

## Effect of post-insemination clitoris massage on conception rate and duration of oestrus in crossbred cows with prolonged oestrus

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### ABSTRACT

The effect of post insemination clitoris massage (CM) on conception rate and duration of oestrus in crossbred dairy cattle was studied. Sixty inseminations each were done under category A (insemination immediately followed clitoris massage CM) and category B (without CM). Those continued to show heat signs on second day in both categories were re-inseminated as on the previous day, while AI was not repeated even though few animals continued to show heat signs on the third day. Inseminated animals were observed for return to oestrus and non-returns were verified for pregnancy 2 months after AI.

Prolonged oestrus was noticed on the second day in 13.34% of cases under category A, and 46.67% in category B. Oestrus persisted even on the third day in 5 and 18.34% of cases, respectively, under categories A and B. Conception rate of AI was 38.33% in category A and 41.67% in category B. Though conception rate of AI was better in category B, more double inseminations were involved, making actual number of AI per conception to be 3.52 as compared to 2.96 in category A. It could be concluded that incidence of prolonged oestrus was lesser in cows subjected to CM reducing the necessity for double AI and number of AI per conception. Hence clitoris massage forms a better management strategy in herds with increased incidence of prolonged heat and service failures.

**Key words:** Cattle, Clitoris massage, Infertility, Insemination, Oestrus

Increasing incidence of service failure has been reported among dairy cattle bred through AI (Ray and Misra 2000, Royal *et al.* 2000, Lucy 2001, Kutty and Ramachandran 2003). One of the major reasons for this includes untimely insemination consequent to difficulty of detecting the right time of heat (Senger 1994). Various aberrations of oestrus manifestations are experienced in animals bred continuously through AI, which include long duration of heat lasting for 2 or 3 days (Kutty and Ramachandran 2000, Nasir 2003) instead of 12–18 h normally expected. Prolongation of heat is attributable to insufficient luteinisation of granulosa cells and delayed ovulation leading to service failure unless AI is done more than once during the same heat period (Arthur *et al.* 1989, Duchens *et al.* 1995, Gafius *et al.* 2002).

Clitoris and vestibule are the two segments of the female reproductive tract provided with sensory innervations (Roberts 1971, Bearden and Fuquay 1980). Anatomical features of clitoris gives indication of its biological role in conveying biological signals to the female system regarding

mating process so as to initiate hormonal mechanisms leading to ovulation and cessation of heat signs (Nikolakopoulos *et al.* 2000). Chance of stimulating clitoris and vestibule appears to be minimal in the process of AI and the consequent lack of sensory stimulation in AI bred animals can be thought to affect the biological signals for ovulation (Randel *et al.* 1975, Pant *et al.* 2001) leading to persistence of heat signs, delayed ovulation and low success rate. Hence the present study was carried out with the objective of finding out the effect of clitoris massage along with AI on duration of oestrus and conception rate in crossbred cattle.

### MATERIALS AND METHODS

The study was carried out at the Livestock Research Station, Thiruvazhamkunnu of Kerala Agricultural University. Heat detection was done by manual observation 2-or 3- times each day and the major indicator for heat detection was mucous flow since most of the behavioural signs were feeble or absent (Nasir 2003). AI was carried out in the afternoon each day after confirming the animals being in heat through clinico-gynaecologic examination. Frozen semen with around 20 million sperms in 0.25 ml straws and

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having post thaw motility not less than 40.00% at frequent verification was used for AI.

The study was carried out for over 1 year. A total of 120 oestrus among cows ( $n = 72$ ) showing first to third heat postpartum were considered for the study. Animals reported to be in heat based on external features and behavioural changes were subjected to gynaecologic examination by the investigator and heat was confirmed based on characteristics of the mucus discharge and changes in the tubular reproductive tract and ovaries. The cows in proper heat were allocated randomly for insemination under categories A and B, and 60 inseminations were done under each of the categories. In category A insemination was followed by gentle massaging of the clitoris 2-to-3-times using the thumb inserted into the ventral commissure of vulval lips, while category B served as control having done insemination without any clitoris massage.

