

Late-night suckling inhibits onset of postpartum oestrous activity in beef cows

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To determine whether suckling of calves late at night would prolong lactation anoestrus, 51 Hereford-type cows (21 – 29 days postpartum) were divided into three treatment groups. In treatments 1 (night suckling) and 2 (day suckling) cows suckled their calves five times every 24 hours; the calves in treatment 1 did not suckle between 09:00 and 17:00 and those in treatment 2 between 21:00 and 05:00. Cows in treatment 3 suckled only once daily at 17:00. By 80 days after calving a significantly greater proportion ($P < 0.001$) of the night-suckled cows had not returned to oestrus than those from the other treatments. The mean interval to first oestrus was significantly ($P < 0.001$) longer in treatment 1 (67.0 ± 1.5 days) than in treatments 2 (42.2 ± 2.5) and 3 (45.7 ± 3.2). Lactation anoestrus does not appear to be dependent on the number of sucklings per 24 hours, but rather on the time of night when suckling occurs.

Om te bepaal of suiping van kalwers laat in die nag, laktasie anestrus sal verleng, is 51 Hereford-tipe koeie, 21 – 29 dae na kalwing, in drie groepe verdeel. In behandelings 1 en 2 het die koeie hul kalwers vyf keer elke 24 uur laat suip. Die kalwers is in behandeling 1 (nag suiping) nie tussen 09:00 en 17:00, en dié in behandeling 2 (dag suiping) nie tussen 21:00 en 05:00 toegelaat om te suip nie. Die koeie in behandeling 3 het net eenkeer per dag om 17:00 laat suip. 'n Betekenisvolle groter verhouding ($P < 0.001$) van die koeie wat gedurende die nag laat suip het (behandeling 1), het nie estrus teen 80 dae na kalwing, as by die ander behandelings groepe getoon nie. Die gemiddelde interval tot eerste estrus was betekenisvol ($P < 0.001$) langer by behandeling 1 (67.0 ± 1.5 dae) as by behandelings 2 (42.2 ± 2.5) en 3 (45.7 ± 3.2). Klaarblyklik is laktasie anestrus van die tyd van die nag wanner suiping plaasvind afhanklik en nie van die aantal suipings per 24 uur nie.

Keywords: Beef cow, night suckling, postpartum anoestrus.

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Introduction

When Williams (1990) reviewed suckling-induced anoestrus in cattle, he stated that either the frequency, intensity or duration of suckling had been considered to be the major controlling factor. Although species differences are undoubtedly important, a similar conclusion has been reached for nursing women (McNeilly *et al.*, 1985), with the evidence favouring frequency as opposed to duration (Robyn *et al.*, 1985). To date, no relationship between suckling frequency and onset of oestrus, postpartum, has been detected in bovines (Williams *et al.*, 1984; Day *et al.*, 1987). However, it appeared that two or three sucklings per day would delay breeding after parturition in cows (Williams, 1990), while in women, ovarian activity was suppressed by suckling frequencies exceeding five times, and more than 65 minutes in duration (McNeilly *et al.*, 1982; 1983). Thus, an early resumption of oestrous cycles, by limiting suckling to once daily, has been achieved in beef cows (Randel, 1981; Wells *et al.*, 1986; Browning *et al.*, 1993).

Studies have been undertaken (Short *et al.*, 1972; McVey & Williams, 1988; Viker *et al.*, 1989; 1993) in an attempt to elucidate which aspect of suckling behaviour is responsible for the inhibition of reproductive cycles. When attention was focused on the hours of darkness, it appeared that regardless of the natural suckling frequency, cows initiated breeding only if a consistently long interval (more than 7 hours)

between sucklings occurred (Stewart *et al.*, 1993a). Furthermore, it appeared that this long interval tended to occur late at night. Based on research with human females, indirect support for this hypothesis has been provided by Howie & McNeilly (1982), who proposed that the absence of breastfeeding between midnight and 08:00 might favour the recovery of the hypothalamic-pituitary-ovarian axis to permit ovulation. When cows were separated from their calves, either during the night or day, the former treatment resulted in more cows exhibiting oestrus during the 90-day breeding period (Stewart *et al.*, 1993b). This study was therefore conducted to establish whether cows that continued to suckle their calves late at night would exhibit a prolongation of postpartum anoestrus.

