

## **Oestrus synchronization and conception rate in Kedah-Kelantan cattle following treatment with CIDR-B and cloprostenol**

(Penyelarasan estrus dan kadar kebuntingan lembu Kedah-Kelantan selepas rawatan dengan CIDR-B dan kloprostenol)

J. A. Johari\*, E. Daud\*, M. Y. Mohd. Khusahry\*\* and M. A. Dollah\*\*

Key words: CIDR-B, cloprostenol, oestrus synchronization, KK cows

### **Abstrak**

Keberkesanan penggunaan alat “Controlled Internal Drug Releasing” (CIDR-B) selama 12 hari berbanding dengan dua kali suntikan kloprostenol untuk menyelaraskan estrus pada lembu betina Kedah-Kelantan (KK) telah dinilai. Sebanyak 148 ekor lembu betina KK digunakan dalam tiga percubaan yang dijalankan di Stesen Penyelidikan MARDI di Kluang, Johor. Lembu betina KK dibahagikan secara rawak kepada dua kumpulan. Kumpulan 1 diberi dua suntikan kloprostenol pada jarak waktu 12 hari dan kumpulan 2 dimasukkan CIDR-B ke dalam farajnya selama 12 hari. Lembu betina dikesan estrus dan disuntik dengan air mani beku 12 jam selepas estrus dikesan. Kebuntingan lembu ditentukan dengan ujian aras hormon progesteron dalam darah dan kepalpatan melalui rektum 60 hari selepas pernianian beradas. Kadar estrus kumpulan 2 lebih tinggi ( $p < 0.05$ ) daripada kadar estrus kumpulan 1. Hanya 32% dan 52% lembu betina KK kumpulan 1 dan 2 masing-masing menunjukkan estrus 48 jam selepas rawatan. Kadar kebuntingan kedua-dua kumpulan lembu rendah berdasarkan kepalpatan rektum berbanding dengan kadar kebuntingan yang berpandukan ujian aras hormon progesteron dalam darah. Kadar kematian embrio pada peringkat awal kebuntingan tinggi. Kadar kelahiran lembu KK yang diberi rawatan CIDR-B selama 12 hari (44.3%) setanding dengan kadar kelahiran lembu betina yang diberi dua suntikan kloprostenol (42.2%). Kajian ini menunjukkan bahawa estrus yang subur boleh diaruhkan dengan rawatan CIDR-B selama 12 hari setanding dengan dua suntikan kloprostenol. Bagaimanapun, kedatangan estrus yang diaruhkan tidak serentak bagi membolehkan pernianian beradas dilakukan pada waktu yang ditetapkan.

### **Abstract**

The effectiveness of CIDR-B treatment for 12 days compared with the double injections of cloprostenol for oestrus synchronization in Kedah-Kelantan (KK) cows was evaluated. A total of 148 KK cows were used in three trials conducted at MARDI Research Station, Kluang, Johor. Cows were allocated randomly into two groups. Group 1 was given two injections IM of cloprostenol at 12 days apart; group 2 inserted with CIDR-B devices for 12 days. The cows in both groups were detected for oestrus behaviour and were artificially inseminated (AI) 12 h later. Pregnancy rate was diagnosed by plasma progesterone assay and

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\*MARDI Research Station, Kluang, P. O. Box 525, 86009 Kluang, Johor, Malaysia

\*\*Livestock Research Centre, MARDI Headquarters, P. O. Box 12301, 50774 Kuala Lumpur, Malaysia

Authors' full names: Johari Jiken Abdullah, Daud Endam, Mohd. Khusahry Mohd. Yusof and Mohamad Aziz Dollah

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by rectal palpation at day 60 after insemination. Oestrus response within 96 h post-treatment in the CIDR-B treated group was significantly higher ( $p < 0.05$ ) than in the cloprostenol-treated group. Only 32% and 52% of the KK cows treated with cloprostenol and CIDR-B respectively were detected for standing oestrus and were AI within 48 h post-treatment. The pregnancy rates of KK cows in cloprostenol and CIDR-B treated groups diagnosed per rectum were lower than the pregnancy rates diagnosed based on plasma progesterone profile. High early embryonic mortality was observed. The calving rate (44.3%) of KK cows treated with CIDR-B for 12 days was comparable with that of KK cows treated with the standard double injections of cloprostenol (42.2%). This study shows that CIDR-B treatment for 12 days is effective in inducing fertile oestrus and is comparable with the standard double injections of cloprostenol regime. However, it did not produce a tightly synchronized oestrus necessary for fixed predetermined inseminations.

