

AN ABSTRACT OF THE THESIS OF

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Title: HORMONAL STIMULATION OF BREEDING ACTIVITY IN  
DIFFERENT GENETIC GROUPS OF EWE LAMBS

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An investigation of the effects of exogenous gonadotropic treatments on fertility of various genetic groups of ewe lambs were run over a six-year period. Since hormonal treatments varied and the experimental animals were different from one breeding season to the next, each year comprised a separate study. Hormone treatment included the use of Pregnant Mare Serum, progesterone, human chorionic gonadotropin, 17- $\beta$ -estradiol and crude pituitary extracts on predominantly Willamette and Suffolk ewe lambs.

In general, treatment of ewe lambs with various gonadotropins did not increase fertility. The Suffolk lambs, in fact, were adversely affected in that either treatment per se or the handling necessary for treatment caused lower fertility in most cases.

In the 1969 study, histological examination of the reproductive tracts of treated and control ewe lambs were made. No differences

were observed among breeds or between the various treatment and control groups. Pituitary glands were biologically assayed for LH content. Lutenizing hormone was present in measurable quantities in all pituitary glands assayed.

The ewe lambs from a three breed diallel cross were the experimental animals utilized for the 1970 phase of the study. Treatment consisted of three injections of pituitary extract at two-week intervals. Variables measured included ewe birth weight, ewes weight at mating, ewe lamb fertility and lamb birth weight. Heterosis was found to be highly significant for ewe weight at mating and ewe lamb fertility ( $P < 0.01$ ). This significance was not due to a high positive correlation between the variables. It is due to true heterotic effects. General combining ability was also significant for fertility and ewe weight at mating. Reciprocal effects and breed differences accounted for 31 and 23 percent of the total variation in ewe weight at mating. Fertility of the ewe lambs was not effected by treatment.

Hormonal Stimulation of Breeding Activity in  
Different Genetic Groups of Ewe Lambs

by

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# HORMONAL STIMULATION OF BREEDING ACTIVITY IN DIFFERENT GENETIC GROUPS OF EWE LAMBS

## INTRODUCTION

As commercial industry and population growth continue to compete for agricultural land in the United States, the first to suffer will be the livestock industry. The sheep industry, in particular, has experienced declining numbers due to the fact that income from wool and lamb have not offset increasing operating expenses. Means of alleviating the problem are few and include 1) utilizing less desirable range land for production since sheep adapt to it well, 2) finding means of increasing lamb production.

A primary goal of the sheep industry is to produce as many strong, viable lambs from a ewe as possible. It is feasible to conclude that the productive life of a ewe can be extended at least one year if that ewe can be bred to carry a lamb during the first year of her life. However, it is common practice for sheepmen to carry ewes over until the second season and breed them to lamb first at two years of age. It is felt that immature breeding may have adverse effects on future production performance and growth of the ewe. There is evidence to indicate that future production is not affected but final mature size may be.

It is the purpose of the present study to examine the

reproductive performance of the ewe lamb. An attempt was made to determine what effects exogenous gonadotropin, breed of ewe, and other factors may have on reproductive performance.

## LITERATURE REVIEW

An appreciable amount of research has been done on the use of exogenous gonadotropins in stimulating breeding activity in anestrus ewes. However, relatively little work has been done recently on inducing fertility in ewe lambs. Bowstead (1930) reported that the mating of mature males with immature females is believed by most breeders to cause serious deleterious effects on a ewe's future performance. Some of these adverse effects are: (1) reduced mature size of dam and offspring, (2) reduced vitality of offspring, (3) increased difficulties at parturition, (4) decreased future reproductive performance, (5) accumulation of effects if immature mating is practiced over more than one generation. In favor of breeding ewe lambs, Bowstead found that females bred as ewe lambs produced better as two and three year olds than did ewes that were bred first as yearlings. Also, whether lambs from ewe lambs gained as well during the suckling period as wether lambs from yearling mothers. These results tend to disprove some of his initial statements in that production may not be adversely affected by early breeding. Spencer, et al. (1942) found that ewes conceiving as lambs were lighter at their second mating than ewes unmated as lambs. The difference reported as 18.2 pounds was highly significant. These differences disappeared by the third, fourth and fifth years, and are in agreement with results

reported by Bowstead (1930) and Briggs (1936).

Spencer et al. (1942), Bowstead (1930) and Briggs (1936) report that ewe lambs that conceive during their first season tend to be larger and better developed than ewes that do not conceive. This indicates earlier sexual maturity and possibly a higher level of fertility. Spencer followed production over a five year period and found that lamb-bred ewes produced 2.572 more pounds of lamb than ewes bred first as yearlings. Over six years, Briggs (1936) found that lamb-bred ewes produced an average of 0.69 lamb or 30.96 pounds more lamb than did the late or yearling-bred ewes. He also reported that early bred ewes had more broken mouths after six years which indicates that early breeding may have some adverse effect on normal tooth development.

Evidence points to lactation and not early breeding per se as being the cause of delayed mature size in early bred females. Bogart et al. (1939) found that early bred animals may sustain a setback in their growth curve due to the drain of lactation but they eventually reach the same mature size as the late bred animals. Asdell and others (1941) studied the effects of early and late breeding on the female rat and found that females that are allowed to reproduce and lactate when very young are not as productive in later life as are females that are permitted further development following puberty before they are bred. However, they also found that breeding late

in life limits the reproductive performance of the female rat by reducing litter size and increasing the interval between litters. Also, late-bred females have more parturition difficulties and are generally poor mothers.

Early estrus and subsequent conception may indeed be a reliable indicator of lifetime reproductive performance. Terrill (1946, 1962) has shown that early lamb production is positively correlated with later performance. In 1952, Hulet, Wiggins and Ercanbrack initiated a comprehensive study on estrus in range lambs of three breeds. They found that lambs that showed estrus their first winter had a greater cumulative lamb production record than those ewes that did not show estrus their first season. This was true for all three breeds over the first three years of production records, but the effect was more evident in the Rambouillet breed. Their records show that early-estrus lambs had heavier weaning and yearling fall body weights, were older and had lower inbreeding coefficients, higher condition scores and less face covering.

#### Effects of Gonadotropins

Many researchers have investigated the effects of various gonadotrophic substances on inducing breeding activity in anestrus ewes. Again, however, very little information was found indicating the effects of pituitary gonadotropins on immature ewes.

Cole and Miller (1935) succeeded in inducing estrus in anestrus ewes by the injection of estrogenic hormone alone. They obtained their estrogenic hormone from pregnant mare's urine and found that large doses, 800 to 3200 rat units, were most effective in causing regular estrus. Combined injections of estrogen and gonadotrophic hormone were not successful in causing both estrus and ovulation. Subcutaneous injections of small doses of a partially purified pituitary extract were found to cause ovulation in 3-5 month old ewe lambs (Casida, 1934). McKenzie and Bogart (1935) produced estrus in 70% of the ewes injected with pregnant mare serum (PMS) or Antuitrin - S during anestrus, whereas only 35% of the non-treated controls exhibited estrus in the same time period. Casida's work (1934) shows that inducing ovulation is possible in the very immature female; however, the difficulty in obtaining pregnancy is probably due to the lack of a psychological estrus along with ovulation.

Hunt (1964) reports that using PMS or human chorionic gonadotropin (HCG) after synchronization of estrus with progesterone, either injected or fed, produced no trend in the number of ovulations even when varying dosages of PMS and HCG were given. In ewes given varying dosages of PMS and horse anterior pituitary extract (HAP) there were significant differences in ovarian response. PMS caused a greater overall ovarian response than HAP but there was no difference in the number of ovulations. Examination revealed that



PMS causes a much higher percentage of persistent follicles, true particularly at higher dose levels. At lower dosages no significant difference was found in the proportion of follicles that ovulated (Shelton and Moore, 1967). Van Rensberg (1964) compared HAP and PMS at a one-dose rate and found a greater yield of ova from ewes treated with HAP and observed that as the percentage of persistent follicles went up the percentage of ova fertilized went down. It is suggested that HAP may be more reliable than PMS for initiating ovulation (Hammond, Hammond and Parks, 1942; Casida, Warwick and Meyer, 1944; Moore and Shelton, 1964).

Hunt (1964) has also reported that HAP shows promise in inducing super-ovulations. Subcutaneous injections of 75 mg HAP for ewe lambs and 100 mg for mature ewes given in six equal injections 12 hours apart beginning on the 12th day following estrus resulted in 1-21 ovulations with an average of 11.3 ovulations in 74 treated ewes.

Curl, Nix and Hudson (1968) treated 47 Rambouillet ewes with 33 mg FSH intramuscularly on the 13th day following estrus, followed by 25 mg LH administered intravenously at the onset of estrus. Thirty-six ewes not treated served as a control group. Lambs from the control group and single lambs from the treatment group were significantly heavier at birth but the treatment did not increase the lambing rate based on lambs at two weeks of age. Treated ewes had a significantly higher proportion of multiple births than the controls

( $P < 0.01$ ) but a significantly greater proportion of control ewes lambed as a result of first mating than did the treated ewes ( $P < 0.01$ ).

One widely reported procedure for inducing fertile estrus in anestrus ewes involves pretreatment with progesterone followed by injections of various combinations of PMS, FSH, HCG, pituitary extract and/or LH. Varying degrees of success have been reported.

Dutt and Casida (1948) and O'Mary, Pope and Casida (1950) demonstrated that ovulation and estrus could be prevented during the breeding season by injections of progesterone and that when treatment ceased estrus and ovulation would occur within a few days in most cases. Pursel and Graham (1962) were successful in inducing estrus (91.3%) and ovulation (98.4%) in anestrus ewes that received 9-19 days pretreatment with progesterone and a single injection of from 20-50 mg FSH. Injection of 25 mg FSH without progesterone pretreatment resulted in only 17.6% estrus in 17 treated ewes. Oral administration of 6 methyl-17-acetoxyprogesterone successfully replaced progesterone injections as pretreatment.

Mansour (1959) studied the effects of progesterone, PMS, and CG (chorionic gonadotropin) on the ovaries of 1-10 week-old ewe lambs. He found 1-week old lambs entirely unresponsive to PMS treatment. However, 4-week old lambs treated with progesterone and PMS or PMS alone, showed stimulated follicular growth. In 8-week old lambs, PMS caused follicular growth, progesterone plus

PMS induced follicular lutenization, and PMS plus CG stimulated ovulation. Endogenous LH was not present in sufficient strength to induce ovulation in the PMS-alone or progesterone-plus-PMS treated females until they were 16 weeks old.

There is evidence to indicate that fertility, at the induced estrus following treatment with progesterone and PMS, is markedly reduced but fertility at subsequent estrus periods is uniformly high and normal. Ova recovered following synchronization treatment had fewer spermatozoa in the zona pellucida than did ova from untreated control ewes. It has been suggested that the progesterone has some detrimental effect on either sperm transport or capacitation and this then is the cause of lowered fertility at first estrus (Cullen and Shearer, 1964; Cullen, Hovell and Shearer, 1968). These data agree closely with results discussed earlier by Curl, et al. (1968); however, they presented no hypothesis for the lower conception rate at first estrus.

As mentioned previously, most of the work on the use of gonadotropins to increase fertility in sheep has been done on mature, anestrus ewes, and very little on ewe lambs. The reluctance of breeders to expose ewe lambs during their first breeding season has been due to a belief that future performance is adversely affected (Bowstead, 1930) but this does not appear to be true. Extremely low conception rates (35-50%) among ewe lambs have been the primary

problem at this institution, and has been attributed to probably hormonal imbalance. However, Mansour's work (1959) would tend to negate this possibility.