Materials and Methods

This study was conducted at the Cedara Research Centre (longitude $30^{\circ}17'$, latitude $29^{\circ}32'$). Between 21 and 29 days postpartum, spring-calving, Hereford-type cows (aged 3 – 7 years) and their calves were randomly allocated to 1 of 3 treatment groups (Table 1). Cows were allocated in batches (ranging in calving date by not more than 7 days) to the treatment groups. The first cow calved on 27 July and the last cow on 25 September. The cows in treatments 1 and 2 suckled their calves every 4 hours five times every day. When not suckling, the calves were kept separate from their dams by

Table 1 Time of the day when cows were allowed (+) or not (0) to suckle their calves

Treatment	Suckling times					
	09:00	13:00	17:00	21:00	01:00	05:00
1 - Night suckling	+	0	+	+	+	+
2 - Day suckling	+	+	+	+	0	+
3 - Once daily	0	0	+	0	0	0

confining them to a small pen within the cow-holding area (1600-m² feedlot). At the appointed suckling times the cows were allowed to enter the calf pen and the calves suckled for only 5 minutes whereafter the cows were again removed. Free contact between cows and calves was possible through the sides of the pen, except that suckling was prevented.

In treatment 1, the calves ($n = 15$) did not suckle for 8 hours from 09:00 to 17:00 (night suckling), while in treatment 2, this long non-suckling period was from 21:00 to 15:00 (day suckling; $n = 17$). On 25 August, when the first cows entered the experiment, sunrise and sunset were at 06:25 and 17:37, respectively. The experiment ended on 4 December, at which stage sunrise and sunset occurred at 04:56 and 18:43, respectively. Cows in treatment 3 suckled their calves for a total time (25 minutes) equivalent to that of treatments 1 and 2, but only at 17:00 each day (once daily suckling; $n = 18$). In this study emphasis was placed on the postpartum stage at which the cow could have become pregnant, i.e. the day on which serve first became possible. Accordingly, at each suckling time (Table 1), regardless of whether the cows had been suckled, observations for oestrus were conducted for 20 minutes after their separation from the calves. Oestrus was defined as standing while being mounted by other cows. Additional signs of behavioural oestrus, such as nasal exploration of the perineal area by other cows, not in oestrus, and of mucous discharge from the vagina, were also carefully noted. In order to minimize stress due to the experimental procedures, no rectal palpations to determine the presence of corpora lutea were conducted. Similarly, to avoid stimulating

reproductive activity, the examination of the ovarian status per rectum was excluded. However, in order to eliminate cows that may have ovulated prior to the commencement of treatments, jugular blood samples were collected into heparinized tubes for progesterone assay of the plasma, one day before the treatment was initiated. The method of Butcher *et al.* (1974) was used for progesterone assay.

The calves had access to drinking water and hay while the cows were fed maize silage. *Eragrostis curvula* hay and a mineral lick (equal parts salt, maize meal, urea and di-calcium phosphate) so as to maintain a condition score of 2.5 (0 = thin, 5 = fat). Cows that had not shown oestrus by 80 days after calving (once daily suckling, $n = 2$; night suckling, $n = 13$) were removed from the experiment to facilitate re-breeding for other experiments. First-calvers which could not compete effectively for feed and which thus lost condition severely, were also omitted from the analysis (treatment 2, $n = 3$).

