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GENOTYPE-PASTURE INTERACTION IN THE FERTILITY OF ROMNEY MARSH EWES

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Massey University College, Palmerston North

SUMMARY
A comparison was made of reproductive performance of Romney Marsh ewes grazed on an oestrogenic red clover sward and on a control pasture. Ewes grazed on oestrogenic red clover were lower in fertility than ewes grazed on control pasture. Not in the control ewes but among the red clover ewes there was a differential reduction in fertility according to whether the ewes were themselves born as singles or as twins. The results of present work are discussed with reference to the planning of experiments for the study of reproduction in the ewe.

INTRODUCTION
In recent years a considerable amount of research work has been devoted to the study of phenomena known as heredity-environment interaction. The existence and the extent of such an interaction is usually inferred from the differential response of an array of genotypes when exposed to different sets of environmental conditions. The present study reports an observed differential response in fertility between two genotypes within a group of Romney Marsh ewes grazed on two contrasting types of pasture.

EXPERIMENTAL
Pasture
The control pasture was predominantly a ryegrass/white clover association which is normally available for sheep grazing in the Manawatu. The contrasting type of pasture was practically a pure sward of Montgomery red clover (Trifolium pratense, late-flowering variety).

Red clover is known to contain in varying quantities, the isoflavones—biochanin A, genistein and formononetin (Pope, 1951) and, more recently, Wong (1962) has detected, in the red clover, another isoflavone—daidzein. While all of the above-mentioned isoflavones contribute to the oestrogenic properties of the red clover, Wong and Flux (1962) reported that most of the oestrogenic activity in a sample of New Zealand Montgomery red clover was due to biochanin A.
Samples of red clover pasture grazed by sheep in the present study were tested for oestrogenic activity by the mouse uterine weight technique. These tests showed that the red clover was oestrogenic during the period of utilization (D. S. Flux, R. E. Munford and T. S. Ch'ang, pers. comm.).

**Genotype of Ewes**

The ewes in this study were drawn from an experimental flock maintained at Massey University College. No selection for or against fertility has been practised in this flock since its founding in 1944. Rae and Ch'ang (1955) reported that, in this flock, ewes which were born as twins had a higher lambing percentage than ewes which were born as singles. Wallace (1958), presenting evidence from a Romney selection experiment, showed that a higher fertility was obtained from a flock in which most of the ewes were born as twins than another flock in which most of the ewes were born as singles. Furthermore, from a selection experiment for multiple births in Merino ewes, Newtown-Turner *et al.* (1962) recorded a higher lambing percentage among ewes born as twins than those born as singles. All the above evidence suggests that there is a difference in average genotype between twin-born and single-born ewes with respect to fertility. Therefore, in the present study, a distinction was made between ewes according to whether they were born as singles or twins.

**Sheep, Grazing and Mating Management**

On December 7, 1959, forty-five 5½-year-old Romney ewes were randomized into two groups. The group which subsequently grazed on the red clover pasture was allotted 25 ewes and designated as group R. The control group, consisting of 20 ewes, was designated as group C. Except for a period of 3½ months from January 12 to April 26 in 1960, when ewes of group R were grazed on the red clover pasture, both groups of ewes were managed and grazed as one mob on the control pasture during 1960 and until the end of the experiment in December, 1961.

In 1960, the rams were joined with the ewes for 55 days from March 2 to April 26. Since the ewes in group R and group C were confined separately on the red clover and the control pasture during mating, interchange of rams between these two groups was made at weekly intervals throughout the mating period.
During 1961, the ewes were mated as one mob on the control pasture from March 3 to April 28. After weaning in December, 1961, all ewes were slaughtered and examined for endometrial cysts and other abnormalities. None was found.

RESULTS

LIVESTOCK OF EWES

The ewes were weighed on December 7, 1959, at the start of the experiment, on March 2, 1960, when the ewes were joined with the rams, and again on April 26, 1960, when the rams were withdrawn. Liveweights of ewes were not available for 1961.

TABLE 1: AVERAGE LIVESTOCK (LB) AND CHANGES OF AVERAGE LIVESTOCK DURING THE MATING PERIOD

<table>
<thead>
<tr>
<th></th>
<th>7.12.59</th>
<th>2.3.60</th>
<th>2.3.60 to 26.4.60</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Red Clover</td>
<td>Control</td>
<td>Red Clover</td>
</tr>
<tr>
<td>Ewes born</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>as singles</td>
<td>.....</td>
<td>148 (14)*</td>
<td>148 (10)</td>
</tr>
<tr>
<td>Ewes born</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>as twins</td>
<td>.....</td>
<td>143 (11)</td>
<td>143 (10)</td>
</tr>
</tbody>
</table>

* (Number of ewes.)
† Significant at the 1% level.