### Diallel Cross

The diallel mating scheme was first proposed by the Danish geneticist, J. Schmidt, and was meant to evaluate the breeding value of two sires on the basis of successive mating to the same dam (Pirchner 1969). More recently "diallel" has come to mean a cross of P lines with  $P^2$  possible combinations of which P are the inbred combinations and  $P(P-1)$  are the crossbreds and reciprocals. Sprague and Tatum (1942) originally defined the terms 'general and specific combining ability' with regards to diallel crossing. General combining ability has been defined as "performance of the offspring of all the crosses for which a given line has supplied a parent" (Pirchner, 1969). Specific combining ability is defined as "those cases in which certain combinations do relatively better or worse than would be expected on the basis of the average performance of the lines involved" (Griffing, 1956).

In animal breeding experiments, reciprocal variation is due primarily to sex-linked and maternal effects and it is therefore important that a means of determining these differences be considered. Eisen et al. (1966) has proposed models for diallel analysis

that account for sex-linked and maternal effects and also take into consideration:

1. whether the male or female is the homogametic sex, and
2. whether the trait is measured on male or female progeny or both.

Of the four models proposed, only one is relevant to this study and it is as follows:

$$X_{ij} = \mu + A_i + L_i + A_j + M_j + L_j + S_{ij} + E_{ij}$$

Where:

$X_{ij}$  = the progeny mean of the cross i times j,

$\mu$  = overall mean,

$A_i$  = cumulative additive effect of the autosomal genes for line of sire,

$A_j$  = cumulative additive effect of the autosomal genes for line of dam,

$L_i$  and

$L_j$  = cumulative additive effect of the sex-linked genes,

$M_j$  = average maternal effect,

$S_{ij}$  = cumulative non-additive effect (specific combining ability of the cross i times j),

$E_{ij}$  = random effect of environment and genetic segregation.

The assumptions for the proposed model are: (1) Mendelian diploid segregation, (2) an inert Y chromosome.

Eisen compared models prepared by Henderson (1948) and Griffing (1956) and concluded that neither model was sufficient to separate sex-linked effects from either maternal or additive autosomal genetic effects so long as data on only one sex of progeny is analyzed. It must be kept in mind however, reciprocal differences due to sex-linkage and maternal effects are of little importance in plant studies (Griffing, 1956).

### Heterosis

Heterosis is defined as the difference between the mean of the crossbred progeny and the average of the two parental lines or breeds (Pirchner, 1969; Willham, 1970). The cause of heterosis is dominance effects and does not occur when the traits are only influenced by additive genetic effects. Traits that show heterotic effects are sometimes referred to as heterotic characters and there is a preponderance of evidence to indicate that fertility is a heterotic character. Sidwell et al. (1962) found that average increases in percent lambs weaned of ewes bred for 2, 3, and 4 breed crosses were 2.1, 14.9 and 27.1 respectively over the same averages for the purebred parents. They also generalized by reporting overall reproductive ability is higher for crossbred matings than for purebred. Heterosis for fertility and livability was found in cattle by Gaines et al. (1966). They reported a 10% increase in calves weaned from crossbred dams.

Bowman and Falconer (1960) inbred ten lines of mice through 12 generations by breeding double first cousins as the 0 generation and followed with full-sib matings for the remainder of the experiment. They maintained a non-inbred control line as selected and non-selected lines within the inbred population. Selection was based on litter size. Three of the inbred lines survived to 81% inbreeding and these lines were crossed. They found that the litters produced by the crossbred progeny were larger than the non-inbred control by approximately two mice per litter. They credited this gain from heterosis to selection for performance among the inbred lines.

Botkins and Paules (1965) studied straight-bred and crossbred Suffolk and Corriedale ewes and found that crossbred ewes had higher average lambing percents and higher weight of lamb produced than either of the parent breeds. Corriedale ewes mated to Suffolk rams were the most productive of the crossbreds. Lower fertility for inbred lines of cattle have been reported by Stonaker (1954, 1958) and Hawk, Tyler and Casida (1955).

There have also been reports of no heterosis in fertility. Donald (1963) found no heterosis for crossbred sheep based on adjusted records for the first two lamb crops; however, a higher crossbred advantage was postulated for more mature sheep.

In a study done by Fox and McArthur (1963), crossbred ewe lambs were found to be significantly more fertile than comparable

purebreds. The crossbred lambs were of Columbia x Hampshire and Targhee x Hampshire breeding. An equal number (16) of purebred Hampshire lambs were used as control comparisons. They obtained a ewes lambing percent of 44 and 94 for the purebreds and crossbreds respectively, based on the number of ewes exposed to ram. They also obtained a lambing percentage of 100 for purebreds and 153 for the crossbreds based on the number of ewes that lambed. Fifty percent of the crossbreds produced multiple births against none for the purebreds. They postulated that this difference in fertility could be due to nonadditive gene action.



## METHODS, RESULTS AND DISCUSSION

### Introductory Comments

In 1965 a series of studies were initiated to determine if exogenous gonadotropin treatment would effect the fertility of ewe lambs. The program includes six separate studies; one study for each of the breeding seasons from 1965 to 1970. It was necessary to separate and analyze the data in this manner due to the fact that each fall new experimental animals were used. Also, treatment procedures varied greatly from one season to the next. Therefore, analysis and discussion of the data by year was necessary because of the lack of biological continuity from one year to the next. There are, however, several aspects of the program that are consistent and these will be discussed first. In all instances the lambs were born in February and March and were first bred the following September or at 6-8 months of age. All of the ewe lambs in this program were of the Willamette and/or Suffolk breed except for the last year. The last year of the study will be discussed in more detail later. Management practices were maintained as uniformly as possible over the entire six years. All the ewes and rams were maintained at the Oregon State University Hill Pasture Sheep Farm under the care of one herdsman for the entire program.

The rams used in this program were different for each breeding season. All rams were semen tested using the criterion of sperm motility. Semen samples were obtained just prior to the breeding season by use of an electro-ejaculator. Motility of the sperm was evaluated by placing a drop of semen on a slide and subjectively rating motility from 1-10, with 10 being the most active. As mentioned, this test was subjective but it was a basis for determining the most "fertile" ram. Only rams with a rating of 9 or 10 were used in the breeding program.

Breeding records were kept daily during the time the rams were with the young ewes. Each ram was equipped with a chest marking harness. Colors were changed in the harness about every two weeks. Ewes were considered to have been serviced if they were marked clearly on the rump. Wet weather caused some problems with this method but not enough to make it unusable.

The data were collected and prepared for analysis as uniformly as possible from one year to the next. For example, fertility was represented throughout the study by letting the numbers 1, 2 and 3 indicate that a ewe lamb was barren, carried a single lamb, or carried twins, respectively. In no year did a ewe lamb give birth to more than twins. Even though no analysis was done across years, weights of ewes and their lambs were taken as near the same intervals as practical. The term "fertility" will be used to refer to

whether a ewe lamb conceived and carried a lamb to term. Spontaneous abortion was not a problem with no more than 2-3 cases over the five-year program.

#### 1965 Study

Forty-two ewe lambs, thirty-one Willamette and eleven Suffolk, were available for the initial study involving the use of PMS. The Willamette lambs were divided into treatment and control groups but all of the Suffolk ewes were placed on treatment due to the small number. All ewes were subjected to fifteen days of pretreatment with Repromix. Repromix and PMS were supplied by the Upjohn Company, Kalamazoo, Michigan. The Repromix contained 3.24 grams Medroxyprogesterone acetate per pound. The ewes were fed 3/4 of a pound of untreated ground grain beginning August 20, 1965 for seven days to allow them to become accustomed to it. On August 27, Repromix was mixed with the ground grain so that each ewe received 50 mg per ewe per day in 3/4 pound of grain. Pretreatment ended on September 9 and rams were placed with the ewes on September 10. The rams used were two years old and of the same breed as the ewes.

On September 13, 17 Willamette and 11 Suffolk ewes were injected intramuscularly with 400 IU of PMS. The PMS was Upjohn's "Gonadogen" which was mixed in two ml of sterile saline. The same procedure was repeated on September 27. The fourteen non-injected

Willamette ewes were maintained with the treated. The two breeds were maintained separately.

Breeding dates were observed and recorded by the use of marking harnesses on the rams. Colors were changed at the time of the second PMS injection.

### Results and Discussion

The use of Repromix on ewe lambs was not completely successful. Approximately 65% of the ewe lambs showed evidence of having estrus synchronized. Deweese, et al. (1970) compared oral medroxyprogesterone acetate given orally and in vaginal sponges to mature crossbred ewes and found that both methods are effective for synchronizing estrus. They also reported no significant differences due to oral MAP or the vaginal sponges in total lambing rate. From the present study, it appears that ewe lambs do not respond as effectively to orally administered MAP as do mature ewes.

There were no significant differences between treatment and control Willamette ewe lambs in percentage that conceived. However, the treated ewes had a higher lambing percent, based on ewes lambing, than did the control lambs. Lambing percent, based on ewes exposed to the ram, was approximately equal for treated and control ewes. (Table 2). There were slightly significant differences ( $P < 0.10$ ) between the treated Willamette and Suffolk ewe lambs, with the

Willamette lambs showing a greater response to the treatment than the Suffolk lambs (Table 1). Only three of the eleven Suffolk ewes carried a lamb which gave a percent of ewes lambing of 27.3%. The Suffolk ewes were comparable in size and age to the Willamettes. Therefore, the only conclusion is that treatment or handling during treatment has some adverse effect on the Suffolks. Another assumption may be that Suffolk ewe lambs do not mature at as early an age as Willamettes. Shelton and Moore (1967) concluded that PMS, particularly at high dosages, may cause persistent follicles. This, however, should be a general problem and not specific to one breed. It is possible that if persistent follicles are an effect of PMS in mature ewes whose reproductive systems are fully mature and functional, the problem may be accentuated in ewe lambs.

Table 1. Analysis of variance comparing fertility in the treated Willamette versus Suffolk ewe lambs.

Source	df	MS	F
Willamette vs. Suffolk	1	1.407	2.919 <sup>+</sup>
Error	30	0.482	
Total	31		

<sup>+</sup> P < 0.10

Table 2. Summary of breeding and lambing results for 1965.

	Breed		Overall
	Willamette	Suffolk	
Treated			
No. of ewes	19	11	30
No. of ewes lambing	9	3	12
No. of lambs born	11	3	14
Percent of ewes lambing	47.4	27.3	40.0
Lambing percent <sup>a/</sup>	57.9	27.3	46.7
Lambing percent <sup>b/</sup>	122.2	100.0	116.7
Control			
No. of ewes	12	--	12
No. of ewes lambing	6	--	6
No. of lambs born	7	--	7
Percent of ewes lambing	50.0	--	50.0
Lambing percent <sup>a/</sup>	58.3	--	58.3
Lambing percent <sup>b/</sup>	116.7	--	116.7

<sup>a/</sup> Based on ewes exposed to ram.

<sup>b/</sup> Based on ewes lambing.

1966 Study

In 1966, 34 Willamette and 27 Suffolk ewe lambs were available for hormone study. Ewes within each breed were randomly divided into two groups; one being the treatment group, the other the controls.

No attempt was made to synchronize estrus. The treatment consisted solely of a series of three injections of PMS in the form of "Gonadogen" supplied by the Upjohn Co., Kalamazoo, Michigan. On August 26, 1966, the Willamette and Suffolk treatment groups received 400 IU PMS via subcutaneous injection in two ml sterile saline. On August 27 breeding groups were established by randomly allotting ewes to groups such that both treated and control ewes were placed in all breeding groups. Breed integrity was maintained but treated and control ewes were mixed within breed to reduce differences due to the ram. On September 9, the treatment ewes received another injection of PMS, 200 IU in one ml sterile saline. A third and final injection of 400 IU was given on the following day, 10 September.

Semen tested ram lambs of the same breed as that of the ewes to be bred were turned in with the ewes on August 27. Ram lambs were utilized in all breeding groups except one Suffolk group in which a two year old ram was used. Daily breeding records were kept until the rams were removed on October 20, 1967.