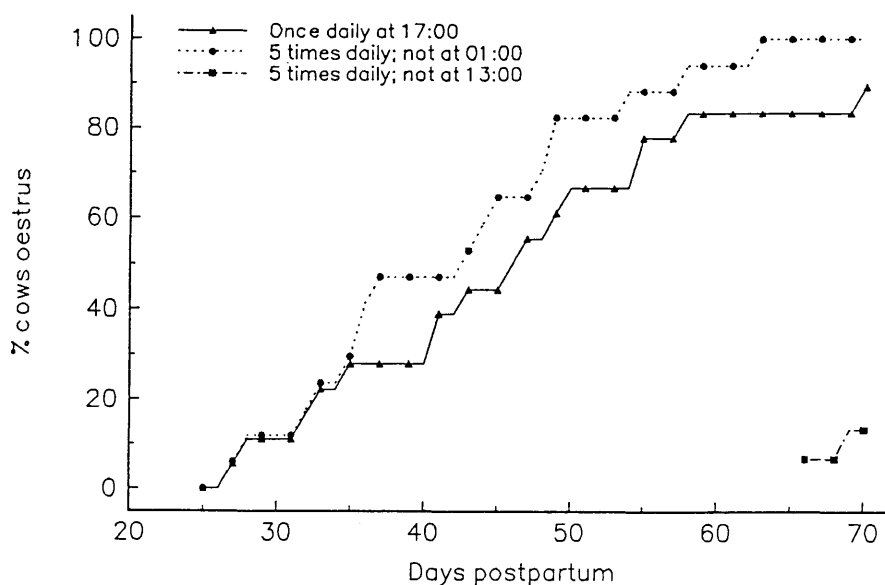
Statistical analysis

The proportions of cows which had not exhibited oestrus by 80 days postpartum were compared by the Chi-square test. The number of days to first oestrus for the three treatment groups was compared using the log rank test (Armitage & Berry, 1987) which is appropriate for survival data. The days for cows that had not shown oestrus by 80 days postpartum were treated as censored observations.

Results

Night suckling (treatment 1) increased ($P < 0.001$) the proportion (13/15) of cows that did not cycle (Figure 1) compared with cows suckled only once daily (treatment 3) or suckled during the day (treatment 2).

The difference in mean days to first oestrus between cows suckled once daily (treatment 3, $\bar{x} = 45.7 \pm 3.2$) and those that missed the suckling at 01:00 (treatment 2, $\bar{x} = 42.2 \pm 2.5$ days) was not statistically significant at the 5% level, although there was some evidence (Figure 1) that treatment 2

**Figure 1** The cumulative % of cows exhibiting first oestrus as influenced by frequency and time of suckling.

had a slightly shorter mean interval to first oestrus ($P < 0.07$). Cows that suckled at night, but not at 13:00 (treatment 1, $\bar{x} = 67.0 \pm 1.5$ days), showed oestrus later ($P < 0.0001$) than treatments 1 and 3 (Figure 1).

Discussion

Just as in the beef cow, nursing of the infant is believed to be the primary cause of lactational acyclicity in women (Short, 1984). As the infant grows older, the frequency and duration of nursing decrease and this facilitates the resumption of ovarian cycles in the lactating mother (Short, 1984). However, among !Kung women, where the period between births frequently exceeds 3 years (Howell, 1979), the interval between nursings was strongly correlated with the age of the infant, but not the duration of each nursing (Konner & Worthman, 1988). Similarly, no effect of age of the calf on duration of suckling bouts has been recorded (Drewry *et al.*, 1959; Odde *et al.*, 1985; Wells, 1986; Stewart *et al.*, 1993a).

Konner & Worthmann (1988) maintained that the critical change that occurred as the infant aged was a lengthening of the period between nursings. Obviously, the number of nursings per 24 hours must then decrease accordingly. A similar response has been seen in beef calves where the suckling frequency decreased with the age of the calf (Drewry *et al.*, 1959; Hutchinson *et al.*, 1962; Walker, 1962; Reinhardt & Reinhardt, 1981; Wells, 1986; Day *et al.*, 1987). In contrast, Odde *et al.* (1985) have reported no effect of calf age on suckling frequency.

The association between frequency of suckling and interval to first ovulation, postpartum, may not be as clear-cut as presumed. Thus, Stewart *et al.* (1993) observed that at about the time cows could be expected to end postpartum anoestrus, i.e. when their calves were about 45 days or older, the calves were less likely to suckle from 22:00 h to dawn than younger calves. Attention has been drawn to the important ovulation-delaying effect of night-time breast feeding (Howie & McNeilly, 1982; Short, 1984) and, in the discussion following Frisch's (1985) paper, McNeilly proposed that breast feeding before 23:00 was as effective in disrupting onset of normal ovarian activity as suckling between 23:00 and 06:00. McNeilly (pers. comm., 1994) now maintains that the time of day or night is unimportant.

