

## Using the same CIDR up to three times for estrus synchronization and artificial insemination in dairy goats

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**ABSTRACT.** The objective of this study was to evaluate the effectiveness of reusing a controlled internal drug release (CIDR) device for up to three times in the reproductive performance of dairy goats raised in the semi-arid zone of northeastern Brazil. Forty-five goats were allocated into three hormone treatments, as follows: CIDR1x, treated with new CIDR during nine days. Two days prior to device removal, injections of 75 µg d-cloprostenol and 300 IU equine chorionic gonadotropin (eCG) were administered. For the other treatments, the same hormone protocol was used, differing only by the use of the same CIDR for a second time in CIDR2x and for a third time in CIDR3x. The interval from device removal to the onset of estrus ( $13.3 \pm 1.1$ h vs.  $13.8 \pm 2.6$ h vs.  $13.3 \pm 1.4$ h), as well as estrus duration ( $33.6 \pm 7.3$ h vs.  $29.6 \pm 3.2$ h vs.  $32.8 \pm 4.5$ h), did not differ ( $p > 0.05$ ) among groups CIDR1x, CIDR2x and CIDR3x, respectively. All synchronized females were found to be in estrus. The overall fertility and prolificacy after artificial insemination were 82.2% and 1.9 kids, respectively, without significant difference ( $p > 0.05$ ) among treatments. The use of the same CIDR for up to three times was effective using 9-day estrus synchronization protocols in dairy goats.

**Keywords:** hormone treatment, small ruminant, Tropical region, vaginal device.

**RESUMO.** Uso do mesmo CIDR por até três vezes para sincronização do estro e inseminação artificial de cabras leiteiras. Objetivou-se avaliar o efeito da utilização do mesmo dispositivo de liberação controlada de drogas (CIDR) por até três vezes sobre o desempenho reprodutivo de cabras leiterias exploradas no semiárido do Nordeste Brasileiro. Foram utilizadas 45 cabras divididas em três tratamentos de sincronização do estro, sendo: CIDR1x, tratadas com CIDR novo durante nove dias. Dois dias antes da retirada do dispositivo, foram aplicados 75 µg de d-cloprostenol e 300 UI de gonadotrofina coriônica equina (eCG). Para os demais tratamentos, foi utilizado o mesmo protocolo hormonal, diferindo apenas pelo uso do mesmo CIDR pela segunda vez no grupo CIDR2x e uso pela terceira vez no grupo CIDR3x. O intervalo entre a retirada do dispositivo e o início do estro ( $13,3 \pm 1,1$ h vs.  $13,8 \pm 2,6$ h vs.  $13,3 \pm 1,4$ h), bem como, a duração do estro ( $33,6 \pm 7,3$ h vs.  $29,6 \pm 3,2$ h vs.  $32,8 \pm 4,5$ h) não diferiram ( $p > 0,05$ ) entre os grupos CIDR1x, CIDR2x e CIDR3x, respectivamente. Todas as fêmeas sincronizadas foram identificadas em estro. As médias de fertilidade e prolificidade média após inseminação artificial foram, respectivamente, de 82,2% e 1,9 crias, não havendo diferença significativa ( $p > 0,05$ ) entre os tratamentos. A utilização do mesmo CIDR por até três vezes foi viável na sincronização do estro de caprinos leiteiros.

**Palavras-chave:** tratamento hormonal, pequenos ruminantes, região Tropical, dispositivos vaginais.

### Introduction

An efficient Artificial Insemination (AI) program requires the use of protocols that assure acceptable pregnancy rates with low variability in the response within flocks. An efficient pregnancy rate is closely linked to the synchronization of ovulations obtained in treated females. In goats, estrus synchronization is commonly performed with

vaginal sponges impregnated with fluorogestone (FGA) (FERNANDEZ-MORO et al., 2008; FREITAS et al., 1996) or medroxyprogesterone acetates (MAP) (FONSECA et al., 2005; LEHLOENYA et al., 2005), or using controlled internal drug release (CIDR) devices impregnated with progesterone (AMORIM et al., 2008; MENCHACA et al., 2007). All of these intravaginal devices are

associated with equine chorionic gonadotropin (eCG) and prostaglandin- $F_{2\alpha}$  analogues, such as d-cloprostenol. Intravaginal sponges and CIDRs have been found to be equally effective in controlling estrus and ovulation in goats (MOTLOMELO et al., 2002; OLIVEIRA et al., 2001; ROMANO, 2004).

CIDR devices offer important advantages. Firstly, they contain low natural doses of progesterone (WHEATON et al., 1993). Moreover, unlike intravaginal sponges, CIDRs do not absorb or obstruct drainage of vaginal secretions, resulting in less foul-smelling discharge upon removal (MOTLOMELO et al., 2002; ROMANO, 2004). Finally, these devices also induce earlier and more compact synchronization and have a better retention rate during treatment (MOTLOMELO et al., 2002).