## Results and Discussion

Analysis of variance was used to determine if differences existed between treatment and control groups or between breeds. There were no significant differences due to treatment or breed. The breed x treatment interaction was also non-significant. From Table 3 it is evident that Suffolk ewe lambs responded adversely to treatment in that only 3 of the 16 ewes lambled. However, of the 3 ewes lambing, 2 ewes had twins. Of the Willamette ewe lambs, one ewe gave birth to twins. In the control group there were no twin births. The overall lambing percent based on ewes lambing was 130.0% and 100.0% for the treated and controls respectively.

It is the opinion of the author that this program was begun too early in the breeding season. Fox and McArthur (1963) found that purebred ewe lambs came into first estrus approximately 17 days later than crossbred ewe lambs of the same age. Their breeding season began on September 14 and rams were removed on October 30. Since crossbred ewe lambs were bred earlier in the season, they hypothesized the remote possibility that one cause of reduced fertility in the purebreds could have been due to reduced fertility of the rams later in the season. Overall results from the present program indicate that ewe lambs generally come into estrus later than mature ewes of the same breed.



Table 3. Summary of breeding and lambing results for 1966.

	Breed		Overall
	Willamette	Suffolk	
Treated			
No. of ewes	18	16	34
No. of ewes lambing	7	3	10
No. of lambs born	8	5	13
Percent of ewes lambing	38.9	18.8	29.4
Lambing percent <sup>a/</sup>	44.4	31.3	38.2
Lambing percent <sup>b/</sup>	114.3	166.7	130.0
Control			
No. of ewes	16	11	27
No. of ewes lambing	7	4	11
No. of lambs born	7	4	11
Percent of ewes lambing	43.8	36.4	40.7
Lambing percent <sup>a/</sup>	43.8	36.4	40.7
Lambing percent <sup>b/</sup>	100.0	100.0	100.0

<sup>a/</sup> Based on ewes exposed to ram.

<sup>b/</sup> Based on ewes lambing.

The lack of a significant difference between breeds is not consistent with results found in other years of this program. Breed differences approached significance in 1964 and were significant in 1967. Either there was sufficient environmental influence to negate the difference in breeds or the adverse effect of treatment on the Suffolk ewe lambs noted for other years was not as evident during this year.

Pregnant mare serum alone was not sufficient to influence fertility in these ewe lambs. There was a slight response in the number of multiple births but the total number of ewes lambing was not affected by treatment.

#### 1967 Study

In 1967 a more extensive treatment program was undertaken utilizing 39 Willamette and 28 Suffolk ewe lambs. From a total of 67 ewe lambs, 37 were randomly allotted to the treatment group and the remaining 30 were maintained as controls.

The treatment began on August 25, 1967 with all ewe lambs in the treated group receiving 16 mg "Promone" (Medroxyprogesterone acetate, supplied by the Upjohn Company, Kalamazoo, Michigan) per day for the first four days via intramuscular injection. On the fifth day each ewe received a single injection of 5 mg 17-beta-estradiol also intramuscularly. On the day following estradiol

treatment the ewes were placed on a two-week regimen of Repromix, fed at the rate of 50 mg per day mixed in 3/4 pound of ground grain per ewe. On September 9, 1967 each ewe lamb in the treated group received a final injection of 700 IU PMS and the rams were turned in on the same day.

Ewes in the control group were subjected to the same handling procedures as were the treated ewes. However, no attempt was made to simulate injections on the control animals with inert material. It was felt that the controls should be maintained as unmolested as possible as far as treatment procedures were concerned.

All ewes, treated and control, were allotted randomly and equally into breeding groups to eliminate differences due to the ram. All breeding groups were pasture-mated and breeding records were kept daily through the use of crayon marking harnesses on the rams. Mature rams of the same breed which had been semen tested and known fertile through previous breeding records were used. Rams remained with the ewe lambs until October 31, 1967 when they were removed.

### Results and Discussion

Overall lambing results were higher for 1967 than for either of the previous two years. Of the 39 Willamette ewe lambs there were 23 lambs born including four sets of twins. The Suffolk ewe

lambs did not respond adversely to treatment during this year. Eighteen of the 28 young Suffolk ewes lambled including six sets of twins. Multiple births were spread over both treatment and control groups with five twin births in each group. The percent of ewes lambing for the Willamette breed over treatment and control was 49 percent. For the Suffolk ewe lambs the percent of ewes lambing was 64 percent. The mean difference in percent ewes lambing between the two breeds is significant at  $P < 0.05$ .

There was no significant difference between the treated and control groups. The overall lambing percent based on ewes exposed to the ram was highest in the treated group, 73.0% in contrast to 66.7% for the controls. However, the lambing percent based on ewes lambing was highest in the control group, 133.3% versus 122.7% for the treated ewes. The percent of ewes lambing was approximately 10% higher for the treated group but as mentioned it is not statistically significant and cannot be attributed directly to treatment.

Table 4. Analysis of variance for ewe lamb fertility in 1967.

Source	df	MS	F
Treatment Total	3	1.170	1.87
Treatment	1	0.501	$P < 1$
Breed	1	2.742	4.38*
B x T	1	0.268	$P < 1$
Error	73	0.626	
Total	76		

\*  $P < 0.05$

Table 5. Summary of breeding and lambing results for 1967.

	Breed		Overall
	Willamette	Suffolk	
Treated			
No. of ewes	28	9	37
No. of ewes lambing	16	6	22
No. of lambs born	19	8	27
Percent of ewes lambing	57.1	66.7	59.5
Lambing percent <sup>a/</sup>	67.9	88.9	73.0
Lambing percent <sup>b/</sup>	118.8	133.3	122.7
Control			
No. of ewes	11	19	30
No. of ewes lambing	3	12	15
No. of lambs born	4	16	20
Percent of ewes lambing	27.3	63.2	50.0
Lambing percent <sup>a/</sup>	36.4	84.2	66.7
Lambing percent <sup>b/</sup>	133.3	133.3	133.3

<sup>a/</sup> Based on ewes exposed to ram.

<sup>b/</sup> Based on ewes lambing.

### 1968 Study

In 1968 a total of 90 ewe lambs, 45 Willamette and 45 Suffolk, were available for study. All lambs were placed on treatment and fed MGA (Melengestrol acetate premix, MGA-100, Upjohn Co.) at 0.3 mg per lamb per day in 0.5 lb. ground grain mixture starting on September 6 and continuing for 10 days or until September 15, 1968. They were taken off MGA for three days during which each lamb received a daily injection of 0.25 mg 17-beta-estradiol, but grain feeding was continued. On September 19, the lambs were placed again on the premix treatment of 0.3 mg MGA per head per day in 0.5 lb. of ground grain. The administration of MGA was continued for 14 days or until October 2. On October 2, the lambs were allotted randomly within breed to two groups of 22 and 23 ewes each. One group was given 700 IU of PMS and the other 500 IU of PMS by intramuscular injection on October 2. Breeding groups within breed were established and rams that were proven fertile by semen test and previous records were put with the ewes on October 2. On October 3, the lambs were randomly allotted within breed and level of PMS to two groups. One group within each breed and within each level of PMS was given 800 IU APL (chorionic gonadotropin, Ayerst Laboratories Inc.) by injection and the other group received no APL. Ewe lambs were allotted to treatment groups as follows:

<u>Breed</u>	<u>MGA</u>	<u>PMS</u>	<u>Chorionic gonadotropin</u>
Suffolk			
45	45	23 at 700 IU	12 at 800 IU 11 at 0 IU
		22 at 500 IU	12 at 800 IU 10 at 0 IU
Willamette			
45	45	22 at 700 IU	11 at 800 IU 11 at 0 IU
		23 at 500 IU	12 at 800 IU 11 at 0 IU

Breeding records were kept on a daily basis using crayon marking harnesses on the rams for identifying ewes in heat. The color of the crayon was changed every 14 days going from a light color initially to a darker one later.

### Results and Discussion

Analysis of variance was used to analyze the data for differences in fertility due to breed, treatments (PMS and HCG), and the interactions among treatments and breeds. The breed of ewe lamb had no significant effect on fertility which is not consistent with results found in other aspects of this study. All ewe lambs in this study were treated; there were no control animals. From Table 8 it can be seen that the breed x HCG interaction approached significance ( $P < 0.10$ ) which indicates that the interactions between breeds and treatments could have caused the absence of a significant breed difference.

In general, PMS was the only treatment that approached having a significant effect on ewe lamb fertility ( $P < 0.10$ ). Many workers have found PMS to be very effective in enhancing the fertility and off-season breeding in mature ewes (Roberts and Edgar, 1966; Moore and Shelton, 1964; Brunner, Hansel and Hogue, 1964). More lambs were conceived in the ewe lambs treated with 500 IU PMS than in the group receiving 700 IU PMS. This is in agreement with work by Shelton and Moore (1967) who found that PMS in high dosages tends to cause persistent follicles. This could explain the lowered fertility rate at the higher dose level.

The presence or absence of HCG had no significant effect on the fertility of the ewe based on the number of ewes lambing that were exposed to the ram. However, it is interesting to note that all multiple (twin) births occurred in the treatment groups that received 800 IU of HCG; therefore, the HCG had some small multi-ovulating effect in both breeds of ewe lambs.

Except for the breed x HCG interactions, the other interactions analyzed were non-significant. As stated, the breed x HCG interaction only approached significance.

Birth weights of the lambs from the treated ewes were also analyzed with regard to the treatments and interactions previously mentioned. Total treatment effect was statistically significant ( $P < 0.05$ ). However, of the factors measured only breed had any



influence on lamb birth weight and that effect was highly significant ( $P < 0.01$ ). The lambs from the Willamette ewe lambs were approximately 2.5 pounds heavier at birth than the Suffolk lambs. Several of the Suffolk ewe lambs, however, gave birth to lambs in the 2.0 to 4.8 lb. range which accounts for the large standard deviations among the birth weights of the Suffolk lambs. These lambs in general were born dead; however, they were carried full term. There is no apparent reason for the abnormally small lambs from some of the Suffolk ewe lambs as there appeared to be no correlation between lamb birth weight and weight of the ewe.

Table 6. Means and standard deviations of birth weights of lambs from treated ewe lambs.

Treatment	Breed	
	Willamette	Suffolk
	Lbs.	Lbs.
Treatment I <sup>a/</sup>	10.49 ± 1.52	7.55 ± 3.29
Treatment II <sup>b/</sup>	9.40 ± 1.92	8.25 ± 1.39
Treatment III <sup>c/</sup>	9.40 ± 2.00	6.38 ± 2.03
Treatment IV <sup>d/</sup>	10.75 ± 2.06	8.74 ± 3.19
Breed mean	10.13	7.53

<sup>a/</sup> 500 IU PMS, no HCG

<sup>b/</sup> 700 IU PMS, no HCG

<sup>c/</sup> 500 IU PMS, 800 IU HCG

<sup>d/</sup> 700 IU PMS, 800 IU HCG

Table 7. Summary of breeding and lambing results for 1968.