The results obtained here clearly demonstrate (Figure 1) that night suckling prolonged lactation anoestrus. In contrast, when suckling was prevented between 21:00 and 05:00, this was as effective as once daily suckling in stimulating the onset of first oestrus. Thus, it would appear that there may be a time of the night when beef cows are particularly sensitive to the inhibitory influence of suckling. The possibility that the hours after onset of darkness or before first light may be of greater importance than the actual time needs to be considered.

The mechanism by which suckling delays the time of first ovulation after parturition has been the subject of speculation and research. It has been proposed that the release of prolactin in response to nipple stimulation (Short, 1984) may not be the cause of the hypothalamic inhibition observed. A more likely mechanism appears to be the suppressive effects of beta-endorphins on the release of gonadotrophin-releasing hormone (Short, 1984). Williams (1990) has expanded on this

thesis as it pertains to the beef cow and Viker *et al.* (1993) have shown that the teat may not be the only sensitive area of the udder. Recent results have shown the mother-offspring bond to be a vital component of the mechanism by which suckling delays ovulation (Silveira *et al.*, 1993) and somatosensory cues appear to play no role in suckling-induced anoestrus (Williams *et al.*, 1993).

A further possibility involves the mechanism by which melatonin mediates the effect of variations in day length on seasonal breeding (Yellon *et al.*, 1992). This hormone is released in a characteristic pattern during darkness in many mammalian species (Deveson *et al.*, 1992; Lincoln, 1992) and the period of exposure to elevated levels of melatonin may be important in modifying the inhibitory influence of night-time suckling. While seeking an explanation, it should not be forgotten that there is also a negative association between calving date and delay to first oestrus (Peters & Riley, 1982; Hauser, 1984). Whatever the final explanation, our results provide strong supportive evidence for Short's (1984) contention that the abandonment of night-time feeding might remove the suckling inhibition of GnRH release from the hypothalamus. This explanation is also supported by the finding that the return of ovarian function in breast-feeding women is associated with night-time pulsatile secretion of LH (Tay *et al.*, 1989). In order to obtain further support for the conclusions regarding late-night suckling, cows in treatment 1 that had reached 80 days postpartum without having exhibited oestrus were then prevented from suckling between 21:00 and 05:00, but continued to suckle their calves five times per 24 hours, as for treatment 2. Of the 7 cows treated in this way 57% were in oestrus within 18 days.

The failure to observe a positive association between milk yield and duration of postpartum anoestrus in beef cows (Day *et al.*, 1987; Beal *et al.*, 1990) also needs to be accounted for. This is especially important for selection in the beef industry where there has been much attention on improving weaning weights. The associated increased pre-weaning growth rates are at least partly dependent on higher milk production of the dams (Beal *et al.*, 1990), with attendant greater nutritional demands. If such demands are not satisfied, the cows could fail to re-breed in time. To date, experimental results have not supported such a trend (Deutscher & Whiteman, 1971; Holloway *et al.*, 1975; Montaña-Bermudez & Nielsen, 1990). At the other end of the scale, cows which fail to produce sufficient milk to satisfy the appetite of the calf are likely to be suckled frequently (Drewry *et al.*, 1959; Odde *et al.*, 1985). Consequently, the chances of suckling occurring during the 'critical' hours of darkness are increased. Such cows may thus be anoestrus for longer than cows producing more milk.

The above conclusions support the proposal (Short, 1984) that supplementation of energy to nursing mothers may decrease the frequency of suckling as a consequence of the improved milk yield. These additional calories might result in night-time nursing being curtailed. The lack of an association between milk yield and duration of lactation anoestrus in beef cows can then be explained in terms of suckling occurring outside the sensitive period of the night.