### Introduction

The main reason for synchronizing oestrus in cattle is to increase the use of artificial insemination (AI) in extensively managed beef cattle and dairy heifers (Mauleon 1974). In Malaysia, the use of AI in beef cattle has been very limited mainly due to unsatisfactory conception rate as a result of poor oestrus detection (Mohamad 1983). The adaptation and adoption of oestrus synchronization technology would, therefore, enhance the used of AI in an effort to upgrade the productive performance of Kedah-Kelantan (KK) cattle through the use of semen from exotic bulls. In embryo transfer programme, synchronization of oestrus between donor and recipient cows is necessary for optimum pregnancy rate (Rowson et al. 1972; Sreenan et al. 1975). Synchronization of oestrus can be achieved through the use of prostaglandin F<sub>2</sub>α (PGF<sub>2</sub>α) (Roche 1974b) and its analogues (Jackson et al. 1983; Seguin et al. 1983), the use of progesterone (Lamond 1964; Roche 1974a) or its analogues (Randel et al. 1972), a combination of progesterone and estradiol (Roche 1976), and a combination of prostaglandin and progestagen (Smith et al. 1981; Smith et al. 1984; Roche 1976, 1986; Spolt et al. 1983; Whittier et al. 1996). Prostaglandin and its synthetic analogues, such as cloprostenol (*Estrumate* and *Coopers*) and fenprosteno (l) (*Prosalvin* and

*Intervet*), have been used for many years as the main means of oestrus synchronization in cattle. They have luteolytic effect on existing functional corpus luteum (CL) but is ineffective in the early stage of the oestrus cycle (Battista et al. 1984; Kiracofe et al. 1985). The recommendation of fixed time AI, at 72 h and 96 h following two injections of PGF<sub>2</sub>α 11–12 days apart, was seen as a major breakthrough (Lauderdale 1972; Roche 1974a). Fertility rate of cattle following oestrus synchronization with PGF<sub>2</sub>α and its analogue, and fixed time AI, however, have been low and variable (Lauderdale et al. 1974; Nancarrow 1976; Jainudeen and Cameons 1977; Nordin et al. 1980; Tan et al. 1984; Tan et al. 1986). These were attributed to lack of precise synchrony of oestrus following treatment with PGF<sub>2</sub>α and its analogue. Optimum fertility rate was achieved when AI was performed after oestrus was observed following induction with PGF<sub>2</sub>α or its analogue (Roche 1974a; Hafs and Manns 1975; McIntosh et al. 1984; Tan et al. 1986).

Progesterone was the hormone originally used to synchronize oestrus in cattle (Lamond 1964). Later, potent analogues of progesterone, such as melengesterol acetate (MGA) (Randel et al. 1972), 6-methyle-17 acetoxy-progesterone (MAP) and *Synchro-Mate B* (SMB) (Hixon et al. 1981), were developed and used for

oestrus synchronization. Long-term treatment with progesterone of more than 14 days appeared to produce better synchrony of oestrus than the PGF2 $\alpha$  regimes, but the extended use of exogenous progesterone has been shown to reduce pregnancy rate (Roche 1974a). The development of progesterone releasing intra vaginal device (PRID) has enhanced the use of progesterone as a synchronization drug. Although the device is very effective, skill and experience are required to place the device into the vagina. The PRID may cause trauma and discomfort to the cattle, and sometimes up to 10% of the devices may be lost from the cows before the treatment is terminated (Roche 1976).

An alternative intra vaginal device, Controlled Internal Drug Releasing (CIDR-B) device, was developed jointly by Raukura Agriculture Research Centre and the Agriculture Division of the Carter Holt Harvey Plastic Products in Hamilton, N.Z. (Macmillian et al.1991). The CIDR-B device produced similar result as PRID but caused little trauma and less discomfort to the animal. This device has great potential for the strategic usage of progesterone to improve reproductive efficiency in cattle (Macmillian and Peterson 1993). This study was, therefore, conducted to evaluate the effectiveness of 12-day treatment with CIDR-B compared with the standard double injections of cloprostenol for oestrus synchronization in KK cattle.

### Materials and methods

A total of 148 KK cows were used in this study conducted at MARDI Research Station, Kluang, Johor. The KK cows were 4–8 years of age and were between 60 and 90 days postpartum. All cows weighed at least 220 kg and had a body condition score of 5 or more (1 = emaciated; 10 = obese). They were confirmed by rectal palpation to have normal reproductive tracts and were observed to exhibit normal oestrus cycles. The cows were allocated randomly without reference to the stage of oestrus cycles to

Table 1. Experimental design

Trial	Month/year	No. of cows treated	
		Cloprostenol	CIDR-B
1	Mar 1994	30	30 (3)
2	June 1994	20	20 (1)
3	Oct 1994	24	24 (1)

( ) No. indicates cows lost CIDR devices 1–2 days after insertion. These cows were excluded from the analysis

two groups: cloprostenol treatment (cows received two intramuscular injections of 0.5 mg cloprostenol, 12 days apart) and CIDR-B treatment (cows were inserted CIDR-B devices into the vagina for 12 days). The one-year experiment consisted of three trials (*Table 1*).

