively). When litter size was included as a factor, there was no significant effect of

Table 3. Mean (±SE) lamb birth weight, preweaning growth and weaning weight of lambs surviving to weaning from the control and superovulated ewes fed at low or high quality ration

Variables	Ration quality				Level of significance		
	Low		High				
	Control	Super-ovulation	Control	Super-ovulation	Super-ovulation	Ration quality	Inter-action
Lamb birth weight (kg)[1]	1.84± 0.14	1.76± 0.22	2.13± 0.26	1.77± 0.15	ns	ns	ns
Lamb weight at 84 d (kg)[2]	7.68± 0.50	10.06± 1.48	8.01± 1.18	9.44± 0.77	ns	ns	ns
Average daily gain (g)	69.52± 7.38	98.81±15.71	70.00±12.61	91.31±10.60	0.07	ns	ns
Milk consumption (liter/lamb/84 d)	21.63± 0.25	24.05± 1.82	22.16± 0.40	24.58± 0.65	**	ns	ns
Milk consumption (ml/lamb/d)	257.50± 2.98	286.31±21.67	263.81± 4.76	292.62± 7.74	**	ns	ns
Milk production (ml/ewe/d)	261.03±17.14	423.19±21.54	312.28± 9.03	469.54±22.31	**	ns	ns

[1] Birth weight of 22 of 40 born lambs surviving to weaning, regardless of litter size.
[2] The number of lambs surviving to weaning per ewe was generally a singleton, except one twin in the nonsuperovulated ewes fed a low quality ration, nonsuperovulates and superovulated ewes fed a high quality ration, respectively.
* $p<0.05$; ** $p<0.01$; ns=nonsignificant.

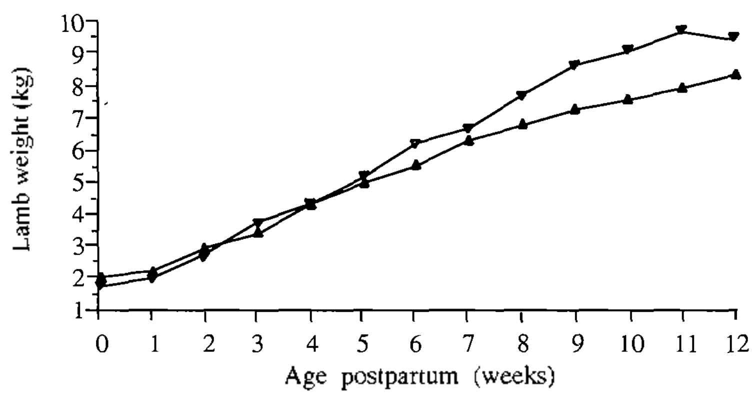

Figure 1. Weekly lamb live weight of the nonsuperovulated (▲) and superovulated (▼) ewes during 84 d postpartum. Standard error of weekly lamb live weight ranged from 0.16 to 0.71 kg in the nonsuperovulated ewes, and from 0.12 to 0.67 kg in the superovulated ewes.

superovulation and ration quality on lamb birth weight of a single lamb (table 2). In the ewes giving birth to a single lamb, lamb birth weights of nonsuperovulated and superovulated ewes were similar (1.97 vs 1.91 kg). Superovulated ewes giving birth to multiple lambs, however, had higher lamb birth weight than nonsuperovulated ewes with the same litter size ($p<0.05$). Mean lamb birth weights in the nonsuperovulated and superovulated ewes giving birth to twin lambs were 1.34 and 1.68 kg, respectively. Superovulated ewes giving birth to multiple lambs had average lamb birth weight similar to nonsuperovulated and superovulated ewes giving birth to a single lamb (1.68 vs 1.97 kg) ($p>0.05$).

