

# Oestrus During Pregnancy in the Cow

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*Veterinary Record (1989) 124:387-390*

Forty-three oestruses were observed in 35 pregnant cows in one beef suckler herd and 17 in a dairy herd; at each oestrus the pregnant cow stood willingly to be mounted by another cow or bull. Such oestruses were observed at all stages of pregnancy, although more often between 121 and 240 days, occurred more than once per pregnancy and were also seen in successive pregnancies. On one farm where the dairy cows were observed for four 30 minute periods daily, oestrus was recorded in 5.7 percent of pregnant cows. The behavioral signs associated with oestrus during pregnancy were indistinguishable from those of true oestrus in non-pregnant animals and although its duration was shorter (mean 5.6 hours), its intensity was comparable to that of the true oestrus. Pregnant cows showing oestrus were usually seen interacting with other oestrus cows in the sexually active group. Pregnant cows showing oestrus had a higher mean condition score ( $3.9 \pm 0.64$ ) than control pregnant cows ( $3.0 \pm 0.36$ ). Physiological changes in the genital tract normally associated with true oestrus were not observed in pregnant cows showing oestrus. There was no ovulation or metoestrous bleeding. The characteristics of cervical mucus, including ferning patterns, were similar to those of pregnant cows at the same stage of pregnancy. Hormonal changes associated with oestrus in non-pregnant cows were not observed in the pregnant cows exhibiting oestrus. Seven of nine pregnant cows at oestrus stood willingly to be mounted by a bull. On seven occasions, bulls exposed only to cervical mucus from pregnant cows showing oestrus did not display flehmen. The ferning test, and other characteristics of cervical mucus, and an on-farm milk progesterone assay were used to confirm oestrus in pregnancy.

Oestrus is that restricted period of sexual receptivity, characterized by intense sexual desire, when the female will seek the male and even have periods of homosexual activity in which a cow mimics a bull. True oestrus begins when the female assumes the mating stance so that the male may mount and copulate. It does not normally occur during pregnancy in farm animals, although it is known to occur sporadically in cattle.

The incidence of oestrus during pregnancy has been

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reported to lie between 1 percent and 10 percent.<sup>3,4,5,19,16</sup> M.J. Ducker (personal communication) in a study of progesterone concentrations at the time of insemination, showed that in a herd with 20 percent mistimed inseminations, 1 percent to 2 percent occurred during pregnancy, but that in a herd with 40 percent mistimed inseminations, over 20 percent occurred during pregnancy.

Oestrus during pregnancy has several important practical implications for herd management: in the absence of accurate pregnancy diagnosis, animals which show oestrus may be thought to be repeat breeders and sold as infertile. Alternatively, the insemination of already pregnant cows may result in the loss of the embryo or fetus, and the consequent lengthening of the calving interval.<sup>7</sup>

The mechanism responsible for the exhibition of oestrus during pregnancy is not known. In the oestrus cycle in non-pregnant cows, hormonal changes are responsible for the manifestation of oestrus behavior, but no information is available on the hormonal status of the pregnant cows showing oestrus. A method for the diagnosis of oestrus during pregnancy is desirable; the present study was designed to examine the potential use of the characteristics of the cervical mucus, in particular the ferning pattern, and the concentration of progesterone in milk. The characteristics of the behavior of cows showing oestrus during pregnancy, especially their interactions with other cows and the bull, were also studied.

## Materials and Methods

A circular was sent to clients expressing the present authors' interest in the phenomenon of oestrus in pregnancy. On being notified of such animals, the farm was visited immediately, and again 14 days later. Pregnant cows showing various signs of oestrus, such as mounting, but not actually standing, were ignored. Only pregnant cows standing to be mounted at least twice on the first visit were investigated further. Details of another cow in the same herd and at a similar stage of pregnancy, but not showing oestrus, were also recorded for control data.

The following information was obtained about each cow: breed, condition score,<sup>12</sup> housed or at grass, presence of skin irritation, parity, stage of lactation, dates of oestrus during pregnancy, subsequent calving date, sex of calf,

signs of oestrus observed, intensity and duration of oestrus, other cows involved, reaction of bull to cow and to cervical mucus, presence of ovarian follicles and corpora lutea, uterine tone, vaginal and vulval oedema, and patency of the cervix.