It can be seen from Table 1 that, at the start of the experiment, there was no difference in the average liveweights between ewes allotted to various groups. The differences in average liveweight between the red clover and the control groups at the beginning of the mating period were statistically significant. The lower liveweight of the control ewes was probably due to the fact that they were confined for periods during January and February in 1960 as a precaution against facial eczema. The red clover ewes, however, maintained their liveweight while they remained on the red clover pasture. The changes in the average liveweights were negligible during the mating period.

FERTILITY OF EWES

A more detailed account of the extent of reduction in fertility of Romney ewes grazed on red clover pasture has already been published (Ch'ang, 1961). For the present purpose, the lambing records available for comparison
consisted of the date of lambing and the performance of the ewe at lambing.

(1) *Average date of lambing:* The coded dates of lambing are presented in Table 2. Analysis of variance of the date of lambing within each year showed that the effects due to type of birth, pasture and the interaction between them were not significant at the 5% level.

**Table 2: Average Dates of Lambing**

<table>
<thead>
<tr>
<th></th>
<th>1960</th>
<th>1961</th>
<th>Grazed on Red Clover 12.1.60 to 26.4.60</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Red Clover</td>
<td>Control</td>
<td></td>
</tr>
<tr>
<td>Ewes born as singles</td>
<td>16 (6)*</td>
<td>12 (9)</td>
<td>19 (10)</td>
</tr>
<tr>
<td>Ewes born as twins</td>
<td>19 (9)</td>
<td>19 (8)</td>
<td>27 (7)</td>
</tr>
</tbody>
</table>

* (Number of ewes which lambed.)

(2) *Lambing percentage:* The lambing percentage was measured by all lambs born, dead or alive, as a percentage of the total number of ewes put to the rams, excluding ewe deaths in each year. The results for 1960 are given in Table 3.

**Table 3: Average Lambing Percentages in 1960**

<table>
<thead>
<tr>
<th></th>
<th>Red Clover</th>
<th>Control</th>
<th>Difference</th>
</tr>
</thead>
<tbody>
<tr>
<td>Ewes born as singles</td>
<td>64 (14)*</td>
<td>140 (10)</td>
<td>-76</td>
</tr>
<tr>
<td>Ewes born as twins</td>
<td>118 (11)</td>
<td>140 (10)</td>
<td>-22</td>
</tr>
<tr>
<td><strong>Interaction</strong></td>
<td></td>
<td></td>
<td>-54</td>
</tr>
</tbody>
</table>

* (Number of ewes.)

It can be seen that there was an overall reduction in lambing percentage of the ewes grazed on red clover pasture in comparison with the ewes grazed on the control pasture. In addition, there was a differential reduction in lambing percentage according to the type of birth of the ewes. Thus, while the ewes born as singles were 76% lower in lambing percentage than that of the control group, the ewes born as twins grazed on the same oestrogenic red clover were less severely affected. The extent of this differential reduction in lambing percentage was 54%.
Table 4 shows the lambing percentages of these same groups of ewes all grazed and mated on the control pasture during 1961.

The difference in lambing percentages between the groups of ewes within each type of birth, as shown in Table 4, was negligible. Furthermore, there was no evidence of differential response in fertility between the single-born and twin-born ewes.

While the lambing percentages given in Tables 3 and 4 were informative, no valid method could be found to test the statistical significance of the genotype-pasture interaction in data thus arranged. However, the lambing percentage is a function of the proportions of barren, single- and twin-bearing ewes, and therefore the data were re-arranged for statistical analysis in the following manner:

1. The number of barren ewes in each subclass was expressed as a percentage of total ewes in that subclass, and
2. The number of ewes producing twin lambs in each subclass was expressed as a percentage of the number of ewes actually lambed in that subclass.

The data were thus regarded as samples from the binomial distribution. A method involving the angular transformation of percentages, as outlined by Mather (1951) was used to test the statistical significance of the genotype-pasture interaction in barrenness and in the incidence of twinning. Statistical tests showed that the genotype-pasture interaction was significant in the percentage of barrenness in 1960. These results, together with those on lambing percentages given earlier, suggest the existence of a genotype-pasture interaction in the fertility of aged Romney ewes.