Of the 22 ewes weaning at least one lamb at day 84 postpartum, superovulation and ration quality did not affect lamb birth and weaning weights (table 3). The weekly live weights of lambs reared by nonsuperovulated and superovulated ewes is presented in figure 1. During the first 5 weeks postpartum, growth of lambs from nonsuperovulated and superovulated ewes was similar. However, from week 6 to weaning, superovulated ewes tended to have higher lamb live weights than the lambs of nonsuperovulated ewes. Average daily gain of the lamb in the superovulated ewes during 84 days postpartum did not significantly different from that of nonsuperovulated ewes ($p=0.07$).

Average daily gain of the lambs from the superovulated ewes was 98.81 and 91.31 g in those fed a low and a high quality ration, respectively, as compared to 69.52 and 70.0 g in the nonsuperovulated ewes fed a low and a high quality ration, respectively (table 3). In contrast, lamb live weights of the ewes fed a low and a high quality ration were similar during 84 days postpartum (figure 2).

Milk consumption of the lambs during the 84 days is presented in table 3. Total milk consumption of lambs in the superovulated ewes during 84 days postpartum was significantly higher than that of nonsuperovulated ewes ($p<0.01$).

As a comparison, daily milk production of the

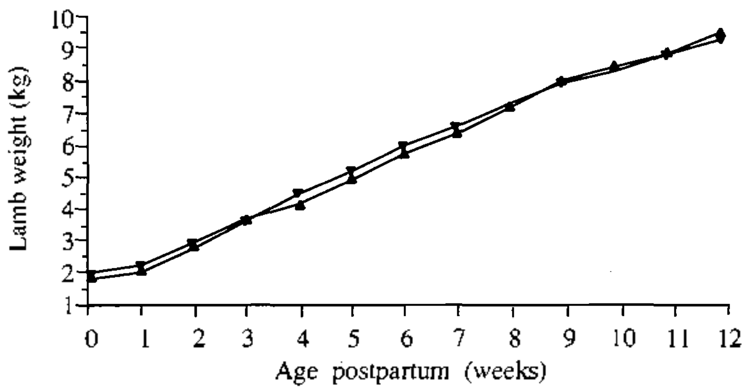

Figure 2. Weekly lamb live weight of the ewes fed low (▲) and high (▼) planes of nutrition during 84 d postpartum. Standard error of weekly lamb live weight ranged from 0.13 to 0.91 kg in the nonsuperovulated ewes, and from 0.06 to 0.69 kg in the superovulated ewes.

experimental ewes is presented (table 3). In the nonsuperovulated ewes, the amount of milk produced was only enough to meet the daily consumption of a single lamb, especially after week 5 postpartum. In the superovulated ewes, as a contrast, the amount of milk produced was enough to meet daily consumption of 1.5 lambs. The profile of milk production and milk consumption in the experimental lambs is presented in figure 3. From milk production and milk consumption profiles in figure 3, during the first 5 weeks postpartum, milk consumption was lower than milk production in the nonsuperovulated and superovulated ewes, and in the ewes fed low and high quality rations. However, the difference was greater in the superovulated ewes and the ewes fed a high quality ration. After week 5 postpartum, in the nonsuperovulated ewes, milk production and milk consumption decreased to a low level, while superovulated ewes still produced milk more than was consumed by the lambs. Profiles of milk production by the ewes and milk consumption by the lambs similar in the ewes fed a low and a high quality ration.

DISCUSSION

The results of the experiment indicated that superovulation of ewes prior to mating improves prenatal growth of multiple fetuses as shown by the increased birth weight.

Previous studies have indicated that ewes with higher progesterone concentrations during pregnancy have heavier lambs at parturition (Manalu and Sumaryadi, 1998d; Manalu and Sumaryadi, 1999). Superovulation of ewes prior to mating increases the number of corpora lutea, maternal progesterone concentrations, uterine and fetal weight during early pregnancy (Manalu et al., 1998). The increased number of corpora lutea and maternal serum progesterone concentrations in the superovulated ewes is associated with an increase in uterine and fetal weight during pregnancy in sheep. The level of maternal progesterone concentrations during pregnancy highly correlates with uterine, placental, and fetal growth (Manalu, 1999). In addition, placental weight positively correlates with placental lactogen concentration and fetal weight (Schoknecht et al., 1991) and estrogen and progesterone concentration (Rasby et al., 1990). Other studies report that placenta lactogen and insulin-like growth factor have positive correlations with fetal size in the end of pregnancy in sheep (Gluckman and Barry, 1988). The effect of superovulation on uterine and fetal growth could be partly associated with the increased secretory activities of the uterine gland and the size of the placenta due to the increased hormonal stimulation produced by the corpus luteum, and estrogen and progesterone are reported to change secretory activity of the uterine glandular epithelium (Murray, 1992).