Samples of cervical mucus were obtained with the aid of a vaginal speculum and a 45 cm x 0.5 cm plastic tube, the tip of which was inserted 1 to 2 cm into the external cervical os. An aspirated sample was spread on a clean, dry glass slide, allowed to dry and examined with a lower power microscope for ferning patterns.

Heparinised peripheral blood samples were taken from the tail by venepuncture, centrifuged at 1000 g for 10 minutes and the plasma stored at -20°C. Oestradiol and progesterone concentrations in plasma were measured by the verified radioimmunoassays of Dobson and Dean (1974)<sup>2</sup> and Kanchev and others (1976)<sup>10</sup> respectively; the interassay coefficients of variation were 13 and 12 percent respectively over the range of concentrations found in the present study.

Milk progesterone concentrations were assayed immediately on-farm, using an ELISA kit (Ovucheck; Cambridge Life Science).

The condition scores, plasma oestradiol and progesterone concentrations of the cows displaying oestrus in pregnancy were compared with the values obtained for the control cows, using Student's paired *t* test.

## Results

Forty-three incidences of true oestrus during pregnancy were investigated in 35 animals from 18 herds (17 dairy and one beef suckler) over a two-year period. Thirty-eight occurred in Friesian cows, four in Jersey cows and one in a Hereford cow. The dairy cows reflected the percentage of different dairy breeds in the practice, but this was not so for the beef animals. An incidence of 5.7 percent was recorded in one herd observed for four 30 minute periods each day.

The pregnant cows showing oestrus had significantly greater condition scores than the controls (Table 1). Oestrus was observed in 16 cows at grass and 27 cows which were housed for the winter. Sixty percent of the cows stood to be mounted in the loafing area near the cubicle shed, 31 percent were seen in the field and collecting yard, 10 percent in the collecting yard, and only 9 percent in the cubicle shed. The times of occurrence of oestrus reflected the seasonal calving patterns and sizes of the herds in the practice.

Pregnant cows showed oestrus during the first to the ninth lactations but 56 percent of the cases occurred during the fourth and fifth lactations. A peak incidence was recorded between 121 and 240 days of gestation (Fig. 1) but 18 percent occurred in the dry period and 7 percent

in nulliparous heifers. Five cows displayed oestrus in two successive pregnancies; one of them had been reported to show numerous periods of oestrus during all its pregnancies. No difference was observed between the sex of the calves born to the cows which showed oestrus, and the control cows.

TABLE 1: Comparison between the condition score of pregnant cows in oestrus and control cows at the same stage of pregnancy which were not in oestrus

Number of cows	Condition score							
	2	2½	3	3½	4	4½	5	
Pregnant cows in oestrus	-	2	6	8	18	5	4	
Pregnant cows not in oestrus	1	8	26	6	2	-	-	

Mean (± sd) score of cows in oestrus : 3.9 ± 0.64  
 Mean (± sd) score of cows not in oestrus : 3.0 ± 0.36, P < 0.001

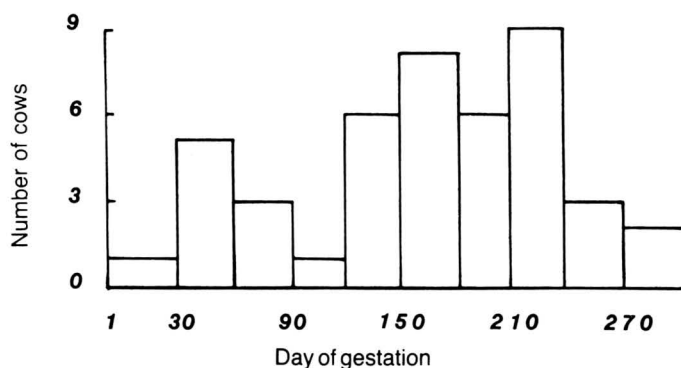


FIG 1: The occurrence of oestrus at different times during pregnancy

## Behavior

As can be seen in Table 2, mounting other cows, restlessness and increased walking were observed very frequently at oestrus during pregnancy. The mean ± sd duration of oestrus was 5.7 ± 2.5 hours (range one to 12 hours) with 70 percent lasting between four and eight hours. When the intensity of oestrus was estimated as weak, normal or strong, 60.5 percent of the oestrus were classed as normal. On 77 percent of the occasions when pregnant cows were in oestrus, one or more non-pregnant cows were simultaneously in oestrus, with two to five other cows in the interacting group. Of cows in this group, 46 percent were in true oestrus and 15 percent were in pro-oestrus. However, 34 percent were also in the last three months of gestation. On nine occasions it was possible to expose a bull to a pregnant cow in oestrus, and at the same time to a control cow. Seven pregnant cows in oestrus were picked out by the bull. After sniffing the genital region, and displaying flehmen in five cases, together with chin resting, the bull mounted and served seven cows, which willingly stood. There was no sniffing, flehmen display, chin resting or mounting of the control pregnant cows.

