Artificial Induction of Lactation: 
A Comparative Study in Heifers

W. J. Fulkerson

Wokalup Research Station, Wokalup, W.A. 6221.
Present address: Department of Animal Science and Production, Institute of Agriculture, 
University of Western Australia, Nedlands, W.A. 6009.

Abstract

A comparison was made between four methods for artificial induction of lactation in fertile maiden heifers. Animals lactating after pregnancy were used as controls. Twenty-nine heifers were allocated to five groups and injected subcutaneously as follows:

- **Group 1:** 35 mg β-oestradiol plus 87.5 mg progesterone twice daily for 7 days.
- **Group 2:** 10 mg β-oestradiol-3-benzoate plus 100 mg progesterone every 3 days for 42 days.
- **Group 3:** 2.4 mg β-oestradiol-3-benzoate plus 600 mg progesterone every 3 days for 42 days.
- **Group 4:** As for group 3.
- **Group 5:** Heifers calving at the same time as the remaining heifers began to milk.

Heifers in groups 2, 3 and 4 received 20 mg dexamethasone 3 days later for two consecutive days. All heifers were milked from 5 days after the last injection except group 4 heifers which were suckled by calves for 4 weeks. For 300 days milk production was 2051 ± 429, 1965 ± 283, 1986 ± 200, 1862 ± 637, and 3205 ± 501 kg for groups 1–5 respectively.

If relative costs of each treatment are considered, treatment groups 1 and 2 offer the most promise. However, heifers in both groups, particularly group 1, displayed continuous nymphomania for prolonged periods.

Introduction

There have been numerous attempts to induce lactation artificially using the hormone oestrogen, alone or in combination with progesterone (Folley 1952; Meites 1961). These hormones were either implanted (Folley and Malpress 1944; Hammond and Day 1944; Reineke *et al.* 1952) or injected subcutaneously (Williams and Turner 1962). However, the variable response, below-normal milk production and the long period of treatment made these methods commercially unattractive.

Since then, workers in the U.S.A. (*Smith et al.* 1973; Smith and Schanbacher 1973, 1974) have reported relatively successful induction of lactation in sterile cows by injecting oestrogen plus progesterone for only 7 days. As with previous methods, however, the success rate remained relatively low (60–70%) and milk production was below normal.

More recently Fulkerson and McDowell (1974) found that normal milk yields could be obtained in all maiden ewes following a series of injections of oestrogen plus progesterone then a synthetic glucocorticoid. A preliminary experiment in maiden heifers, using a similar treatment adjusted for differences in body weight, also gave promising results (Fulkerson and McDowell 1975).

There is now a need to compare these methods for induction of lactation in an experiment incorporating normal animals, lactating after pregnancy, as controls.
Materials and Methods

Animals
The 24 Friesian and 5 Jersey heifers used in the experiment were grazed on irrigated pasture supplemented with good quality hay. Their ages varied from 22 to 32 months at the commencement of milking and their body weight was approximately 350 kg. The heifers were allocated by restricted randomization to four groups of six and one group of five using age, breed and live weight as the deciding criteria.

Hormones
β-Oestradiol-3-benzoate and β-oestradiol (Sigma Chemical Co., St Louis, U.S.A.) and progesterone (Diosynth, OSS, Holland) were dissolved in ethanol and administered either as such or suspended in peanut oil. Dexamethasone (Dexafort) (3 mg/ml) was obtained from Intervet Laboratories Ltd, Cambs., U.K. The schedule of hormone treatments is shown in Table 1.

Table 1. Schedule of hormone treatments for groups 1–5
All injections were administered subcutaneously; those for group 1 were in 2 ml ethanol and those for groups 2–4 were in 8 ml peanut oil. E2, β-Oestradiol; OB, β-oestradiol-3-benzoate; P, progesterone