Normal duration of heat being less than 24 h, cases with persistence of heat signs beyond 24 h were categorised as prolonged heat. All the inseminated animals were observed for persistence of heat signs on subsequent days and those suspected to be in heat based on mucus discharge and other external signs were subjected to clinico-gynaecological examination for confirming persistence of the heat. Animals with prolonged heat on the second day were re-inseminated using semen of the same bull along with clitoris massage if it was done on the previous day. While AI was not repeated in few animals in heat even on the third day. The investigator himself did all the inseminations under the trial to avoid individual variations. Out of 72 cows inseminated, 41 that returned to heat in the subsequent cycle were included in the trial again as fresh cases if they have not undergone 3 inseminations already, while non-returns were verified for pregnancy after 2 months of AI. The data were recorded and analysed for description and significance (Chi-square test) of the findings.

## RESULTS AND DISCUSSION

Details of prolonged heat in animals subjected to AI with or without CM are compared in Table 1. Out of the 60 inseminations done on the first day of heat with CM, 21.67% animals showed heat signs on the second day, and upon verification 13.33% were in heat. Again on the third day 6.67% of those inseminated on the first day had external indications of heat, however, only 5% were still in heat. In contrast, among 60 inseminations done without CM, 55 and

26.67% were suspected to be in heat on second and third days of AI, respectively, and out of them 46.67% and 18.33%, respectively, were confirmed to have prolonged heat. Three animals that showed no heat signs on second day were also there among the animals in heat on third day attributable to split heat. Even on the fourth day few animals showed certain external signs suggestive of heat and 5 (8.33%) of them were still having internal changes of heat. There was a difference of 33.33% and 13.33%, respectively, between proportions of animals in experimental and control groups showing prolonged heat on second (13.33% versus 46.67%) and third days of AI (5.00% versus 18.33%).

Details of success rate of AI under experimental and control categories are shown in Table 2. Out of the 60 oestrous cycles in which inseminations were done under experiment category 23 (38.33%) became pregnant and the total number of AI required was 68 with 2.96 inseminations per conception. While in control category, 25 conceptions (41.67%) were obtained from 60 oestrous cycles requiring a total of 88 inseminations making AI per conception 3.52.

Conception rate obtained appears to be better in controls, and is attributable to many double AI done consequent to prolonged oestrus in this category. However the benefit of double AI appears to be marginal since 20 (33.33%) inseminations were done in control category in excess to that in experimental group to get additional 2 conceptions (3.34%). Influence of double AI on conception rate in experimental and control categories is compared in Table 3.

In experimental group conception rate was the same irrespective of whether double AI was done or not. However in control group conception rate was almost double (57.14% versus 28.13%) in animals given double AI compared to those not given. Also in both categories together conception rate was much better in double AI cases than no double AI (52.78% versus 34.52%). This in turn shows contribution of double AI in improving conception rate in animals manifesting prolonged heat, and is a useful strategy for animals managed in farms. However, household production system prevalent in villages especially in Kerala makes double AI very inconvenient and escalates the cost of breeding (Kutty and Ramachandran 2000).

Low proportion of double AI in animals inseminated under category A may be attributed to shortening of the heat period in these animals consequent to clitoris massage. Conception rate in those manifested prolonged heat even after CM did not improve by double AI indicating conception failure in

Table 1. Persistence of heat signs on the second and third day from the onset of signs in animals subjected to AI with and without CM

| Category | Oestrus on first day | Oestrus persisting on second day |             | Oestrus persisting on third day |             |
|----------|----------------------|----------------------------------|-------------|---------------------------------|-------------|
|          |                      | Suspected                        | Confirmed   | Suspected                       | Confirmed   |
| A        | 60                   | 13 (21.67%)                      | 8 (13.33%)  | 4 (6.67%)                       | 3 (5.00%)   |
| B        | 60                   | 33 (55.00%)                      | 28 (46.67%) | 16 (26.67%)                     | 11 (18.33%) |