The nonsignificant difference in lamb birth weight of superovulated and superovulated ewes, especially in the single litter size was probably related, partly, to the space limitation of the placental growth (Schoknecht et al., 1991; Robinson et al., 1995) in this small size of sheep, since the fetuses and placenta of superovulated ewes in the same breed, with a similar weight and age, were heavier than those of nonsuperovulated ewes until week 15 of pregnancy, regardless of litter size (Manalu et al., 1998). The effect of nutrient availability in the maternal circulation (Harding and Johnston, 1995; Hay, 1995) could probably be eliminated since the ration with 12% CP and 65% TDN had similar effect on lamb birth weight to that with 15% CP and 75% TDN. Furthermore, the maximum growth of the fetus in this particular age, weight, and breed of sheep was probably achieved. In the lambs raised until weaning, the growth of the lambs of nonsuperovulated and superovulated ewes was similar until age 5 weeks postpartum. Thereafter, the

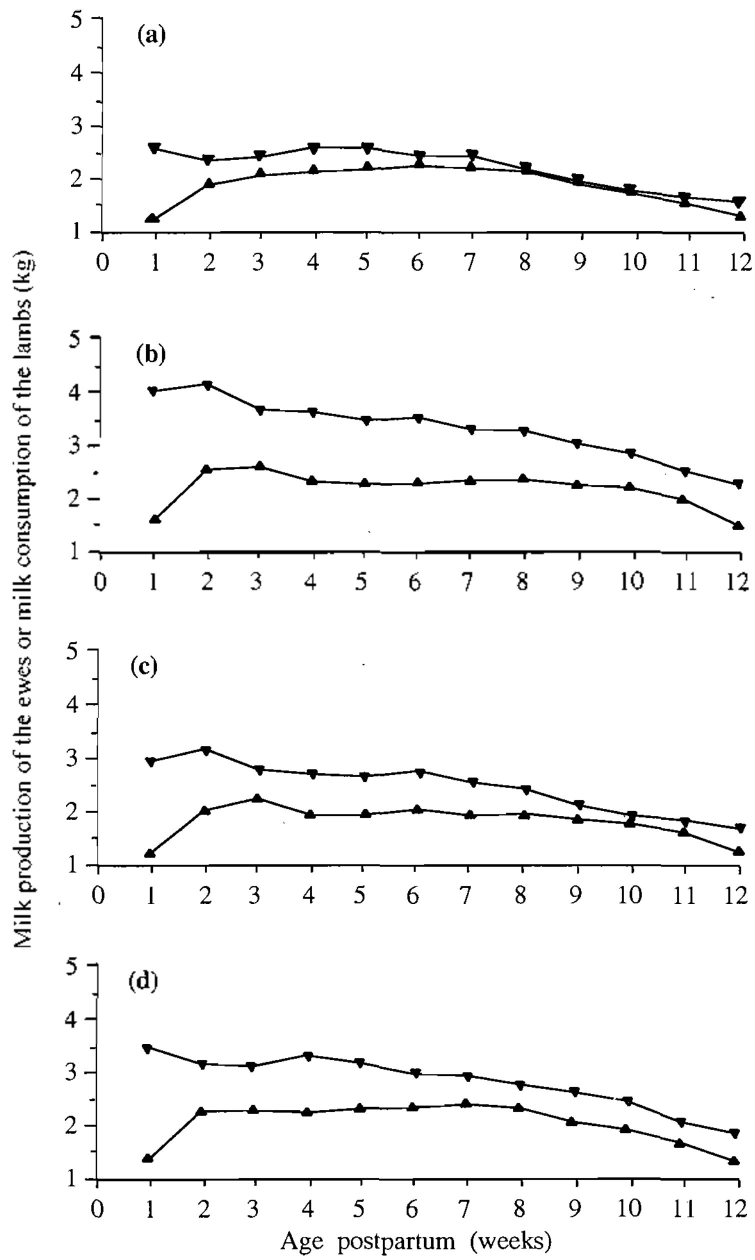

Figure 3. Pattern of weekly milk production of the ewes (▼) and weekly milk consumption of their lambs (▲) in the (a) nonsuperovulated ewes, (b) superovulated ewes, (c) ewes fed at a low quality ration, and (d) ewes fed at a high quality ration in Javanese thin-tail sheep. Standard error of weekly milk production ranged from 0.1 to 0.39 kg in the nonsuperovulated, and from 0.1 to 0.4 kg in the superovulated ewes, and from 0.1 to 0.36 kg in the ewes fed on low quality ration, from 0.1 to 0.4 kg in the ewes fed a high quality ration. Standard error of weekly milk consumption ranged from 0.04 to 0.06 kg in the nonsuperovulated, and from 0.05 to 0.13 kg in the superovulated ewes, and from 0.06 to 0.15 kg in the ewes fed on low quality ration, from 0.06 to 0.09 kg in the ewes fed a high quality ration.

lambs of superovulated ewes had greater average daily gain than those of nonsuperovulated ewes. From milk production and milk consumption profiles in figure 3, apparently the pattern of milk production of the ewes and milk consumption of the lambs was comparable with the lamb preweaning growth. The pattern of milk production of the ewes and milk consumption of the lambs in the ewes fed a low quality ration was similar to that of ewes fed a high quality ration, giving a similar pattern of preweaning growth. The pattern of milk production of the ewes and milk consumption of the lambs in the superovulated ewes, however, was different than that of nonsuperovulated ewes, and the lamb growth curve was different especially after week 8 postpartum. These data indicated that when the lambs consumed all milk produced by their ewes, the difference would be greater. However, in this particular experiment, the lambs were fed with their ewes milk twice a day after each milking.