However, CIDRs are considered to be expensive when the benefit/cost ratio is evaluated and compared to vaginal sponges. For this reason, some researchers such as Guido et al. (1999) and Amorim et al. (2008) have evaluated the repeated use of CIDR in order to reduce costs without altering reproductive performance. Moreover, these researchers have used this device for up to two consecutive times, without significantly reducing protocol cost. The repeated use of CIDR should make its usage cheaper.

To reduce the cost of using CIDR for estrus synchronization, this study evaluated the effectiveness of using the same CIDR up to three consecutive times in the reproductive performance of dairy goats raised in the semi-arid zone of northeastern Brazil.

## Material and methods

### Animals and location

The experiment was carried out according to the International Laws for Animal Research and Experimentation. The experiment was conducted during the dry season on a private farm located in Santa Maria da Boa Vista, Pernambuco State, northeastern Brazil (8° 48' 00" S, 39° 49' 12" W). Mean annual rainfall is 463.10 mm, distributed from November to April. The average annual temperature at the site is 25.5°C.

A total of 24 Saanen and 21 Alpine dairy does were used as experimental animals in the present study. All were cycling, multiparous, at the end of lactation and non-pregnant. They were allocated into three homogeneous groups in terms of breed, age and body condition score (BCS). The animals were 3.72 years old and had BCS equal to 2.50 (1-5 scale) at the beginning of the experiment.

The animals were submitted to a semi-intensive production system. In the morning, they were kept in irrigated pasture of *Panicum maximum* cv. Tanzânia, and during the afternoon they received indoor triturated Napier grass (*Pennisetum purpureum* Schum.) and commercial concentrate with 16.0% crude protein. Commercial mineral supplement and water were offered *ad libitum*.

### Estrus synchronization treatments

The does were allocated into three consecutive estrus synchronization treatments: In CIDR1x (n = 15), the females were treated for nine days with a new CIDR vaginal device (Eazi-Breed CIDR®, InterAg, New Zealand) containing 0.3 g progesterone. Two days (48h) prior to device removal, intramuscular injections of 75 µg d-cloprostenol (Ciosin®, Coopers, Brazil) and 300 IU equine chorionic gonadotropin (eCG; Novormon®, Syntex, Argentina) were administered. For the other treatments, the same hormone protocol was used, differing only by the use of the same CIDRs used for the CIDR1x group for a second time in CIDR2x (n = 15) and for a third time in CIDR3x (n = 15).

The CIDRs used for the CIDR2x and CIDR3x groups were cleaned in physiological solution and dried immediately after use. No storage was applied for CIDRs between CIDR1x and CIDR2x or CIDR2x and CIDR3x, as the CIDRs were used for the subsequent group immediately after being cleaned.

### Estrus detection

Does were tested for estrus with two vasectomized bucks starting 12h after device removal and every four hours for 60h. Does were considered to be in estrus when they allowed mounting by the vasectomized bucks.

### Artificial insemination

Semen was collected by using an artificial vagina from a proven fertile Alpine buck. Only ejaculates over 0.5 mL, mass activity  $\geq 3$  (0-5 scale) and sperm motility greater than 70% were used. Semen was extended in a coconut water solution as reported by Nunes (1998) in order to obtain a dose of at least  $300 \times 10^6$  spermatozoa in 0.25 mL. All treated does showing estrus behavior were inseminated once between 16 and 20h after the onset of estrus by transcervical route, using a vaginal speculum and cervical traction.

### Fertility rate and prolificacy

The pregnancy rate was confirmed by the occurrence of parturition, and prolificacy by the number of kids born.

### Statistical analysis

The parameters concerning breed, estrus response (interval from device removal to onset of estrus and estrus duration) and prolificacy were analyzed using analysis of variance (ANOVA). The Tukey test was used to determine the difference between treatments. Percentages of females in estrus and fertility rate were compared between treatments by using the Chi-square test. Results were presented as mean  $\pm$  SEM or percentage for statistical significance at 95% confidence interval.

### Results and discussion

In this study, differences in pregnancy rate and additional parameters between treatments were not affected by breed ( $p > 0.05$ ), and data were pooled for these factors.

All females (100.0%) from the three experimental groups submitted to estrus synchronization were identified to be in estrus. There was no significant difference ( $p > 0.05$ ) among treatments for the interval to onset of estrus, estrus length, fertility rate and prolificacy (Table 1).

**Table 1.** Percentage of does in estrus, mean ( $\pm$  SEM) interval to onset of estrus, estrus length, fertility rate and prolificacy of dairy goats.

Group	N*	Females in estrus (%)	Onset of estrus (h)	Estrus length (h)	Fertility (%)	Prolificacy (%)
CIDR1x	15	100.0	13.3 $\pm$ 1.1	33.6 $\pm$ 7.3	93.3	2.5 $\pm$ 0.3
CIDR2x	15	100.0	13.8 $\pm$ 2.6	29.6 $\pm$ 3.2	73.3	1.7 $\pm$ 0.4
CIDR3x	15	100.0	13.3 $\pm$ 1.4	32.8 $\pm$ 4.5	80.0	1.5 $\pm$ 0.3
Overall	45	100.0	13.5 $\pm$ 0.5	32.0 $\pm$ 1.4	82.2	1.9 $\pm$ 0.3

\*Number of animals. No significant differences were observed between treatments in the same column ( $p > 0.05$ ).