Table 2. Success rate of AI compared between experimental and control categories

| Category | Oestrus cycle studied | Inseminations done |            |       | Pregnancy recorded | Conception rate per |               |
|----------|-----------------------|--------------------|------------|-------|--------------------|---------------------|---------------|
|          |                       | First day          | Second day | Total |                    | Oestrus cycle       | Inseminations |
| A        | 60                    | 60                 | 8          | 68    | 23                 | 38.33%              | 33.82%        |
| B        | 60                    | 60                 | 28         | 88    | 25                 | 41.67%              | 28.41%        |

them due to other reasons. Altogether 16 inseminations in these animals resulted only in 3 conceptions making number of AI per conception more than 5. Beneficial effect of CM on conception rate was reported in dairy cattle (Singh *et al.* 2001) and beef cattle (Short *et al.* 1979) and is believed to be through enhancement of ovulation and thus causing early cessation of heat (Pant *et al.* 2001 and Singh *et al.* 2001), so that availability of active sperms are ensured at the time of ovulation. Otherwise delayed ovulation causes fertilization failure through ageing of sperms unless fresh insemination is done in the form of double AI (Arthur *et al.* 1989).

In 32 (53.33%) oestrus periods, single insemination was done without any clitoris massage (category B) and the success rate was only 28.13% (Table 3), showing the usual conception rate expected in the herd without any of the interventions. This can be attributed to the high incidence of oestral aberrations in the herd like gradual disappearance of behavioural signs and increasing incidence of prolonged heat (Nasir 2003). Remaining 28 cases of category B required double inseminations indicating high incidence (46.67%) of prolonged oestrus in the herd. With clitoris massage occurrence of prolonged heat reduced to 13.34% in category A, making a difference of 33.33% from AI without CM (Table 1). Moreover conception rate of single AI with CM increased by 10% from that of AI without CM (38.46% versus 28.13%) taking an average of 2.60 and 3.56 AI per conception respectively (Table 3).

Highest conception rate of oestrus was obtained for double

AI without clitoris massage (57.14%), even though number of AI required per conception was 3.5. This strategy escalates the breeding cost very much and is very inconvenient especially in small holder production systems. Clitoris massage – a very simple intervention benefits through reducing the occurrence of prolonged heat considerably ( $P < 0.01$ ), thus reducing the necessity for double AI, number of inseminations per conception ( $P < 0.01$ ) and improving the success rate of single AI, thus causing overall reduction in the cost of breeding. This is very clear from the figure that in category A, 23 conceptions were obtained from just 68 inseminations, while in category B, 25 conceptions necessitated 88 inseminations. Extra cost incurred for additional 20 inseminations done at village level for 2 extra pregnancies has to be given due concern. Hence practising clitoris massage with AI appears more economic management strategy than double AI in herds with prolonged heat, while in larger herd with breeding facilities, clitoris massage combined with double AI of those with persistence of heat signs can be recommended.

It is concluded that clitoris massage shortens the oestrus duration in crossbred cattle herds with prolonged oestrus, in terms of number of days of showing the heat signs. This in turn reduces the necessity for double AI and improves the conception rate of single AI, lowering number of services per conception. Hence clitoris massage can be recommended as a simple management tool to improve conception rate in animals exhibiting prolonged heat and thus to reduce the

Table 3. Influence of double AI on conception rate in experimental and control categories

|               | Double AI done |        | Double AI not done |        | Total |        |
|---------------|----------------|--------|--------------------|--------|-------|--------|
|               | Number         | %      | Number             | %      | Total | %      |
| Category A    |                |        |                    |        |       |        |
| Total         | 8              | 100.00 | 52                 | 100.00 | 60    | 100.00 |
| Conceived     | 3              | 37.50  | 20                 | 38.46  | 23    | 38.33  |
| Not conceived | 5              | 62.50  | 32                 | 61.54  | 37    | 61.67  |
| Category B    |                |        |                    |        |       |        |
| Total         | 28             | 100.00 | 32                 | 100.00 | 60    | 100.00 |
| Conceived     | 16             | 57.14  | 9                  | 28.13  | 25    | 41.67  |
| Not conceived | 12             | 42.86  | 23                 | 71.88  | 35    | 58.33  |
| Category A+B  |                |        |                    |        |       |        |
| Total         | 36             | 100.00 | 84                 | 100.00 | 120   | 100.00 |
| Conceived     | 19             | 52.78  | 29                 | 34.52  | 48    | 40.00  |
| Not conceived | 17             | 47.22  | 55                 | 65.48  | 72    | 60.00  |

breeding cost. However, further studies are very much indicated to understand the hormonal mechanisms associated with clitoris massage, so that the intervention can be standardised and popularised.

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