**TABLE 2: Incidence of signs of oestrus in pregnant cows in oestrus**

Signs of oestrus	Number of oestruses	Percentage
<b>Behavioural signs</b>		
Standing to be mounted	43	100
Mounting other cows	37	86
Restlessness and increased walking	32	74
Sniffing other cows	19	44
Bellowing	6	14
Head-to-head mounting	1	2
<b>Circumstantial signs</b>		
Ruffled hair over rump	26	60
Muddy back and sides	25	58
Steaming back	14	33
Raw tail head	10	23

### Anatomy

Erythema, or oedema of the vulva was not observed in either the pregnant cows in oestrus or in the control cows. However, vulval oedema and relaxation was increasingly noticed in both types of cows in the last month of pregnancy, as calving approached. The mucosa of the vagina and cervix was pale in the pregnant cows in oestrus and in the control cows. The cervix was closed, although from 220 days of pregnancy onwards one finger could be inserted in the external os of seven cows and their controls. None of the cows investigated had any significant uterine tone.

Both ovaries were palpated per rectum between 22 and 145 days of pregnancy, after which it became too difficult. Follicles ranging in size from approximately 1 cm to 1.5 cm were felt on both ovaries of five of 12 pregnant cows in oestrus and in three of 12 control cows. The presence of the follicles was confirmed by ultrasonography; they were soft fluctuant structures protruding from the surface of the ovary, and yielded to pressure. Palpation two days later failed to detect evidence of ovulation, and by 14 days later secondary corpora lutea were not palpable. Metoestrous bleeding was not observed.

### Mucus

In general, the cervical mucus was cloudy or creamy, sparse and jelly-like, with little elasticity (Table 3). However, during the last week or so of gestation, the characteristics of the mucus changed; it became clean, increased in quantity and became more fluid and elastic. Similar observations were made in the control cows. Typical ferning patterns, i.e., regular fine patterns fully developed throughout the smear, were not seen in the mucus from any pregnant control cows. However, samples collected from 35 non-pregnant cattle in true oestrus all showed typical ferning. Atypical ferning (with coarse patterns in widely distributed pockets which lacked the elegance and fine architecture of typical ferning) was seen in 20 pregnant cows on the day of oestrus, and in 15 control cows on the same day. Fourteen days later, 13 of

the pregnant cows in oestrus and 11 control cows showed atypical ferning. Absence of ferning (with a high cell content) was first recorded in 23 pregnant cows in oestrus and in 28 control cows; 14 days later the position was almost the same. There was an increase in the proportion of the ferning in the atypical groups in cows near parturition. The ferning was coarse and did not present a diagnostic problem, because it was obvious that calving was imminent.

**TABLE 3: Number of pregnant cows in oestrus and control pregnant cows not in oestrus with cows cervical mucus showing different characteristics**

Characteristics of cervical mucus	On the day of oestrus	Control cow	14 days after oestrus*	Control cow 14 days later*
<b>Colour</b>				
Creamy	9	12	11	14
Cloudy	30	26	26	25
Clear	4	5	4	2
<b>Quantity</b>				
Little	39	38	37	36
Some	4	5	4	5
Large amounts	—	—	—	—
<b>Consistency</b>				
Thick, jelly-like	41	41	41	41
Fluid	2	2	—	—
Watery	—	—	—	—
<b>Elasticity</b>				
Little	39	38	35	37
Some	4	5	6	4
Very elastic	—	—	—	—

\* Two cows were not examined as they had already calved

A bull was present on five farms and on nine occasions, when a sample of cervical mucus from either a pregnant cow in oestrus or a control cow was presented on a plastic glove to the bull, interest and licking of the samples was observed on samples from three of the pregnant cows in oestrus. There was no display of flehmen or general arousal of the bull.