<table>
<thead>
<tr>
<th>Group No.</th>
<th>No. of injections</th>
<th>Time of day (h)</th>
<th>E2 (mg)</th>
<th>OB (mg)</th>
<th>P (mg)</th>
<th>Ratio (E2 or OB)/P</th>
<th>Dexamethasone A</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>14 (twice daily)</td>
<td>0800, 1600</td>
<td>35</td>
<td>—</td>
<td>87·5</td>
<td>1/2·5</td>
<td>—</td>
</tr>
<tr>
<td>2</td>
<td>14 (every 3 days)</td>
<td>1100</td>
<td>—</td>
<td>10</td>
<td>100</td>
<td>1/10</td>
<td>+</td>
</tr>
<tr>
<td>3</td>
<td>14 (every 3 days)</td>
<td>1100</td>
<td>—</td>
<td>2·4</td>
<td>600</td>
<td>1/250</td>
<td>+</td>
</tr>
<tr>
<td>4</td>
<td>14 (every 3 days)</td>
<td>1100</td>
<td>—</td>
<td>2·4</td>
<td>600</td>
<td>1/250</td>
<td>+</td>
</tr>
<tr>
<td>5</td>
<td>Controls</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

A Dexamethasone injections, 20 mg, were given 3 and 4 days after the last injection of oestrogen and progesterone.

Experimental Procedure
The induced heifers (groups 1–4) received their last injection of hormones on the same day and the following day was taken as day 1 of the experiment.
Heifers in groups 1, 2 and 3 were machine milked once daily from day 4 to day 7 at 1600 h and then twice daily. Heifers in group 4 were suckled by an average of 1½ young calves per heifer at 1500 h and machine milked at 0600 h from day 4 to day 28, then machine milked twice daily. Milk yields from heifers in groups 1–4 were recorded on days 4, 7, 14, 21 and 28 and from heifers in group 5 at 7, 14, 21 and 28 days after calving. Thereafter all groups were recorded monthly for a further 9 months. To obtain total daily milk production of heifers in group 4, milk consumption of calves was found by weighing them before and after suckling.

Chemical Analyses
A composite sample of mammary secretion was taken from heifers in all groups on days 7, 14, 21 and 28 and monthly thereafter.
Samples of mammary secretion were analysed for protein, lactose and fat using the infrared milk analyser (Biggs 1972). The freezing point depression and somatic cell numbers of milk were determined on samples taken 33 days after the commencement of milking.

Statistical Analysis
Treatment differences between groups in yield and content of fat, protein and lactose of milk were determined by separate analyses of variance for each sampling date indicated and for overall 300-day yields.

Mating
All heifers were artificially inseminated as they came into oestrus after 3 months of milking. If they had not conceived after three inseminations they were mated to a Hereford bull.
Results

Milk Production

Although milk production of heifers in group 1 was significantly \( (P < 0.05) \) less than production from the other induced heifers initially, by day 21 there was no difference between the milk yields of induced heifers and this relationship persisted throughout lactation (see Fig. 1). Average total milk production for induced heifers was 60–70% of that for heifers in group 5 (normal) and this difference was statistically significant \( (P < 0.001) \).

![Fig. 1. Daily yield of mammary secretion for the five groups of heifers commencing on the first day of milking. Plotted points represent mean values for six heifers in groups 1, 2, 4 and 5 and five heifers in group 3. Standard errors are shown as vertical bars. ⚫ Group 1. ● Group 2. ▲ Group 3. ■ Group 4. △ Group 5.](image)

Milk Composition

The fat content of secretion was significantly \( (P < 0.05) \) higher for heifers in groups 2–4 than for heifers in group 1 and 5 on days 7 and 14 (see Table 2). Total protein was also significantly \( (P < 0.05) \) higher in the induced heifers than in heifers of group 5 (normal) from day 7 to day 90. In contrast, lactose content of secretion from induced heifers was significantly \( (P < 0.05) \) lower than in group 5 heifers on day 4. After this initial difference milk composition in all groups was similar.

Live Weight

There was no significant difference in live weight between the groups at the time heifers in group 5 were mated or at the end of lactation.

Reproductive Performance

The reproductive performance of heifers, as assessed by the number of inseminations to conception (1·3) and the number of animals failing to conceive after three artificial inseminations and three natural matings (30%), was similar in all groups. Palpation of the uterus and cervix at the time of mating indicated that these organs had developed to a stage similar to that of parous animals.

Animal Behaviour

Three heifers out of six in group 4 were observed to develop a maternal association towards the calves after 3–4 days of contact.
Table 2. Concentrations (\% w/v) of fat, protein and lactose in mammary secretion for the five groups of heifers  
Values given are means ± s.e. for six heifers in all groups except group 3 which contained five animals. Mean values for groups 2-4 do not differ significantly so have been grouped together. —, No secretion; n.a., not available.