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References

- ARMITAGE, P. & BERRY, G., 1987. Statistical methods in medical research. Blackwell Scientific Publications, Oxford.
- BEAL, W.E., NOTTER, D.R. & AKERS, R.M., 1990. Techniques for estimation of milk yield in beef cows and relationships of milk yield to calf weight gain and postpartum reproduction. *J. Anim. Sci.* 68, 937.
- BROWNING, R., NEUENDORFF, D.A. & RANDEL, R.D., 1993. Effects of postpartum nutrient intake and once daily suckling on return to estrus and endocrine profiles in fall-calving Brahman cows. *J. Anim. Sci.* 71 (suppl. 1), p. 447 (Abstr).
- BUTCHER R.L., COLLINS, W. & FUGO, N., 1974. Plasma concentration of LH, FSH, prolactin, progesterone and estradiol-17 β throughout the 4-day estrous cycle of the rat. *Endocrinology*, 94, 1704.
- DAY, M.L., IMAKAWA, K., CLUTTER, A.C., WOLFE, P.L., ZAL-ESKY, D.D., NIELSEN, M.K. & KINDER, J.E., 1987. Suckling behavior of calves with dams varying in milk production. *J. Anim. Sci.* 65, 1207.
- DEUTSCHER, G.H. & WHITEMAN, J.V., 1971. Productivity as two-year-olds of Angus-Holstein crossbreds compared to Angus heifers under range conditions. *J. Anim. Sci.* 33, 337.
- DEVESON, S.L., ARENDT, J. & FORSYTH, I.A., 1992. The influence of the pineal gland and melatonin on the reproductive performance of domesticated ungulates. *Anim. Reprod. Sci.* 30, 113.
- DREWRY, K.J., BROWN, C.J. & HONEA, R.S., 1959. Relationships among factors associated with mothering ability in beef cattle. *J. Anim. Sci.* 18, 938.
- FRISCH, R.E., 1985. Maternal Nutrition and Lactational Amenorrhoea: Perceiving the Metabolic Costs. In: Maternal Nutrition and Lactational Infertility. Ed. Dobbing, J., Raven Press, New York.
- HAUSER, E.R., 1984. Seasonal effects on female reproduction in the bovine (*Bos taurus*) (European Breeds). *Theriogenology* 21, 150.
- HOLLOWAY, J.W., STEPHENS, D.F., WHITEMAN, J.V. & TOTUSEK, R., 1975. Performance of 3-year-old Hereford, Hereford \times Holstein and Holstein cows on range and in drylot. *J. Anim. Sci.* 40, 114.
- HOWELL, N., 1979. Demography of the Dobe !Kung. Academic Press, New York.
- HOWIE, P.W. & MCNEILLY, A.S., 1982. Effect of breast-feeding patterns on human birth intervals. *J. Reprod. Fert.* 65, 545.
- HUTCHINSON, H.G., WOOF, R., MABON, R.M., SALEHE, I. & ROBB, J.M., 1962. A study of the habits of zebu cattle in Tanganyika. *J. Agric. Sci.* 59, 301.
- KONNER, M. & WORTHMAN, C., 1988. Nursing frequency, Gonadal function, and birth spacing among !Kung hunter-gatherers. *Science* 207, 788.
- LINCOLN, G.A., 1992. Photoperiod-pineal-hypothalamic relay in sheep. *Anim. Reprod. Sci.* 28, 203.
- MCNEILLY, A.S., GLASIER, A. & HOWIE, P.W., 1985. Endocrine Control of Fertility. I. In: Maternal Nutrition and Lactational infertility. Ed. Dobbing, J., Raven Press, New York.
- MCNEILLY, A.S., HOWIE, P.W., HOUSTON, M.J., COOK, A. & BOYLE, H., 1982. Fertility after child-birth: adequacy of postpartum luteal phases. *Clin. Endocrinol.* 17, 609.
- MCNEILLY, A.S., GLASIER, A.F., HOWIE, P.W., HOUSTON, M.J., COOK, A. & BOYLE, H., 1983. Fertility after childbirth: pregnancy associated with breast feeding. *Clin. Endocrinol.* 19, 167.
- MONTAÑO-BERMUDEZ, M. & NIELSEN, M.K., 1990. Reproductive performance and variation in body weight during annual cycles for crossbred beef cows with different genetic potential for milk. *J. Anim. Sci.* 68, 2289.
- McVEY, W.R. & WILLIAMS, G.L., 1988. Mammary somato sensory masking in suckled anestrous cows: Endocrine, reproductive and lactational characteristics. *Biol. Reprod.* 38 (Suppl. 1), 109(Abstr).