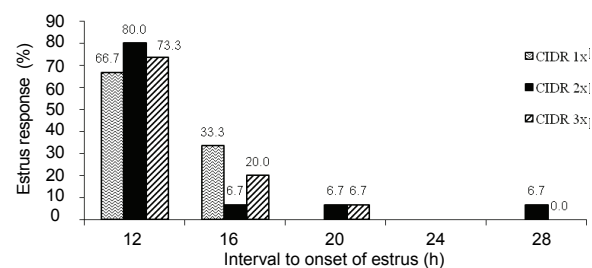
The use of the same CIDR devices for up to three consecutive times with 9-day protocols showed that CIDRs are efficient in synchronizing estrus in goats. The estrus response was the same for all treatments; this is in agreement with Guido et al. (1999) and Oliveira et al. (2001), who reported 100% of animals in estrus after 10-day and 9-day protocols with CIDR, respectively, using 100 IU eCG in Saanen goats.

In the present study, the mean estrus onset time was similar to Bitaraf et al. (2008), who reported an interval of 21.7  $\pm$  0.4h. However, it was shorter than the intervals reported by Guido et al. (1999) (42.0  $\pm$  4.6h), Motlomelo et al. (2002) (27.2  $\pm$  0.4h), Romano (2004) (40.2  $\pm$  10.5h) and Maffili et al. (2006) (35.0  $\pm$  5.9h).

Although those authors used CIDR for estrus synchronization in goats, the shorter interval from CIDR removal to onset of estrus in this study can be explained by the eCG doses used in each treatment. The higher the dose of eCG, the greater the ovarian

activity and tendency for anticipation of the estrus onset after the end of treatment (FONSECA et al., 2005). Furthermore, the frequency of estrus detection used in this study was greater (6 times day<sup>-1</sup>) than the ones used by those authors, increasing the probability of detection of onset of estrus.

The treatments featured low dispersion and, therefore, a strong synchrony for the onset of estrus (Figure 1), as 100.0, 86.7 and 93.3% had already shown signs of estrus 16h after device removal for, CIDR1x, CIDR2x and CIDR3x, respectively.



**Figure 1.** Distribution of onset of estrus after synchronization treatments in dairy goats

This demonstrates that 9-day protocols with new and reused CIDR can provide efficient estrus synchronization programs, and that these treatments could be used for fixed-time AI programs. The relationship between the early onset of estrus, ovulation and pregnancy rates after AI programs are in agreement with Baril et al. (1993) and Romano (2004). Both reports showed that when goats exhibited estrus until 30h after progestagen treatment, a fertility rate of 65% was observed following AI using frozen-thawed semen, but when estrus was observed at intervals of 49 or 72h after hormone treatment, a pregnant goat rate of only 25% was verified (BARIL et al., 1993).

Only one doe in CIDR3x was observed to show estrus before device removal. That female was inseminated, but did not become pregnant. One possible explanation for this fact is the reduced levels of progesterone in the device. Probably, these levels were not enough to suppress the hypothalamic-pituitary axis, resulting in estrus response (ROMANO, 2004; UNGERFELD; RUBIANES, 2002).

In this study, estrus length was similar for all treatments (Table 1). Mean estrus length was similar to those found by Oliveira et al. (2001) (36.0  $\pm$  4.5h), Motlomelo et al. (2002) (35.2  $\pm$  0.7h) and Maffili et al. (2006) (36.0  $\pm$  7.6h), and shorter than Romano (2004) (39.2  $\pm$  10.9h). These differences can be explained by the higher frequency of estrus

detection in the present study or by individual differences among breeds.

In the present study, mean fertility and prolificacy after AI using fresh extended semen was considered excellent (Table 1), as fertility was greater than those found by Motlomelo et al. (2002) (46.7%), Romano (2004) (63.0%), Bitaraf et al. (2008) (55.0%) and Ungerfeld and Rubianes (2002) (59.6%). Additionally, data obtained for fertility rate in this study are similar to other studies that used controlled mating to fertilize the females. Similarly, Guido et al. (1999), Oliveira et al. (2001) and Maffili et al. (2006) observed fertility rates of 100.0, 95.0 and 50.0%, respectively. Oliveira et al. (2001) did not find any difference in prolificacy between new and reused CIDR, either (1.50 vs. 1.40, respectively).

In this study, the favorable fertility rate results after AI were due to several aspects. Firstly, AI was performed between 16 and 20h after the onset of estrus. Therefore, the semen used in this study was deposited in the reproductive tract of females close to the moment of ovulation. According to Maffili et al. (2006), ovulation occurs  $25.0 \pm 7.0$ h after the onset of estrus. Moreover, the method of AI by cervical traction allowed the insemination gun to get through the cervical canal into the uterus in 100% of does.

## Conclusion

The use of the same CIDR for up to three times was effective in synchronizing the estrus of dairy goats. Further investigations should be done to determine how many times it is possible to reuse this device in 9-day protocols in dairy goats, and how long it is possible to conserve CIDR devices between treatments.

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