	Breed		Overall
	Willamette	Suffolk	
Treatment I <sup>c/</sup>			
No. of ewes	11	10	21
No. of ewes lambing	10	6	16
No. of lambs born	10	6	16
Percent of ewes lambing	90.9	60.0	76.2
Lambing percent <sup>a/</sup>	90.9	60.0	76.2
Lambing percent <sup>b/</sup>	100.0	100.0	100.0
Treatment II <sup>d/</sup>			
No. of ewes	11	11	22
No. of ewes lambing	5	4	9
No. of lambs born	5	4	9
Percent of ewes lambing	45.5	36.4	40.9
Lambing percent <sup>a/</sup>	45.5	36.4	40.9
Lambing percent <sup>b/</sup>	100.0	100.0	100.0
Treatment III <sup>e/</sup>			
No. of ewes	12	12	24
No. of ewes lambing	4	8	12
No. of lambs born	5	10	15
Percent of ewes lambing	33.3	66.7	50.0
Lambing percent <sup>a/</sup>	41.7	83.3	62.5
Lambing percent <sup>b/</sup>	125.0	125.0	125.0
Treatment IV <sup>f/</sup>			
No. of ewes	11	12	23
No. of ewes lambing	6	6	12
No. of lambs born	6	7	13
Percent of ewes lambing	54.5	50.0	52.2
Lambing percent <sup>a/</sup>	54.5	58.3	56.5
Lambing percent <sup>b/</sup>	100.0	116.7	108.3

<sup>a/</sup> Based on ewes exposed to ram.      <sup>e/</sup> Treatment - 500 IU PMS,  
800 IU HCG.

<sup>b/</sup> Based on ewes lambing.

<sup>c/</sup> Treatment - 500 IU PMS, no HCG.      <sup>f/</sup> Treatment - 700 IU PMS,  
800 IU HCG.

<sup>d/</sup> Treatment - 700 IU PMS, no HCG.

Table 8. Analysis of variance for ewe lamb fertility in 1968.

Source	df	MS	F
Treatment Total	7	0.558	1.44
Breed	1	0.63	P<1
HCG	1	0.293	P<1
PMS	1	1.427	3.69 <sup>+</sup>
B x HCG	1	1.476	3.81 <sup>+</sup>
B x PMS	1	0.014	P<1
HCG x PMS	1	0.221	P<1
B x HCG x PMS	1	0.412	1.06
Error	86	0.387	
Total	93		

<sup>+</sup> P<0.10

Table 9. Analysis of variance for birth weight of lambs from treated ewes.

Source	df	MS	F
Treatment Total	7	12.981	2.53 <sup>*</sup>
Breed	1	62.678	12.22 <sup>**</sup>
HCG	1	0.131	P<1
PMS	1	8.326	1.62
B x HCG	1	0.662	p<1
B x PMS	1	5.924	1.16
HCG x PMS	1	12.693	2.48
B x HCG x PMS	1	0.456	P<1
Error	45	5.128	
Total	52		

<sup>\*</sup> P<0.05

<sup>\*\*</sup> P<0.01

### 1969 Study

In 1969 it was decided to simplify the treatment and place the emphasis of the study on the physiological and histological state of the reproductive tract. Toward this end a portion of the ewes from each treatment group as well as untreated control ewes were slaughtered and the major endocrine glands and reproductive tracts were removed for study.

For this study 111 ewe lambs, 70 Willamette and 41 Suffolk, were placed on test. Past experience indicated it may be more wise to use as many animals as possible for treatment at the expense of a control group; therefore, on September 1, 1969, all ewe lambs were weighed and treatments were initiated. Each ewe received 10 mg progesterone ( $\Delta^4$ -pregnen-e, 20-dione, prepared by Sigma Chemical Co., St. Louis, Mo.) given in one ml propylene glycol per day for six days via subcutaneous injection. On September 12, 1969, the ewe lambs were divided into three treatment groups as outlined below. There were not sufficient numbers of Suffolk lambs available; therefore, no Suffolks were placed on 50 mg APE treatment. Anterior pituitary treatment consisted of a single subcutaneous injection of 50, 150 and 450 mg APE suspended in two ml propylene glycol. This particular procedure caused some serious difficulties which will be discussed in more detail later. On September 13, the ewes

were allotted within breed to five breeding groups, each group comprised of ewe lambs from each of the treatment groups and proven fertile rams were put with them. Breeding groups were on non-irrigated pastures until November 6, 1969.

#### Experimental outline for 1969 study

<u>Breed</u>	<u>Progesterone</u>	<u>Anterior Pituitary Extract</u>
Willamette 70	70	3 - slaughter control 21 - 50 mg APE 23 - 150 mg APE 23 - 450 mg APE
Suffolk 41	41	2 - slaughter control 0 - 50 mg APE 20 - 150 mg APE 19 - 450 mg APE

Anterior pituitary extract was obtained from Mann Research Laboratories, New York, N. Y. A preliminary bioassay was conducted to determine whether or not the APE had any LH activity. The procedure is outlined in Yarrow et al. (1964). Ten Sprague-Dawley male rats, 40-50 grams at 21 days of age were obtained from Simonson Laboratories, Gilroy, California. Five rats were allotted randomly into the control group. The remaining five received 6 mg APE per day for four days. Ventral prostate glands were removed on the fifth day and weighed. The ventral prostate glands of the treated rats were significantly larger than those of the control. This indicated LH activity in the APE; however, no attempt was made to

quantitate the amount of LH present.

Twenty-five ewe lambs were slaughtered on September 22, 1969 for the purpose of examining the ovaries, uterus, and endocrine glands. Three Willamette and two Suffolk ewes were withheld from the APE treatment to serve as slaughter controls. Four ewe lambs were selected from each of the treatment groups for slaughter. All the ewes were slaughtered at the Oregon State University Meats Laboratory. The reproductive tracts were removed, trimmed as uniformly as possible and weighed, after which both ovaries were separated from the reproductive tract and weighed together. The ovaries and a cross section through both horns of the uterus were placed in Bouins solution for future histological examination. The adrenal glands were removed, trimmed and weighed but not preserved. The pituitary glands were removed and weighed, then quick frozen on dry ice for subsequent bioassay for lutenizing hormone.

Simple correlations were run using live slaughter weight as the independent variable to determine if endocrine gland and reproductive tract weights were positively associated with body weight. Of the endocrine glands, the ovary was correlated most highly with body weight,  $r = 0.76$ . The coefficient of determination ( $r^2$ ) indicated that 57.2% of the variation in ovarian weight is associated with the variation in body weight. The regression (b) shows a 0.02 gram increase in ovarian weight for each pound increase in body weight

(Figure 1). Of course, it's logical that the organs of the body would be in proportion to the body as a whole and this is a simple explanation for the high ovarian correlation. However, what does not follow is that the correlation between pituitary weight and body weight is only  $r = 0.27$ . This would tend to indicate that size and growth of the pituitary is independent of the rest of the animal as these ewes had not yet reached mature size. Adrenal gland weight was moderately correlated with body weight,  $r = 0.44$ . Again, a fairly high correlation would be expected. Reproductive tract weight was lowly correlated with body weight. Each of these ewes were very near the same stage of estrus at slaughter as determined by histological examination. However, the lack of correlation is undoubtedly due to sampling error rather than it being the actual situation. When the reproductive tracts were removed an attempt was made to trim the fat and non-associated tissue away. It is the author's opinion that the inability to accomplish this uniformly resulted in considerable variation in weight. An analysis of variance was run to determine if differences existed between treatments and breeds for the various weights. The results were non-significant and will not be discussed further.

As mentioned previously, propylene glycol was used as the injection medium for the progesterone and anterior pituitary extract. No problem existed with the progesterone as it completely dissolved

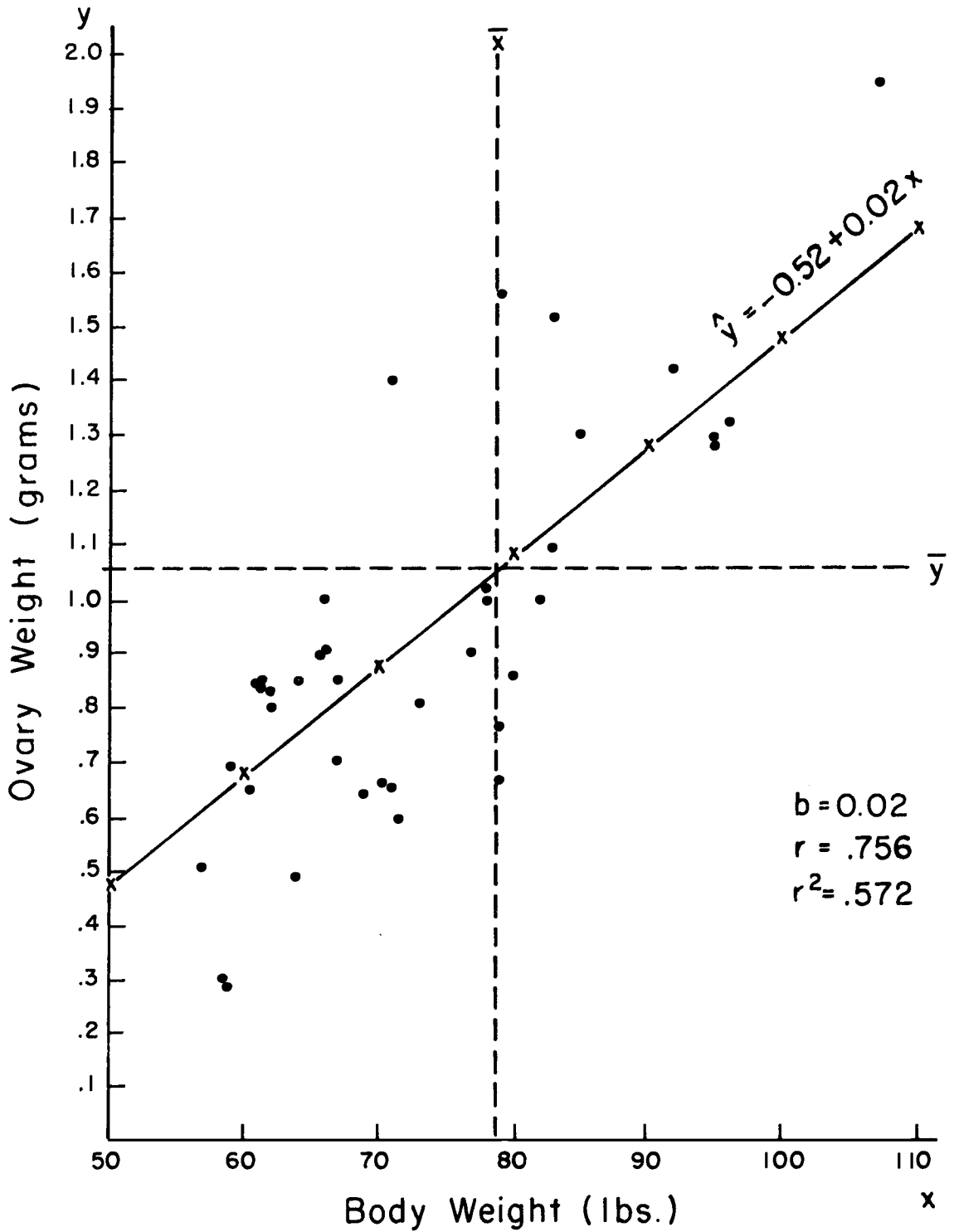


Figure 1. Correlation Scatter Diagram for Ovarian Weight versus Ewe Body Weight



Table 10. Endocrine and reproductive tract weights for slaughter lambs in 1969.

Breed and Treatment Group	Slaughter Wt. (lb)	Pituitary Wt. (mg)	Adrenal Wt. (g)	Ovary Wt. (g)	Reproductive Tract Wt. (g)
Willamette					
50 mg APE <sup>a/</sup>	107.0	548.0	3.25	1.95	53.5
	57.0	346.0	2.21	0.51	23.7
	79.0	346.1	2.51	0.67	28.1
	73.0	491.0	2.40	0.81	25.0
150 mg APE	64.0	393.8	1.98	0.85	36.2
	83.0	342.0	2.00	1.52	33.3
	79.0	450.5	2.27	0.77	31.5
	78.0	446.1	2.75	1.02	31.1
450 mg APE	80.0	425.0	2.22	0.86	33.2
	66.0	491.9	2.19	1.00	30.7
	95.0	578.2	2.25	1.27	33.0
	77.0	450.1	2.15	0.90	29.1
Control <sup>b/</sup>	71.0	440.0	2.30	1.40	25.9
	82.0	520.0	1.86	1.00	37.7
Suffolk					
150 mg APE	64.0	306.2	1.92	0.49	52.8
	96.0	579.5	2.53	1.32	39.3
	83.0	528.3	1.67	1.07	28.7
	62.0	256.0	2.13	0.83	23.0
450 mg APE	67.0	500.0	1.99	0.70	30.7
	79.0	347.0	2.43	1.56	29.7
	78.0	402.0	2.45	1.00	29.7
	92.0	257.0	2.64	1.42	29.8
Control	95.0	204.0	1.95	1.28	51.0
	85.0	435.0	2.13	1.30	38.8

<sup>a/</sup> Anterior pituitary extract.