### Hormones

The concentrations of oestradiol and progesterone in paired plasma samples from pregnant cows in oestrus are shown in Figs. 2 and 3. The results from control cows were comparable and there were no significant differences between the samples taken on the day of oestrus and those taken 14 days later, nor between oestrus and control cows on either day.

All lactating pregnant cows, whether in oestrus or not, had high progesterone concentrations, demonstrated with the on-farm milk progesterone kit, in contrast to the non-pregnant cows in true oestrus, which had low concentrations of progesterone in milk.

### Discussion

Forty-three pregnant cows expressing oestrus, and their paired controls, have been thoroughly investigated.

The incidence of 5.7 percent in one herd was in agreement with Erb and Morrison (1958)<sup>5</sup> and Perez-Garcia and others (1984).<sup>16</sup> The pregnant cows in oestrus had

while they were housed for the winter. The duration of standing oestrus in pregnant cows was shorter (5.6 hours) than in non-pregnant cows in true oestrus (9.2 to 14.9 hours)<sup>13,6</sup> and it was therefore more likely to be noticed when they were housed and under closer observation. However, in a study of the factors determining the expression of oestrus, Britt and others (1986)<sup>1</sup>, found that more cows were willing to stand or mount on dirt than on concrete. Most occasions of oestrus in the present study were observed in the loafing area near the cubicle shed. However, a large percentage was observed in the field, an observation similar to that of Pennington and others (1985)<sup>15</sup> who found that sexual activity usually occurred in the least crowded areas with the best footing.

Oestrus during pregnancy occurred in all parities but was more frequent in the fourth and fifth lactations. Dominant cows were more likely to be involved in sexually active groups, and the degree of dominance was increased by age, weight and time in the herd. Donald (1943)<sup>3</sup> and Erb and Morrison (1958)<sup>5</sup> recorded the highest incidence of oestrus during the first 90 days of pregnancy whereas Mohan and Purbey (1981)<sup>11</sup> and Patti and others (1982)<sup>14</sup> reported an average of 120 days. The present findings concur with those of Perez-Garcia and others (1984)<sup>16</sup> and Britt and others (1986)<sup>1</sup> who found the highest incidence between 120 and 240 days. Most pregnant cows showed oestrus only once, and rarely in successive pregnancies.

In agreement with Donald (1943)<sup>3</sup> there was no association between the sex of the fetus and the occurrence of oestrus during pregnancy; however, Erb and Morrison (1958)<sup>5</sup> found a significantly higher percentage of male calves in pregnant cows which had shown oestrus. Robertson and King (1979)<sup>18</sup> were unable to detect an influence of fetal sex on the oestrogen concentrations in fetal fluids or maternal blood.

Pregnant cows showed the whole spectrum of oestrus behavior shown by non-pregnant cows in oestrous.<sup>9</sup> Cows in the first 150 days of pregnancy did very little mounting, but from 180 days onwards they became increasingly involved in a sexually active group. Considerable follicular activity was observed on the ovaries of both pregnant cows in oestrus and control cows, especially in early pregnancy. However, there was little evidence of any oestrogenic influence on the vagina and cervix, except at the end of pregnancy. No extra ovulations, and no secondary corpora lutea, were recorded.

The cervical mucus in most cases was gelatinous, adhesive and tenacious, and no different from the mucus of control pregnant cows. Ferning patterns in all the pregnant cows were either atypical or absent as previously observed by Ghannam and Sorensen (1967).<sup>8</sup> The ferning test was very accurate in showing whether or not oestrus behavior in a supposedly pregnant cow indicated true oestrus. However, because the technique required the animal to be restrained and strict attention to hygiene was