- ODDE, K.G., KIRACOFE, G.H. & SCHALLES, R.R., 1985. Suckling behavior in range beef calves. *J. Anim. Sci.* 61, 307.
- PETERS, A.R. & RILEY, G.M., 1982. Is the cow a seasonal breeder? *Br. Vet. J.* 138, 535.
- RANDEL, R.D., 1981. Effect of once-daily suckling on postpartum interval and cow-calf performance of first-calf Brahman \times Hereford heifers. *J. Anim. Sci.* 53, 755.
- REINHARDT, V. & REINHARDT, A., 1981. Natural suckling performance and age of weaning in zebu cattle (*Bos indicus*). *J. Agric. Sci.* 96, 309.
- ROBYN, C., MEURIS, S. & HENNART, P., 1985. Endocrine Control of Lactational Infertility. In: Maternal Nutrition and Infertility. Ed. Dobbing, J., Raven Press, New York.
- SHORT, R.V., 1984. Breast feeding. *Sci. Am.* 250, 23.
- SHORT, R.E., BELLOWS, R.A., MOODY, E.L. & HOWLAND, B.E., 1972. Effects of suckling and mastectomy on bovine postpartum reproduction. *J. Anim. Sci.* 34, 70.
- SILVEIRA, P.A., SPOON, R.A., RYAN, D.P., WILLIAMS, G.L., 1993. Evidence for maternal behaviour as a requisite link in suckling-mediated anovulation in cows. *Biol. Reprod.* 49, 1338.
- STEWART, I.B., LISHMAN, A.W. & LOUW, B.P., 1993a. Suckling behaviour and fertility in beef cows on pasture. 1. Suckling behaviour. *S. Afr. J. Anim. Sci.* 23, 176.
- STEWART, I.B., LOUW, B.P. & LISHMAN, A.W., 1993b. Suckling behaviour and fertility in beef cows on pasture. 2. The influence of twelve-hour calf separation on interval to first oestrus after onset of mating period. *S. Afr. J. Anim. Sci.* 23, 180.
- TAY, C.C.K., GLASIER, A. & MCNEILLY, A.S., 1989. Twenty-four hour secretory profiles of gonadotrophins and prolactin in breast-feeding women. *J. Reprod. Fert. Abstr. Series* No.3, p. 10 (Abstr).
- VIKER, S.D., McGUIRE, W.J., WRIGHT, J.M., BEEMAN, K.B. & KIRACOFE, G.H., 1989. Cow-calf association delays postpartum ovulation in mastectomised cows. *Theriogenology* 32, 467.
- VIKER, S.D., LARSON, R.L., KIRACOFE, G.H., STEWART, R.E. & STEVENSON, J.S., 1993. Prolonged postpartum anovulation in mastectomised cows requires tactile stimulation by the calf. *J. Anim. Sci.* 71, 999.
- WALKER, D.E., 1962. Suckling and grazing behavior of beef heifers and calves. *N.Z. J. Agric. Res.* 5, 331.
- WELLS, P.L., 1986. The influence of suckling during early lactation on the resumption of ovarian activity and oestrous behaviour in the Afrikaner cow. PhD. thesis, University of Natal, Pietermaritzburg.
- WELLS, P.L., HOLNESS, D.H., McCABE, C.T. & LISHMAN, A.W., 1986. Fertility in the Afrikaner cow. 3. Once-a-day suckling and its effect on the pattern of resumption of ovarian activity and conception rate in early lactation. *Anim. Reprod. Sci.* 12, 1.
- WILLIAMS, G.L., 1990. Suckling as a regulator of postpartum rebreeding in cattle: A review. *J. Anim. Sci.* 68, 831.
- WILLIAMS, G.L., McVEY, W.R. & HUNTER, J.F., 1993. Mammary somatosensory pathways are not required for suckling-mediated inhibition of luteinizing hormone-secretion and delay of ovulation in cows. *Biol. Reprod.* 49, 1328.
- WILLIAMS, G.L., OSBORN, R.G., KIRSCH, J.D. & TILTON, J.E., 1984. Suckling, milking and calf presence as regulators of tonic gonadotropin release and postpartum interval. 10th Int. Congr. Anim. Reprod. Artif. Insem. Univ. Illinois, Urbana-Champaign, p. 410.
- YELLON, S.M., FOSTER, D.L., LONGO, L.D. & SUTTIE, J.M., 1992. Ontogeny of the pineal melatonin rhythm and implications for reproductive development in domestic ruminants. *Anim. Reprod. Sci.* 30, 91.