<sup>b/</sup> One control Willamette ewe died prior to slaughter.

in the propylene glycol and the consistency of the solution remained unchanged. However, the anterior pituitary extract did not dissolve and therefore the suspension tended to become thicker, making injections more difficult. Several of the ewes developed subcutaneous sores at the injection site approximately three days following the injection. A supplemental experiment outlined in Zarrow, et al. (1964) was undertaken to determine what effects might occur from using propylene glycol as an injection medium for anterior pituitary extract. Twenty-five Sprague-Dawley male rats were obtained from Simonson Laboratories, Gilroy, California. Each rat was 21 days old and within a 40-50 gram weight range upon delivery. The rats were allotted randomly to one of five groups: control, saline + 5 mg APE, saline + 15 mg APE, propylene glycol + 5 mg APE and propylene glycol + 15 mg APE. Each rat was injected once daily with its dose of APE in the loose skin at the back of the neck for four days. On the fifth day the ventral prostate gland was removed and weighed on a torsion balance (Table 11).

Table 11. Weight of rat ventral prostate gland as influenced by physiological saline or propylene glycol as the APE carrier.

	Saline			Propylene glycol	
	Control	5mg APE	15mg APE	5mg APE	15mg APE
Weight/ rat (mg)	23.5	44.0	41.7	32.8	46.0
	52.1	46.2	47.0	53.2	35.0
	46.0	41.4	43.6	34.1	55.4
	45.6	43.6	42.3	42.6	29.0
	43.0	39.0	48.4	28.4	30.4
Mean Wt.	42.0	42.8	44.6	38.2	39.2

Generally, the rats injected with propylene glycol were in very poor condition at the end of the experiment. The injection site was thick and hard. On autopsy it was found that the pituitary extract had been walled off and had not been absorbed. Definite irritation was evident in the propylene glycol injected rats and they were lethargic and thin. Regretably, initial and final whole body weights were not taken because it was not thought the reaction would be so pronounced. None of the adverse effects were noted in the saline injected rats. They remained healthy and normal in all respects.

## Histology of the Reproductive Tract

Cross sections of the uterus and of the ovaries were fixed in Bouins solution for subsequent slide preparation. All tissue samples were left in Bouins for approximately one month. This extended period did not adversely affect sectioning quality. Autotechnicon equipment, materials, and procedures were used in the preparation for sectioning. Following fixing and dehydration, tissues were imbedded in paraffin and mounted on wooden blocks. Serial sections were taken of each ovary and of both horns of the uterus so there were four slides per animal. Sections were cut at 10  $\mu$  on a Spencer, model 820 microtome with very satisfactory results. When mounted the tissues were stained with Hematoxylin-Eosin.

The ovaries showed considerable follicular development with follicles in all stages. There was no apparent difference in follicular development when ovaries from ewes from the different treatment groups were compared. Breed of the ewe had no effect on ovarian development. One abnormality was noticed. One ovary from a Willamette control ewe was found to be completely infantile. No follicular development existed and no primary follicles could be found. The cortex appeared loose and fibrous while the medulary region was very compact with only granulosa cells. No evidence of corpora lutea were found in any of the ovarian samples. This

observation is not consistent with observations of the number of follicles present but it does agree with the premise of the author, that although FSH is present in sufficient quantities to effect follicular development, the proper balance of FSH and LH is not present to cause ovulation. This, of course, is not true for all ewe lambs because a certain percentage of them did breed and conceive. The problem that exists is to determine why some ewe lambs do conceive and others do not.

The uteri were also examined. There was little if any differences evident. Uterine samples were taken from each reproductive tract at as nearly as possible the same region, immediately anterior to the bifurcation of the uterine horns. There was no indication of any treatment versus control differences; however, estrogenic activity was apparent. Each sample had a well developed endometrium with corresponding glandular development. There was no indication that any of the uteri were abnormal in any way. All appeared to be fully functional and ready to accept implantation if fertilized ova were present.

There is no apparent histological reason for lower fertility in ewe lambs. All organs and tissues appear to be fully functional. They are slightly smaller in size than uteri from mature ewes but this is due to the smaller size of the ewe lamb.

## Pituitary Assay

Since examination of the ovaries indicate that FSH is present in the immature lamb in sufficient amounts to cause follicular development, the next question that arises is: are there measurable amounts of LH present to possibly effect ovulation?

As previously mentioned, the pituitary glands were removed from the twenty-five slaughtered lambs and quick frozen on dry ice immediately. The pituitaries were prepared for assay by thawing and separating the anterior pituitary (AP) from the pars intermedia and nervosa. Once separated the AP was completely homogenized in ten ml of 0.9% saline. The homogenate of each AP was placed in a petri dish and freeze dried in a Thermovac vacuum chamber, model FDC-10-JDV15, to remove all moisture. Each AP extract was placed in a plastic vial and sealed until the assay could be run.

In an attempt to maintain uniformity, the same assay method mentioned previously (Zarrow et al., 1969) was used to determine LH levels. This assay is based on the fact that LH stimulates the interstitial cells of the testes to secrete testosterone which then stimulates the development of the accessory reproductive tissues, e. g. the prostate gland (Hafez, 1962). Ninety immature male rats (Sprague-Dawley) were obtained from Simonson Laboratories. It was specified that the rats be male, 20 days of age upon delivery,

and between 40-50 grams in weight to reduce variation between rats as much as possible.

The pituitary extract was prepared for injection in three dosage levels, 4, 8, and 16 mg per rat per day for four days. There was not sufficient pituitary extract from each individual ewe to run a separate assay on each. Therefore, the extract from two ewe lambs of the same breed was mixed, group one containing extract from three ewes, making 12 groups of extract to be assayed (Table 12). Pituitary extract from each group was weighed and mixed in 0.9% saline to make up the three treatment levels. The extract was mixed such that each rat received its particular dose in 0.4 ml of saline. Each rat was injected subcutaneously with its particular dosage each morning for four days so that each injection was about 24 hours apart. Twenty-four hours following the last injection the rats were sacrificed and the ventral lobe of the prostate gland was removed and weighed.

Table 12. Experimental design for the bioassay of ewe lamb pituitary glands for lutenilizing hormone.

Treatment	Group 1 <sup>a/</sup>	Group 2 <sup>b/</sup>	Group 3 . . .	Group 12
4mg/day	2 rats	2 rats	2 rats	2 rats
8mg/day	2 rats	2 rats	2 rats	2 rats
16mg/day	2 rats	2 rats	2 rats	2 rats

<sup>a/</sup> Groups 1, 3-8 Willamette ewe lambs

<sup>b/</sup> Groups 2, 9-12 Suffolk ewe lambs

In addition to the treatment, a control group and a dose response curve was run. Ten of the ninety rats served as a control base for the dose response curve. These rats received no treatment whatsoever. The dose response curve was obtained by injecting purified LH in 0.9% saline in the following doses with two rats per dose: 0.05, 0.25, 0.50, 0.75 and 1.00 mg pure LH per rat per day.

An analysis of variance was run to determine if differences between groups and between treatments existed; also the group-treatment interaction was checked for significance. There was no significant difference between groups which was expected. As a further check breed differences within groups were analysed and no significant difference was found between breeds. There is no biological reason to believe that pituitary LH levels will vary significantly between breeds particularly in this instance. The lambs were all close to the same age and level of sexual maturity and had received pretreatment with progesterone to synchronize estrus. There were highly significant differences between treatments. This indicates that as the treatment level increased from 4.0 mg to 16.0 mg pituitary extract there is a measurable increase in LH activity. It does not indicate that the concentration per pituitary increases. It only means that more pituitary extract contains more LH. The fact that the group-treatment interaction was insignificant lends strength to the actual treatment differences. If the means of treatment



Table 13. Rat ventral prostate weights over 12 groups and 3 treatments.

Groups <sup>a/</sup>	Treatment			CONTROL <sup>b/</sup> Rat prostate weights
	4mgAP/day	8mgAP/day	16mgAP/day	
1	64.0	72.0	63.7	43.5
	57.7	60.0	65.0	
2	49.0	51.0	70.2	43.6
	51.7	59.5	67.6	
3	37.0	65.7	75.0	47.7
	51.0	68.2	52.4	
4	47.7	68.1	65.8	37.6
	44.1	65.7	80.1	
5	46.0	62.3	64.4	38.3
	65.0	43.0	74.1	
6	46.3	57.8	43.0	38.3
	49.0	79.0	57.0	
7	67.6	55.0	61.7	43.4
	52.0	52.3	74.0	
8	36.0	57.8	72.3	48.0
	53.6	59.8	69.9	
9	54.2	56.3	56.9	51.8
	70.0	51.7	86.0	
10	63.6	59.0	61.7	49.0
	59.0	55.6	59.0	
11	63.0	46.0	68.0	
	50.0	58.4	51.8	
12	61.8	52.1	60.0	
	78.3	68.1	55.7	

<sup>a/</sup> Data are for rat ventral prostate weight in milligrams, each group contains anterior pituitary from two sheep.

<sup>b/</sup> Rats received no treatment.