The cattle in both groups were grazed on improved pasture of *Brachiaria decumbens* during the day and kept in the holding yard during the night. Palm kernel cake was given to the animals as supplementary feed at 1 kg/head daily. Water and high cobalt mineral lick-blocks were made available to the animal ad libitum.

The animals were checked for oestrus behaviour continuously 24 h daily, after removal of CIDR-B or after the second injection of cloprostenol. A team of three observers were involved in a 12-h shift for oestrus detection. No bulls were used for oestrus detection.

The animals were observed for oestrus behaviour in the holding yard at night and in the paddock during the day. An animal in oestrus was defined as one that stood to be mounted or showed a clear vaginal mucus discharge (Godke and Kreider 1977). Those cows in “standing” oestrus were inseminated with frozen thawed semen approximately 12 h after the standing oestrus.

Blood samples were collected twice weekly beginning at observed oestrus till 3 weeks after AI to monitor conception rate. The samples were collected from the jugular vein in heparinized vacutainer tubes and chilled immediately in ice. The samples

were centrifuged and the plasma stored at 20 °C until analysis for progesterone. The plasma progesterone concentration was determined by solid-phase radioimmunoassay (RIA). The assay was performed using the coat-a-count progesterone kit (Diagnostic Products Corporation, Los Angeles, USA). The progesterone concentration in the plasma sample was calculated using a log/it-log programme on a computer. The inter-assay and intra-assay variation was 8.5% and 5.5% respectively.

Pregnancy was determined by plasma progesterone profile and later confirmed by rectal palpation approximately 60 days following insemination. The pregnancy rate was defined as the percentage of cows pregnant from the total number of cows treated in the group. The differences in the pregnancy rate and calving rate between the treatment groups in the three trials were tested using analysis of variance (SAS 1984).

**Results and discussion**

The average distribution of intervals to the onset of oestrus after treatment with cloprostenol and CIDR-B are summarized in Table 2. Between trials, variation in the synchrony response was minimal. The proportion of animals detected in standing oestrus at 48 h following CIDR-B removal or the second injection of cloprostenol was 52% and 32% respectively. By 96 h post-treatment, 75.7% (56 out of 74) of animals in group 1 exhibited oestrus behaviour compared with 92.8% (64 out of 69 ) of

animals in group 2. The mean interval from the second injection of cloprostenol or following the removal of CIDR-B to standing oestrus was 61.25 ± 17.3 h and 59.9 ± 19.2 h respectively.

One of the main objectives of oestrus synchronization in cattle is to have all treated animals in oestrus at a precisely known post-treatment intervals so that all the animals can be inseminated at a fixed predetermined time. The 12-day CIDR-B treatment or the two-injection cloprostenol treatment in this study did not produce a tightly synchronized oestrus in a high percentage of KK cows. Lauderdale et al. (1974) indicated that the lack of precision of oestrus in cattle treated with PGF2α could be the cause of low and variable fertility rate following fixed time AI at 72 h and 96 h. The stage of cycle when PGF2α was injected also influenced the degree of synchrony of oestrus (Tanaka and Hann 1984).

Short-term treatment of cattle with CIDR-B for less than 14 days was reported not to produce precised synchrony of oestrus (Macmillan and Peterson 1993). Various studies, however, had reported that combined short-term treatment with progestagen for 7 days and an injection of PGF2α produced relatively precise synchrony of oestrus with satisfactory fertility rates (Roche 1974a; Smith et al. 1981; Smith et al. 1984; Johari et al. 1990). The combined treatment of CIDR-B for 7 days plus an injection of PGF2α at device removal or 2 days before the device

Table 2. Cows detected in oestrus at specific intervals after the second injection of cloprostenol or after removal of CIDR-B device

Group	Treatment	No. of cows treated	No. of cows in oestrus at 3 intervals		
			≤49 h	49–72 h	73–96 h
1	Cloprostenol (2 injections)	74	24(32.4%)a	25(33.8%)a	7(9.5%)a
2	CIDR-B (12 days)	69	36(52.2%)b	15(21.7%)b	13(18.8%)b

Mean values with different letter differ significantly between groups (*p* <0.05)

removal, however, did not improve precision of synchrony of oestrus that would allow every treated animal to be inseminated within 24 h (Macmillan and Peterson 1993). A 12-day treatment with CIDR-B and an injection of cloprostenol 24 h prior to CIDR-B removal treatment regime in donor and recipient cows produced a high degree of synchrony of oestrus between donor and recipients (Johari, J. A., Superovulation and embryo transfer, MARDI, Kluang, unpublished 1995).