The data of this limited experiment indicated that with a similar starting birth weight, average daily gain of the lambs of superovulated ewes was far greater than that of nonsuperovulated ewes even though weaning weight was not significant. Therefore, the effect of superovulation on preweaning weight of the lambs could occur in two ways, one through the effect on prenatal growth during pregnancy, and the other through the effect on mammary growth during pregnancy and milk production during lactation. Ewes with higher progesterone concentrations during pregnancy have better developed mammary glands at parturition (Manalu and Sumaryadi, 1998a,b,c). Superovulated ewes have better mammary growth during pregnancy (Manalu et al., 199b), and higher milk production during lactation has been reported elsewhere (Frimawaty and Manalu, 1999; Manalu et al., 1999a). The effect of superovulation on fetal growth during pregnancy was evident (Manalu et al., 1998). However, the effects of superovulation on the lamb birth weight and preweaning growth merit further study with a larger experimental unit. If the size of experimental unit was large enough to allow separation of single from multiple litter size in the analysis, the effect of superovulation on birth weight, preweaning growth, and weaning weight might be more evident. In addition, if the lambs were allowed to suckle their mothers all times, or if all milk harvested was consumed by the lambs, the effect of higher milk production in the superovulated ewes on the lamb preweaning growth would be more dramatic. Nevertheless, this preliminary information could stimulate other studies on the potential use of superovulation in improving animal production, especially in multiparous mammals. In future experiments the effect of superovulation prior to mating on the preweaning growth of the offspring should be studied in other species of mammals, especially multiparous animals such as swine.

CONCLUSION

Superovulation of ewes prior to mating significantly improved lamb birth weight in multiple litters. Superovulation of ewes prior to mating tended to increase lamb average preweaning daily gain and weaning weight even though lamb milk consumption was far below milk production. Superovulation promises to improve animal production through improvement of prenatal growth during pregnancy and milk production during lactation.

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