### Table 4: Average Lambing Percentages in 1961

<table>
<thead>
<tr>
<th>Grazed on</th>
<th>Red Clover</th>
<th>Control</th>
<th>Difference</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>12.1.60 to 26.4.60</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Ewes born as singles</td>
<td>127 (11)*</td>
<td>125 (8)</td>
<td>+2</td>
</tr>
<tr>
<td>Ewes born as twins</td>
<td>157 (7)</td>
<td>155 (9)</td>
<td>+2</td>
</tr>
<tr>
<td><strong>Interaction</strong></td>
<td></td>
<td></td>
<td>0</td>
</tr>
</tbody>
</table>

* (Number of ewes.)
INTERACTION IN FERTILITY OF EWES

DISCUSSION

At the start of the mating period in 1960, the ewes grazed on the red clover pasture were considerably heavier than those of the control group. In view of the positive relationship usually found between the average liveweight of ewes near mating and their subsequent lambing percentage as shown by Wallace (1961) and Coop (1962), it might have been expected that the red-clover-fed ewes would be higher rather than lower in lambing percentage than the control ewes. Therefore it is unlikely that the reduced lambing percentage of the red-clover ewes could be attributed to their average liveweight at the start of the mating period. It is less likely that the negligible changes of average liveweight during the mating period could account for the difference in reproductive rate between the red-clover and the control ewes. Nor is it likely that the fertility of the rams used in 1960 had differentially affected the lambing percentage of the red-clover and control ewes since regular interchanges of rams between these two groups were made during the mating period.

There appears little doubt that the lower lambing percentage of the red-clover-fed ewes in 1960 was primarily a result of the oestrogenic properties of the legume. That this reproductive disturbance was transient, was shown by the comparable lambing percentages of the various groups in 1961 when all ewes were grazed on the control pasture. In addition, the observed interaction between genotype and pasture in the fertility of the ewes was characteristically associated with the grazing of oestrogenic red clover in 1960 since this interaction was non-existent in 1961 in the absence of the red-clover-grazing treatment.

The present results point to the desirability of having information as to whether the sheep were born as singles or twins in the planning of experiments designed to study reproduction in the ewe. In addition, the controlled grazing of an oestrogenic legume, such as Montgomery red clover, may prove to be a useful experimental technique whereby the expression in fertility between ewes of different types of birth is enlarged to facilitate detailed physiological studies of reproduction under field conditions.

ACKNOWLEDGEMENT

The author wishes to record his thanks to W. R. R. Hewitt and his staff for the care of the experimental sheep; to A. C. Glenday, of the Applied Mathematics laboratory,
D.S.I.R., for advice on a method of statistical analysis; and to Professor A. L. Rae for his interest and helpful comments at various stages of this work.

REFERENCES


DISCUSSION

Q: Could dietary factors other than oestrogenic substances not be involved in the difference of fertility between red clover and control ewes?

T. S. CH'ANG: No studies have been made on dietary factors in the red clover or their effects on the fertility of the ewe.

Q: Is there any evidence as to whether the differences in lambing percentage were due to fewer ova being shed, or to fewer being implanted?

MR CH'ANG: A few ewes from the red clover and the control groups were laparotomized for ovarian examination. No difference was found in counts of corpora lutea between groups. It is possible that the difference in lambing percentage between the red clover fed and the control ewes was either due to failure of fertilization or implantation. More detailed data on this aspect have been published elsewhere (Ch'ang, 1961).

DR A. H. CARTER: The interaction considered here is essentially different from the usual concept, in that the two "genotypes" were distinguished by their phenotypic fertility, itself the subject of investigation. This conditions application of the results to practical selection problems.

Further, the observed interaction could be apparent rather than real, in that it could depend on the recording scale used. That is, a constant reduction in ovulation rate (i.e., no interaction) might be expected to result in a differential reduction in the proportion of barren ewes in "twin" as compared with "single" genotypes.

MR CH'ANG: While Dr Carter's second comment on the recording scale is academically important, it may be pointed out that, in practice, the underlying scale of fertility in the ewe is not known. The recording scale in question—i.e., the proportion of barren ewes—nevertheless, is a scale commonly used as a measure of infertility.