The percentage of cows observed in oestrus by day 4 post-treatment was significantly higher ( $p < 0.05$ ) in the CIDR-B than in the cloprostenol-treated group (Table 2). The low response rate in the cloprostenol-treated group could be due to the effect of stage of oestrus cycle when the treatment was initiated. Cattle injected during day 10 to 15 of the oestrus cycle had greater oestrus response than those injected before day 5 (King et al. 1982).

Prostaglandin or its analogue was unable to precipitate luteolysis in cattle when injected before day 5 (day 0 = day of oestrus) (Lauderdale 1972; Kiracofe et al. 1985).

As the conception rates and calving rates did not differ across trials, the pooled data is presented in Table 3. The conception rate (the number of animals conceived to the number of animals inseminated) based on plasma progesterone assay was 55.3% and 66.8% for group 1 and group 2 respectively. The conception rate based on rectal palpation which was 42.2% and 44.3% for

group 1 and 2 respectively, was not significantly different between the two groups.

Plasma progesterone profile indicated that embryonic mortality occurred from day 21 to 50 post-insemination in the cows diagnosed non-pregnant at 60 days postpartum by rectal palpation. The cause of embryonic death, however, could not be identified in this study. High embryonic mortality has been indicated as a major cause of low fertility in cattle. Tan et al. (1986) also observed that the low conception rate in KK cows treated with cloprostenol and inseminated at observed oestrus was due to early embryonic death.

In this study, the 12-day CIDR-B treatment produced a calving rate comparable with that of two injections of cloprostenol (Table 3). The calving rates obtained were also comparable with the calving rate obtained by Macmillan and Peterson (1993) who used a 14-day CIDR-B treatment regime on dairy heifers. The calving rates reported were 44.4% and 45.0% based on the total number of animals treated and the total number of animals inseminated respectively. It has been reported that treatment of cattle with progestagen for less than 14 days did not reduce conception rate (Wiltbank and Kassan 1968; Roche 1974b, 1976). However, for most of the study, a luteolytic agent was incorporated in the treatment regime. The 12-day CIDR-B treatment or the two-injection cloprostenol treatment in

Table 3. Pregnancy and calving rates of KK cows inseminated by 96 h post-treatment with cloprostenol or CIDR-B

Treatment	No. of cows treated	Cows detected in oestrus and AI		Interval to oestrus post-treatment (mean $\pm$ SD, h)	Pregnancy rate (%)		Calving by AI cows (%)	Calving by total cows (%)
		No.	%		By prog. assay	By rectal palpation		
Cloprostenol (group 1)	74	56	75.7a	61.3 $\pm$ 17.3	55.3a	42.2	53.6	42.2
CIDR-B (group 2)	69	64	92.8b	57.9 $\pm$ 19.2	66.8b	44.3	45.3	44.3

Mean values with different letter differ significantly between groups ( $p < 0.05$ )

this study produced a fertility rate that is comparable with the earlier report (Johari et al. 1990). The calving rate could be improved if the incidence of embryonic death was reduced. The cause of embryonic death has to be identified and remedied to achieve optimum calving rate following oestrus synchronization using either CIDR-B or cloprostenol.

### Conclusion

CIDR-B treatment for 12 days is effective in inducing fertile oestrus and is comparable with the standard double injections of cloprostenol. The degree of synchrony of oestrus, however, was not precise to permit a fixed predetermined insemination. The calving rate of KK cattle treated with CIDR-B for 12 days was comparable with the calving rate of cloprostenol-treated cattle. As with cloprostenol treatment, to obtain optimum conception rate, it is recommended that cattle treated with CIDR-B for 12 days must be observed for oestrus and inseminated 12 h following heat detection. Treated cows should be in good body condition and given adequate plan of nutrition. The benefit of using CIDR-B for 12 days for oestrus synchronization in cattle is that it facilitates the use of AI in a majority of KK cows during a short 4-day period. Although prostaglandin is the hormone of choice for oestrus synchronization in a planned breeding programme, CIDR-B offers alternatives in case where a veterinarian is not available.

More field research should be carried out to develop oestrus synchronization regime which could produce precise synchrony of oestrus necessary for fixed predetermined insemination with high fertility. The technique should be applicable to commercial farms practising planned breeding programme. The cost effectiveness of the treatment regime should also be evaluated.

### Acknowledgements

The authors wish to thank Mr Rusli Abdul Hamid, Mr Hailan Hassan Tami and Mr Ajis Hassan for their assistance in the study. Special thanks are due to Mr Mashodi Surip for inseminating the animals and Mr Chiew Key Szu for the statistical analysis.

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