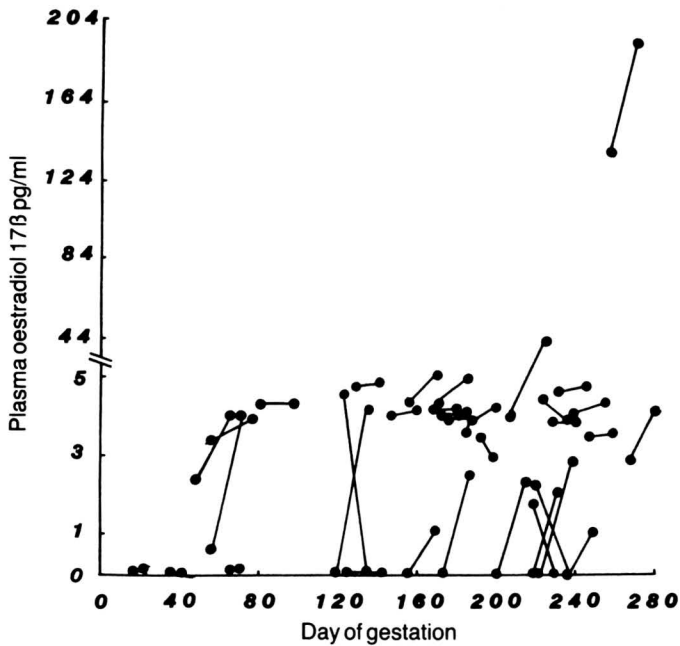


Fig 2

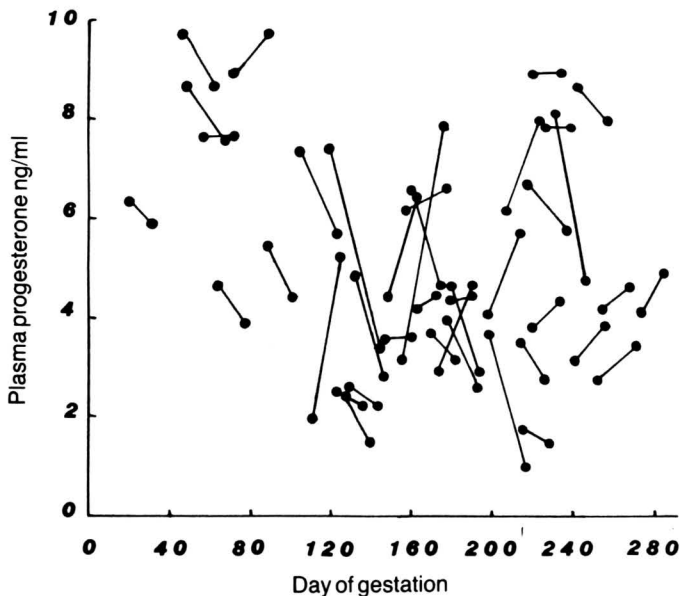


FIG 3: Plasma concentrations of progesterone in pregnant cows in oestrus. Each cow was sampled twice, 14 days apart; the paired results are at the end of each line

significantly higher condition scores than the control cows; non-pregnant cows which are in good condition similarly show more overt signs of oestrus than cows in poor condition.

Most of the pregnant cows were observed in oestrus

necessary, it was considered to be invasive.

When the bull was running with a herd it was not seen to accompany the pregnant cows for a few days before oestrus was observed, unlike his behavior towards the non-pregnant cows in oestrus. However, seven pregnant cows did stand to be mounted and were served by the bull. Subsequent abortions did not occur in this study, and were not reported by Perez-Garcia and others (1984).<sup>16</sup> The bulls did not display flehmen when they were exposed to a plastic glove smeared with cervical mucus taken from a pregnant cow in oestrus, suggesting that insufficient pheromones were present. As the pregnant cows in oestrus were usually in a sexually active group, it is presumed that the attention of the bull was mediated visually.

There was no evidence of increased oestradiol concentrations in the plasma of pregnant cows in oestrus, and high progesterone concentrations were maintained. The on-farm milk progesterone ELISA test was useful, reliable and non-invasive. The present results do not accord with the suggestion of Peters and Ball (1987)<sup>17</sup> that follicles in pregnancy produce sufficient oestradiol to cause oestrus. Circumstantial evidence has been provided to indicate that such follicles do not reach maturity in terms of oestradiol production and ovulation.

It remains a mystery why oestrous behavior occurs during pregnancy.

#### Acknowledgements

This work was carried out by I. Thomas in partial fulfillment of the requirements for the University of Liverpool Diploma in Bovine Reproduction. Gratitude is

expressed to the Royal College of Veterinary Surgeons Leverhulme Trust fund, for financial assistance; to all the farms involved for allowing access to their cows, and to J. Owen and H. Pursell for blood progesterone and oestradiol measurements.

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