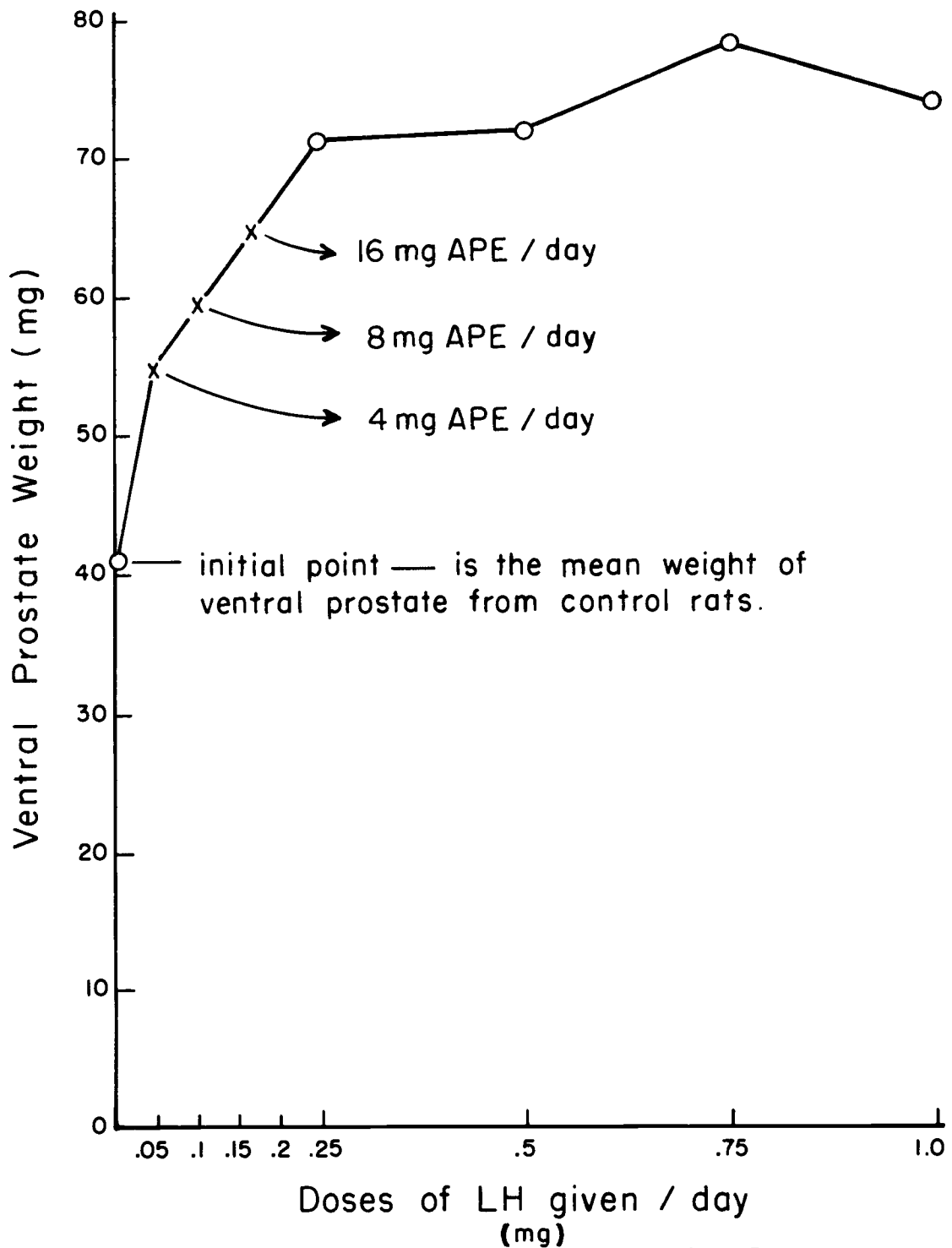


Figure 2. Dose Response Curve for Pure LH

groups are compared and plotted on the dose response curve an estimate of the amount of LH in each treatment level can be obtained.

Table 14. Analysis of variance for ewe lamb pituitary LH activity.

Source	df	MS	F
Groups (G)	10	41.8	P< 1
Breed	1	1.2	P< 1
Treatment (T)	2	590.6	7.47**
G x T	22	132.6	1.68
Error	36	79.1	
Total	71		

\*\* P<0.01

Table 15. Approximate LH levels in treatments as obtained from the dose response curve.

	Control	4mg/day <sup>a/</sup>	8mg/day	16mg/day
Mean Ventral Prostate wt. (mg)	44.1	54.9	59.4	64.8
LH level/treatment <sup>b/</sup>	--	0.04	0.11	0.17

<sup>a/</sup> mg Anterior pituitary extract per day

<sup>b/</sup> mg LH per dose of AP extract per day

Lutenizing hormone is the hormone primarily responsible for ovulation. It appears that between days 3 and 15 of the estrous cycle LH is retained in the anterior pituitary and then released sometime between 4 and 36 hours after the onset of estrus (van-Tienhoven, 1968). It is apparent that the pituitaries from these ewe lambs did contain reasonable amounts of LH. However, it is impossible to determine whether these levels are sufficiently high to cause ovulation when released into the blood. Even the slightest deviation from the intricate hormonal balance may be sufficient reason for ovulation to fail.

#### Breeding Results and Discussion

Treatment with three levels of anterior pituitary extract, as it was given, had no significant effect on increasing fertility in ewe lambs. It is believed that the propylene glycol used as a carrier had such a deleterious effect that treatment effects may have been overshadowed. Also, optimum absorption of the extract was prevented because of the tendency of the body to reject the suspension. It has been shown that APE, both that commercially obtained and that from the slaughtered ewe lambs, does have gonadotropic activity. Shelton and Moore (1967) found that horse anterior pituitary extract may be well suited to produce increase numbers of ova from sheep. Again, however, this was reported for mature anestrus ewes and not ewe

lambs. The present study confirms this in part for ewe lambs in that histological examination of the ovaries showed indications of their being fully functional with several well developed follicles present in all but one ovary. The one non-functional ovary was completely infantile.

The breed x treatment interaction was slightly significant while neither breed nor treatment alone had any effect. The means for ewe lamb fertility are slightly different. As the dose of APE increased fertility in the Willamettes decreased slightly. The Willamette ewes appeared to be more adversely affected by the propylene glycol than did the Suffolks.

Lamb birth weight was also analysed in relation to treatment and breed. Treatment total was highly significant ( $P < 0.01$ ); however, most of the variation was accounted for by breed differences which were also highly significant ( $P < 0.01$ ). Treatment differences were only slightly significant ( $P < 0.10$ ) and therefore had very little effect on birth weight of the lamb. The table of means for lamb birth weight shows this difference quite explicitly. Willamette ewe lambs consistently gave birth to heavier lambs than did the Suffolks. A large standard deviation occurred in lamb birth weights from the Suffolk ewes in the 450 mg APE treatment group. Two of the eight lambs in this group weighed 3.4 and 3.5 pounds at birth. The 3.4 pound lamb was a single lamb, the 3.5 pound lamb was a twin. The range

Table 16. Summary of breeding and lambing results for 1969.

	Breed		Overall
	Willamette	Suffolk	
Treatment I <sup>c/</sup>			
No. of ewes	17	--	17
No. of ewes lambing	8	--	8
No. of lambs born	9	--	9
Percent of ewes lambing	47.1	--	47.1
Lambing percent <sup>a/</sup>	52.9	--	52.9
Lambing percent <sup>b/</sup>	112.5	--	112.5
Treatment II <sup>d/</sup>			
No. of ewes	19	16	35
No. of ewes lambing	11	4	15
No. of lambs born	11	4	15
Percent of ewes lambing	57.9	25.0	42.9
Lambing percent <sup>a/</sup>	57.9	25.0	42.9
Lambing percent <sup>b/</sup>	100.0	100.0	100.0
Treatment III <sup>e/</sup>			
No. of ewes	19	15	34
No. of ewes lambing	9	7	16
No. of lambs born	9	8	17
Percent of ewes lambing	47.4	46.7	47.1
Lambing percent <sup>a/</sup>	47.4	53.3	50.0
Lambing percent <sup>b/</sup>	100.0	114.3	106.3

<sup>a/</sup> Based on ewes exposed to ram.

<sup>b/</sup> Based on ewes lambing.

<sup>c/</sup> Treatment - 50 mg. Anterior Pituitary Extract.

<sup>d/</sup> Treatment - 150 mg. Anterior Pituitary Extract.

<sup>e/</sup> Treatment - 450 mg. Anterior Pituitary Extract.

in lamb birth weights in this group was 3.4 to 10.5 pounds.

Three widely varied treatments of APE were given to the ewe lambs in this study with the idea being that, of the three, one treatment would exhibit optimum effects on fertility. This aspect of the study was not successful due in part to the failure of absorption of the APE in a large number of ewe lambs. It cannot be said that APE will definitely not affect fertility of ewe lambs.

Table 17. Analysis of variance for ewe lamb fertility in 1969.

Source	df	MS	F
Treatment Total	4	0.40	1.18
Treatment	2	0.20	P<1
Breed	1	0.14	P<1
B x T	1	1.00	2.94 <sup>+</sup>
Error	83	0.34	
Total	87		

<sup>+</sup>P<0.10

Table 18. Table of means and standard deviations for lamb birth weight in 1969.

Treatment	Breed	
	Willamette	Suffolk
	Lbs.	Lbs.
50 mg APE	8.86 ± 1.64 <sup>a/</sup>	-----
150 mg APE	10.18 ± 1.53	8.58 ± 1.43
450 mg APE	9.44 ± 1.60	6.69 ± 2.56

<sup>a/</sup> Mean ± standard deviation.

Table 19. Analysis of variance for lamb birth weight in 1969.

Source	df	MS	F
Treatment Total	4	15.09	4.63 <sup>**</sup>
Treatment	2	9.09	2.79 <sup>+</sup>
Breed	1	33.00	10.12 <sup>**</sup>
B x T	1	2.29	P < 1
Error	36	3.26	
Total	40		

<sup>+</sup> P < 0.10

<sup>\*\*</sup> P < 0.01



1970 Study

The principle objective of this aspect of the study was to determine not only treatment effects on fertility but also to examine how heterosis, maternal effects, general combining ability and reciprocal differences affect fertility of ewe lambs and the relationship of these factors to treatment. Traits measured included ewe birth weight, ewe weight at mating, fertility of the ewe and lamb birth weight.

The experimental animals were the female offspring of a three breed diallel cross involving the Willamette, Suffolk, and Hampshire breeds. A total of 179 ewe lambs were available as shown below:

Three breed diallel cross from which ewe lambs were drawn and the number of ewe lambs available from each cell.

		Line of Sire		
		H	S	W
Line of Dam	H	H x H 18	H x S 24	H x W 13
	S	S x H 25	S x S 20	S x W 14
	W	W x H 32	W x S 19	W x W 14

One-half of the ewes from each cell were placed on treatment, the

other half were maintained as controls. All ewes received 1/2-3/4 pound of mixed grain (barley, wheat, oats) beginning in early July and continuing through the breeding season. Otherwise, the ewes were run on non-irrigated pastures of sub-clover, fescue and perennial ryegrass. On August 5, ewes were divided randomly within cell to either the treatment or the control group. The number of each ewe was stamped on her back with red (treatment) or blue (control) marking paint. On the same day, each ewe in the treatment group received a subcutaneous injection of 150 mg anterior pituitary extract in two ml physiological saline. The pituitary extract was supplied by Mann Research Laboratories. All ewes were run together throughout the treatment and breeding season. The colored back marking facilitated distinguishing ewes on treatment from the controls. On August 21 and September 5, each treatment ewe received 200 mg APE via subcutaneous injection.

On September 12, a system of hand mating commenced. Ewes were brought into a small breeding pasture each morning and teaser rams were placed with them. As vasectomized rams were not available, three Willamette rams were fitted with copulation-prevention aprons and used as teasers. Observation of the flock was continuous as long as the teaser rams were with the ewes and any ewe that indicated that she was in estrus was recorded by number. After approximately two hours the entire flock was run through a cutting chute and

the teaser rams and ewes in estrus were removed and the remainder of the flock was returned to pasture. The teaser rams were removed and the ewe lambs in estrus were held for breeding by Shropshire rams. The Shropshire rams were registered and known fertile through past breeding records and semen testing.

Hand mating was decided upon for two primary reasons. Only two Shropshire rams were available during the breeding season, an insufficient number to breed 179 ewes under a pasture breeding system. Past experience had indicated that ewe lambs may fail to stand for servicing even though they are in estrus. By hand mating, this problem became apparent and was solved by holding the ewes that came into estrus for at least one servicing. This system was used until October 14 when the flock was divided in half and a Shropshire ram was run with each group until November 4, at which time the breeding season was terminated. Daily breeding records were kept during the pasture breeding phase.

Least squares analysis for unequal subclass numbers was used to analyze the data from this study. The model is as follows:

$$Y_{ijthk} = \mu + a_h + g_i + g_j + m_j + r_{ij} + t_{ijt} + \text{two-way interactions} + e_{ijthk}$$

where,  $Y_{ijthk}$  is the performance of the  $k^{\text{th}}$  individual from parents of the  $i^{\text{th}}$  and  $j^{\text{th}}$  lines, the  $h^{\text{th}}$  type of breeding and the  $t^{\text{th}}$  treatment.