<table>
<thead>
<tr>
<th>Group No.</th>
<th>Milk component (%)</th>
<th>Days after commencement of milking</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td>4</td>
</tr>
<tr>
<td>1 Fat</td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td></td>
<td>—</td>
</tr>
<tr>
<td>2-4</td>
<td></td>
<td>2.9</td>
</tr>
<tr>
<td>5</td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>n.a.</td>
<td>3.7</td>
</tr>
<tr>
<td>1 Protein</td>
<td></td>
<td>—</td>
</tr>
<tr>
<td>2-4</td>
<td></td>
<td>8.1</td>
</tr>
<tr>
<td>5</td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>n.a.</td>
<td>3.7</td>
</tr>
<tr>
<td>1 Lactose</td>
<td></td>
<td>—</td>
</tr>
<tr>
<td>2-4</td>
<td></td>
<td>4.0</td>
</tr>
<tr>
<td>5</td>
<td>n.a.</td>
<td>5.0</td>
</tr>
</tbody>
</table>

^ Mean for groups 3 and 4 only.

A
There was a marked tendency for some induced heifers to mount other animals. This behaviour began after hormone injections had ceased and lasted for 5–6 weeks. It was most pronounced in heifers in group 1, was less evident in group 2 heifers and absent in heifers of groups 3, 4 and 5. One heifer in group 1 became crippled after treatment but managed to lactate for 300 days.

Discussion

The results of the present experiment are surprising in two respects. Firstly, lactation using any one of four different methods has led to a similar level of milk production and, secondly, 100% of treated animals gave substantial amounts of milk secretion (>5·6 kg/day, peak levels). In contrast, previous workers (Hammond and Day 1944; Spriggs 1945; Smith and Schanbacher 1973) have found that 50–70% of treated animals 'lactated'. However, invariably, the animals were infertile and in fact when 'physiological normal' heifers (Williams and Turner 1962; Smith and Schanbacher 1974; Fulkerson and McDowell 1975) or ewes (Fulkerson and McDowell 1974) were used, all animals responded to treatment.

Total milk production of the induced heifers was about 65% of that of heifers lactating after a normal pregnancy. Production from induced and normal heifers began to decline 3–4 months after the commencement of milking. This lactational pattern appears to be normal and is similar to that observed in heifers induced to lactate by a variety of other methods (Folley and Malpress 1944; Smith and Schanbacher 1973).

The delayed milk production of heifers in group 1 is similar to that reported by Smith and Schanbacher (1974), using the same method. In fact these workers did not commence milking until 14 days after the cessation of hormone injections and this seems a worthwhile modification in a commercial situation.

In contrast to some earlier work (Reece 1943), the removal of milk by calves rather than the milking machine, in the immediate post-hormone period, did not enhance total milk yield in the present experiment. It was hoped that the suckling stimulus would elicit release of endogenous prolactin (Karg and Schams 1974) and/or glucocorticoids (Smith et al. 1972), both considered to be vital components of the hormone complex controlling the initiation of lactation (Fulkerson et al. 1976). Therefore, either the release of these hormones was similar during suckling and milking as has been confirmed in recent work (Fulkerson et al. 1978), or the synthetic glucocorticoid injected just prior to commencement of milking was adequate.

As far as could be ascertained, the composition of milk from induced heifers was normal by day 14 although the level of fat and protein in milk was slightly higher in secretion from induced animals.

One adverse effect of using relatively high doses of oestrogens, as in group 1 or group 2, is prolonged behavioural oestrus after cessation of treatment. Excessive oestrous activity for up to 50 days has also been reported by others (Paape et al. 1973; Narendran and Hacker 1974) and appeared to be due to the formation of a cystic ovary (Erb et al. 1973). This abnormal behaviour in early lactation appeared to have no ill effects on subsequent conception rate which was considered to be normal. In fact, a large proportion of chronically infertile animals have conceived following treatment for induction of lactation (Ferreira et al. 1974; Collier et al. 1975; Fulkerson and McDowell 1975; Fulkerson, unpublished data) and therefore this
treatment offers, as an additional benefit, a method of overcoming specific forms of infertility.

The present experiment provides a much needed comparison of the latest techniques for artificial induction of lactation. In physiologically normal animals a 100% success rate may be expected. However, in animals considered to be infertile, the response to treatment is markedly lower. As the technique will find most application with "infertile" animals, there is clearly a need to be able to select the animals which will respond to treatment. This will result in lowered treatment costs per animal thus making the technique commercially more attractive.

Acknowledgments

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References


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