- $\mu$  is the over-all mean
- $a_h$  is the effect common to all progeny of the  $h^{\text{th}}$  type of breeding (purebred and crossbred), the difference between these being an estimate of heterosis.
- $g_i$  is the general combining ability of the  $i^{\text{th}}$  line, the sire line.
- $g_j$  is the general combining ability of the  $j^{\text{th}}$  line, the dam line.
- $m_j$  is the maternal effect for the  $j^{\text{th}}$  line of dam.
- $r_{ij}$  is the residual of the reciprocal or sex-linked effect with the maternal effect removed.
- $t_{ijt}$  is the treatment effect on the  $i^{\text{th}}$  and  $j^{\text{th}}$  line progeny with the  $t^{\text{th}}$  treatment (treated and control).

two-way interactions included:

Heterosis x Treatment

Purebred x Treatment

General combining ability x Treatment

Maternal effect x Treatment

Reciprocal effect x Treatment

$e_{ijthk}$  is the random error.

Variance components and percentage variation was calculated for each of the factors measured using the following procedure.

$$\text{Where, } \sigma_i^2 = \frac{(\sigma_e^2 + K \sigma_h^2) - \sigma_e^2}{K}$$

$\sigma_i^2$  is the variance component.

$\sigma_h^2$  is the variance component for heterosis.

$\sigma_e^2$  is the error mean square.

K is a function of the number of observations per class with unequal subclass numbers.

Variance components for all factors including the error term were added and the total was divided into each individual component to obtain the percentage variation.

## Results and Discussion

### Ewe Birth Weight

In general, the factors analyzed had little or no effect on ewe birth weight (EBW). Ninety percent of the variation in EBW was relegated to the error. Heterosis effects approached significance at  $P < 0.10$  and accounted for 2.60 percent of the variation in EBW. General combining ability and maternal effects were non-significant; however, each accounted for 1.95 and 2.16 percent of the variation respectively. Comparison of the reciprocal crossbreds with the mid-parent averages show that the crossbreds were 0.6 pounds heavier at birth. Humes (1969) reported that for the female offspring

all linecross groups were smaller at birth than the corresponding inbred beef lines. Gaines et al. (1966) found some evidence for heterosis in birth weights of two-way beef breed crosses. They also found little, if any, evidence of maternal influence in their two-breed crosses which is consistent with results obtained in the present study. Schilling et al. (1968) studied combining abilities of three inbred lines of Suffolk sheep and found that the linecross lambs were generally heavier at birth than the straight-bred lambs.

Even though the effects of factors other than heterosis are statistically non-significant, it can be seen from Table 23 that differences do exist. Willamette and Suffolk ewe lambs were very nearly the same in weight at birth with both being heavier than the Hampshire ewe lambs. General combining ability of the Willamette ewe lambs was greater than that of either the Suffolk or the Hampshire with the Hampshire having the poorest general combining ability. Maternal influence was the greatest in the Hampshire.

The birth weight and subsequent preweaning growth of the lamb is influenced by the inherent genetic potential of the lamb and the maternal influence of the dam. Brown and Galvez (1969) reported that the effect of the cow on birth weight of her calves accounted for 17.6% of the total variation in birth weight of Hereford and 9.3% for Angus calves. Knapp et al. (1942) found that differences between cows accounted for about 19.0% of the total variation in birth weight

Table 20. Summary of breeding and lambing results for 1970.

	Genetic groups									Overall
	HxH <sup>c/</sup>	SxS	WxW	SxW	WxS	HxW	WxH	HxS	SxH	
Treated										
No. of ewes	9	10	7	8	9	7	16	12	13	91
No. of ewes lambing	5	1	5	5	7	4	11	6	10	54
No. of lambs born	6	1	5	7	9	5	13	7	10	63
Percent of ewes lambing	55.6	10.0	71.4	62.5	77.8	57.1	68.8	50.0	76.9	59.3
Lambing percent <sup>a/</sup>	66.7	10.0	71.4	87.5	100.0	71.4	81.3	58.3	76.9	69.2
Lambing percent <sup>b/</sup>	120.0	100.0	100.0	140.0	128.6	125.0	118.3	116.7	100.0	116.7
Control										
No. of ewes	9	10	7	6	10	6	16	12	12	88
No. of ewes lambing	7	6	3	3	8	2	10	9	9	57
No. of lambs born	7	6	3	3	11	3	10	9	9	61
Percent of ewes lambing	77.8	60.0	42.9	50.0	80.0	33.3	62.5	75.0	75.0	64.8
Lambing percent <sup>a/</sup>	77.8	60.0	42.9	50.0	110.0	50.0	62.5	75.0	75.0	69.3
Lambing percent <sup>b/</sup>	100.0	100.0	100.0	100.0	137.5	150.0	100.0	100.0	100.0	107.0

<sup>a/</sup> Based on ewes exposed to ram.

<sup>b/</sup> Based on ewes lambing.

<sup>c/</sup> Dam x Sire, Willamette, Suffolk, Hampshire

Table 21. Least squares analysis of variance for ewe birth weight.

Source	df	MS	F
Heterosis	1	12.789	3.073 <sup>+</sup>
Purebreds	2	1.032	P<1
General combining ability	2	6.223	1.495
Maternal effect	2	5.899	1.417
Reciprocal effect	1	1.674	P<1
Treatment	1	0.196	P<1
H x T	1	15.521	3.729 <sup>+</sup>
P x T	2	2.171	P<1
GCA x T	2	2.690	P<1
M x T	2	0.023	P<1
R x T	1	0.640	P<1
Error	174	4.162	
Uncorrected Total	192		

<sup>+</sup> P<0.10



Table 22. Estimates of variance components and percentage variation in ewe birth weight.

Source	df	$\sigma_i^2$	%
Heterosis	1	0.12	2.60
Purebreds	2	<0.00	0.00
General combining ability	2	0.09	1.95
Maternal effect	2	0.10	2.16
Reciprocal effect	1	<0.00	0.00
Treatment	1	<0.00	0.00
H x T	1	0.15	3.25
P x T	2	<0.00	0.00
GCA x T	2	<0.00	0.00
M x T	2	<0.00	0.00
R x T	1	<0.00	0.00
Error	174	4.16	90.04

Table 23. Means and deviations from the overall mean<sup>a/</sup> of ewe birth weight, 9.73 lbs. as influenced by the factors measured.

	<u>Hampshire</u>	<u>Suffolk</u>	<u>Willamette</u>
Purebreds	9.20 - .53	9.99 + .26	10.00 + .27
General combining ability	9.01 - .72	9.89 + .16	10.29 + .56
Maternal effect	10.17 + .44	9.23 - .49	9.78 + .05
	<u>Purebreds</u>	<u>Crossbreds</u>	
Heterosis	9.43 - .30	10.02 + .30	

<sup>a/</sup> Least squares means and constant effects.

of their calves. In the present study, the maternal effect accounted for only 2.2% of the total variation in birth weight in the crossbred and straight bred ewe lambs.

It is evident that with 90% of the total variation in the error term, the actual causes of variation in birth weight were not measured. The ewes in the diallel cross from which these ewe lambs were obtained were subjected to very different environmental conditions during the breeding season and subsequent gestation period. One-half of the ewes in the entire diallel cross were bred and maintained on irrigated pasture with supplemental alfalfa hay and grain. The remaining one-half were placed on non-irrigated pasture during the breeding season and gestation also with supplemental hay and grain. Therefore, it can be concluded that much of the total variation which was not measured is due to the environmental interaction.

#### Ewe Weight at Mating

Ewe weights were taken on 29 October 1970 or approximately one week prior to the end of the breeding season. Weights were also taken on 5 August 1970. Ewe lambs in the control group gained on the average 11.33 pounds in this period, whereas the ewes in the treatment group gained 9.64 pounds. The overall treatment effect on ewe weight at mating was non-significant. However, as the actual treatment procedure itself was the only difference between groups

(Treated and Control) and there was a difference of 1.69 lb. in weight gain from 5 August to 29 October it can be assumed that the difference is due either to the extra handling required for the injections or to the treatment itself.

The mean ewe weight at mating (29 October) was 95.08 lb. The Suffolk ewes were the heaviest, averaging 104.59 lb. and the Willamette ewes were the lightest, 85.71 lb. This difference among purebreds was highly significant ( $P < 0.01$ ), and accounts for over 23% of the total variation in ewe weight at mating. It is interesting to note that the pattern found for ewe birth weight in general combining ability and maternal effect is nearly the same as that for ewe weight at mating. General combining ability of the Willamettes exceeded that of either of the other two breeds ( $P < 0.05$ ) even though it accounted for only 2.99% of the total variation. Maternal effect was also highly significant for ewe weight at mating ( $P < 0.025$ ). The Hampshire again showed evidence of having a greater maternal influence than either of the other two breeds. It is clear that the Hampshire has a marked maternal influence on both pre- and post-natal growth of the ewe lamb.

The heterotic effect on ewe weight at mating was highly significant ( $P < 0.01$ ), with the crossbred ewes approximately 5.5 lbs. heavier than the purebreds. It is clear that a substantial amount of genetic divergence exists among these breeds to cause the significant

Table 24. Least squares analysis of variance for ewe weight at mating.

Source	df	MS	F
Heterosis	1	1097.19	6.733 <sup>**</sup>
Purebreds	2	1029.65	6.318 <sup>**</sup>
General combining ability	2	523.21	3.211 <sup>*</sup>
Maternal effect	2	739.06	4.535 <sup>+*</sup>
Reciprocal effect	1	1280.71	7.859 <sup>**</sup>
Treatment	1	293.98	1.804
H x T	1	642.04	3.940 <sup>*</sup>
P x T	2	206.89	1.270
GCA x T	2	154.35	P < 1
M x T	2	92.55	P < 1
R x T	1	26.85	P < 1
Error	174	162.96	
Uncorrected Total	192		

\* P < 0.05

+\* P < 0.025

\*\* P < 0.01

Table 25. Estimates of variance components and percentage variation for ewe weight at mating.

Source	df	$\sigma_i^2$	%
Heterosis	1	12.74	2.47
Purebreds	2	119.03	23.05
General combining ability	2	15.44	2.99
Maternal effect	2	32.43	6.28
Reciprocal effect	1	159.40	30.87
Treatment	1	1.79	0.35
H x T	1	6.35	1.26
P x T	2	6.03	1.17
GCA x T	2	<0.00	0.00
M x T	2	<0.00	0.00
R x T	1	<0.00	0.00
Error	174	162.96	31.56

Table 26. Means and deviations from the overall mean<sup>a/</sup> of ewe weight at mating, 95.08 lbs. as influenced by the factors measured.

	<u>Hampshire</u>	<u>Suffolk</u>	<u>Willamette</u>
Purebreds	94.94 - .14	104.59 + 9.51	85.71 - 9.37
General combining ability	88.34 - 6.74	96.00 + .92	100.91 + 5.82
Maternal effect	107.76 + 12.68	90.93 - 4.15	86.56 - 8.53
	<u>Purebreds</u>	<u>Crossbreds</u>	
Heterosis	92.35 - 2.73	97.82 + 2.73	
	<u>Treated</u>	<u>Control</u>	
Treatment	93.67 - 1.42	96.50 + 1.42	

<sup>a/</sup> Least squares means and constant effects.

heterotic effect even though heterosis accounts for only 2.47% of the total variation.

The reciprocal or sex-linkage effect is somewhat difficult to interpret. The effect itself is highly significant ( $P < 0.01$ ) because there have been adjustments made for the maternal effect by the model used. This significance lends support to the presence of substantial genetic diversity between the breeds particularly since the reciprocal effect is corrected for maternal influence.

Ewe weight at mating was the most significant variable of those analyzed. The factors measured accounted for nearly 70% of the total variation in ewe weight at mating. In general the two-way interactions were not significant.

As mentioned previously, it is apparent that ewe birth weight was influenced greatly by the non-uniformity of the environment. This is evidenced by the fact that when the environment was standardized following weaning of the ewe lambs, the variation due to environmental factors was significantly reduced; therefore, it became possible to accurately measure the variation due to the type of breeding, genetic potential and parental influence.

#### Ewe Lamb Fertility

Fertility is a threshold or an 'all or none' character. When it comes to measuring fertility in the sense that it is being used here



the measurement is completely objective. Either a ewe conceives and gives birth to a lamb or she does not. There are intermediate expressions such as failure to implant or spontaneous abortion but these are difficult, if not impossible, to measure. Even if the intermediate expression is observed, it would be extremely difficult to measure objectively. As previously stated, the measure of fertility in the present study involved assigning the values 1, 2 and 3 to a ewe if she was barren or gave birth to a single or to twin lambs respectively.

Overall lambing percent for the study was 71%. This percentage is based on the number of lambs born in relation to the total (179) number of ewe lambs.

The effect of treatment was not significant. However, the least square mean for the treated versus control shows that the lambing percent of the treated group was about 4% higher than that of the controls. The two-way interactions involving treatment were, in general, not significant. The purebred x treatment interaction, however, approached significance ( $P < 0.10$ ). This significant interaction was due to the fact that only one of the ten purebred Suffolks in the treated group lambbed and she gave birth to a single lamb. Of the ten Suffolk ewe lambs in the control group, six gave birth to single lambs. This adverse response of the Suffolks to treatment was fairly consistent throughout the five-year study. The most

Table 27. Least squares analysis of variance for ewe lamb fertility.

Source	df	MS	F
Heterosis	1	2.889	7.081 <sup>**</sup>
Purebreds	2	0.123	P<1
General combining ability	2	1.506	3.691 <sup>+*</sup>
Maternal effect	2	0.608	1.491
Reciprocal effect	1	0.853	2.090
Treatment	1	0.072	P<1
H x T	1	0.430	1.055
P x T	2	1.164	2.854 <sup>+</sup>
GCA x T	2	0.403	P<1
M x T	2	0.583	1.430
R x T	1	0.572	1.408
Error	174		
Uncorrected Total	192		

<sup>+</sup> P<0.10

<sup>+\*</sup> P<0.025

<sup>\*\*</sup> P<0.01

Table 28. Estimates of variance components and percentage variation in ewe lamb fertility.

Source	df	$\sigma_i^2$	%
Heterosis	1	0.033	4.72
Purebreds	2	<0.000	0.00
General combining ability	2	0.047	6.72
Maternal effect	2	0.011	1.57
Reciprocal effect	1	0.063	9.01
Treatment	1	<0.000	0.00
H x T	1	<0.000	0.00
P x T	2	0.104	14.88
GCA x T	2	<0.000	0.00
M x T	2	0.010	1.43
R x T	1	0.023	3.29
Error	174	0.408	58.37

Table 29. Means and deviations from the overall mean<sup>a/</sup> lambing percent of 71.0% as influenced by the factors measured.

	<u>Hampshire</u>	<u>Suffolk</u>	<u>Willamette</u>
	%	%	%
Purebreds	61.0 - 10.0	62.5 - 8.5	89.5 + 18.5
General combining ability	37.9 - 33.1	83.9 + 12.9	91.2 + 20.2
Maternal effect	102.9 + 31.9	57.5 - 13.5	52.6 - 18.4
	<u>Purebreds</u>	<u>Crossbreds</u>	
	%	%	
Heterosis	57.0 - 14.0	85.0 + 14.0	
	<u>Treated</u>	<u>Control</u>	
	%	%	
Treatment	73.2 + 2.2	68.8 - 2.2	

<sup>a/</sup> Least squares means and constant effects.

likely explanation is that the Suffolk ewe lambs are more sensitive reproductively than either of the other two breeds.

The heterotic effect on fertility was highly significant ( $P < 0.01$ ) which is consistent with results reported by Botkin and Paules (1965), Fox and McArthur (1963). The mean lambing percent for the purebreds was 57.0 and for the crossbreds 85.0. Of the 27 purebred ewes that lambed, only one ewe gave birth to twins. However, of the 84 crossbred ewes that gave birth to lambs, 12 ewes had twins. This small difference in the number of multiple births was not analyzed for statistical significance. Of the total variation in fertility, the heterotic effect accounted for 4.72% indicating that the effects of dominance may be small in relation to the total variation but important in its overall influence.

Breed differences were not statistically significant; however, fertility of the Willamette ewe lambs exceeded that of the Hampshire and Suffolk ewe lambs by nearly 30%. The Willamette as a breed is relatively new. It was developed from crosses between the Border Cheviot, Columbia, and Dorset Horn breeds. It is likely that sufficient genetic variation still exists within the Willamette breed to account for an apparent heterotic response; whereas, the older and better established Hampshire and Suffolk breeds responded accordingly.

General combining ability was highly significant for fertility

( $P < 0.025$ ) with the Willamette having the best general combining ability of the three breeds. As in ewe birth weight and ewe weight at mating, the Hampshire was the poorest in general combining ability, 10% below the mean of the population. However, in maternal effect the Hampshire exceeded by far the Suffolk and Willamette breeds. It is generally accepted that the Hampshire ewe gives birth to a larger lamb and supplies more milk to the growing lamb.

#### Lamb Birth Weight

As stated previously, the offspring from all the ewe lambs were sired by Shropshire rams. Lamb birth weight was not significantly influenced by any of the factors measured. The pattern established for the difference in breeds, general combining ability, and maternal effect for ewe birth weight, weight at mating, and fertility was also found for lamb birth weight. The Willamettes produced the largest lambs and were the best in general combining ability. The Hampshires showed the greatest maternal effect. The lack of significant effects of general combining ability and reciprocal effects for lambs produced by linecross ewes is consistent with results reported by Schilling, et al. (1968).

The purebred x Shropshire lambs were slightly heavier than the lambs produced by the crossbred ewes, about 0.36 lbs.

Table 30. Least squares analysis of variance for lamb birth weight.

Source	df	MS	F
Heterosis	1	1.797	P<1
Purebreds	2	3.854	P<1
General combining ability	2	4.881	1.143
Maternal effect	2	2.686	P<1
Reciprocal effect	1	5.724	1.340
Treatment	1	2.606	P<1
H x T	1	0.268	P<1
P x T	2	4.384	1.027
GCA x T	2	4.856	1.137
M x T	2	8.060	1.888
R x T	1	11.595	2.715
Error	105	4.270	
Uncorrected Total	123		

Table 31. Estimates of variance components and percentage of variation in lamb birth weight.

Source	df	$\sigma_i^2$	%
Heterosis	1	<0.00	0.00
Purebreds	2	<0.00	0.00
General combining ability	2	0.04	0.59
Maternal effect	2	<0.00	0.00
Reciprocal effect	1	0.34	5.01
Treatment	1	<0.00	0.00
H x T	1	<0.00	0.00
P x T	2	0.04	0.59
GCA x T	2	0.04	0.59
M x T	2	0.33	4.88
R x T	1	1.70	25.15
Error	105	4.27	63.17



Table 32. Means and deviations from the overall mean<sup>a/</sup> of lamb birth weight, 10.18 lbs. as influenced by the factors measured. <sup>b/</sup>

	<u>Hampshire</u>	<u>Suffolk</u>	<u>Willamette</u>
Purebreds	8.65 - 1.53	10.18 0.00	11.71 + 1.53
General combining ability	10.29 + .11	9.62 - .56	10.63 + .45
Maternal effect	11.27 + 1.09	10.24 + .06	9.03 - 1.15
	<u>Purebred</u>	<u>Crossbred</u>	
Heterosis	10.36 + .18	10.00 - .18	
	<u>Treated</u>	<u>Control</u>	
Treatment	9.95 - .21	10.39 + .21	

<sup>a/</sup> Least squares means and constant effects.

<sup>b/</sup> Lambs are the offspring of purebred and linecross dams x Shropshire sires.

This small heterotic response is not statistically significant; however, the response is in the expected direction. The heterosis of the  $F_1$  lambs should exceed the heterosis of the three breed cross lambs because of the heterotic response already expressed by the crossbred females.

## SUMMARY AND CONCLUSIONS

In 1965 a series of studies were initiated to determine what effects various exogenous gonadotropin treatments would have on fertility of ewe lambs. Treatment procedures over the five programs included the use of pregnant mare serum, anterior pituitary extract, chorionic gonadotropin, and 17-beta-estradiol. These compounds were utilized alone and in various combinations in an attempt to stimulate breeding activity and fertility in ewe lambs. Repromix and other progestational compounds were used in attempts to synchronize estrus in several of the studies but was generally found to be ineffective in young ewes. During the 1969 phase of the study a more in-depth look was taken at the actual effects of treatment. Biological assays of anterior pituitary extract were run to determine its biological activity. Histological examinations of uteri and ovaries taken from treated and control ewe lambs were made to find out if treatment had any effect on the histology of the reproductive system.

Two breeds of lambs (Willamette and Suffolk) were utilized throughout the program until the 1970 phase. This allowed for breed differences to be analyzed as well as breed-treatment interactions. The 1970 phase of the study made use of the female offspring of a three breed diallel cross between the Willamette, Suffolk and Hampshire breeds. This experimental approach provided an opportunity

to determine and measure the genetic aspects of fertility. Variables measured included ewe birth weight, ewe weight at mating, fertility and lamb birth weight. Factors analyzed for their relationship to the variables mentioned were heterosis, general combining ability, maternal effect, reciprocal or sex-linkage effects, the effects of treatment and two-way interactions between treatment and the other factors.

In general, the effects of treatment were found to be statistically non-significant. None of the treatments attempted increased fertility of ewe lambs either among treated and control ewes or among different treatments for a given year. In some cases, particularly in the Suffolk breed, treatment had an adverse effect on fertility by reducing the number of ewes lambing in several instances. One conclusion is readily apparent from this lack of treatment effectiveness. The hormonal balance that influences sexual maturity and subsequent reproductive function is very delicate. Until the relationship between various hormones is better understood and means of accurately measuring intrinsic hormone levels is found, any attempt to augment this balance with exogenous hormone treatment will be arbitrary. Rapid advances, however, are being made in measuring hormone levels through the use of radioimmunological techniques and this will be of great value to further work of this type.

The histological studies showed no apparent reason for

implantation to fail should conception occur. The reproductive tract appeared to be normal in all respects. Glandular development in the endometrium of the uterus was extensive and consistent throughout the samples from the various treatment groups.

Bowstead (1930) listed some of the common problems that exist when ewes are bred as lambs. The smaller size of the ewe lamb at lambing created some difficulties during the course of this program. Several times ewes had to be assisted in giving birth due to the smaller pelvic capacity. Ewe lambs were also generally found to be less effective mothers than mature ewes either because of a lack of sufficient mammary development and lower milk production or disinterest in the lamb once it is born. Usually the ewes that had the most parturition difficulties were the ones that tended to abandon their lambs. Under good management conditions these problems are not sufficient reason for the breeding of ewe lambs to be considered impractical.

The existence of heterosis in fertility indicates that a sound system of rotational crossbreeding may be most beneficial to a commercial lamb producer. Not only would the breeding of ewe lambs allow a ewe to produce an additional lamb, but it would also allow the benefit of achieving the heterotic response normally found for vitality and faster, more efficient growth.

The other genetic factors measured had little or no influence

on fertility in ewe lambs. Maternal effect and reciprocal differences had no effect while general combining ability was an influencing factor. Of the three breeds studied in the last phase of this study the Willamette ewe lambs had a better general combining ability than either of the other two breeds. However, the Willamette ewe lambs had the lowest maternal effect for all factors measured while the Hampshire was far above the Suffolk and Willamette in maternal influence.

Results indicate that the fertility of crossbred ewe lambs is significantly greater than that of the purebreds. From this study it is apparent that the best cross would be between the Hampshire and Willamette with the Hampshire as the dam line. Until more information is available on hormone levels and their effects, exogenous gonadotropin treatment will be relatively ineffective in increasing lamb production from ewe lambs.

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