Original Research

Journal of Advanced Veterinary Research
(2025) Volume 15, Issue 2, 262-266

Controlled internal drug release (CIDR) is superior to improve the conception rate in adult cows and heifers with metestrus bleeding

Mohamed S. Rawy[1*], Hazem M.S. Mesalam[1], Ahmed S.A. Hassaneen[2, 3]

[1]Department of Theriogenology, Faculty of Veterinary Medicine, Minia University, Minia, Egypt.
[2]Department of Theriogenology, Obstetrics and Artificial Insemination, Faculty of Veterinary Medicine, South Valley University, 83523, Qena, Egypt.
[3]Department of Surgery and Theriogenology, Faculty of Veterinary Medicine, King Salman International University, Ras Sudr, South Sinai, Sinai, Egypt.

ARTICLE INFO

Recieved: 06 March 2025

Accepted: 25 March 2025

*Correspondence:

Corresponding author: Mohamed S. Rawy
E-mail address: sadawyvet84@yahoo.com

Keywords:

Fertility, Metrorrhagia, Metestrual bleeding, Post-estrual bleeding, Repeat breeding.

ABSTRACT

Efficient reproductive performance is crucial to maintain high cattle production including dairy and beef production. Cows with metestrual bleeding about 2 days after the end of estrus show lower chance to conceive. The current study aimed to compare the efficacy of double insemination and different hormonal treatments to improve conception rate in animals with metestrual bleeding. In the present study, a total number of 128 cows were used including 89 heifers and 39 pluriparous cows. The used animals were divided into 5 groups; control group (Ctrl; n=15) received no treatment, double insemination group (n=15), prostaglandin (PGF2α)-treated group (n=30), gonadotropin releasing hormone (GnRH)-treated group (n=28), and controlled internal drug release (CIDR; n=40). Ultrasonography to examine the reproductive organs and blood sampling at day 0, 3, 6, 18 were performed and serum samples were stored for estradiol (E2) and progesterone (P4) hormonal assay. Our results showed that, PGF2α, GnRH, and CIDR would be clinically effective in compare to ctrl or double insemination group to improve the conception rate in cows and heifers with metestrual bleeding. CIDR protocol showed superior efficacy with a conception rate of 87.5 % in compared to ctrl (13.3%) or double insemination (33.3%). CIDR treated group showed higher concentrations of both E2 and P4 levels in the treated cows and heifers at day 6 and day 18. While no differences were reported in the measurements of the ovarian cyclic structures between all groups. In conclusion, CIDR protocol for 7 days with PGF2α at day 6 and GnRH after CIDR removal by 48 hours would be the treatment protocol of choice to improve conception rate in cows and heifers with metestrual bleeding.

Introduction

Efficient reproductive performance is essential to improve cattle production (Consentini *et al*., 2021). Cattle production is mainly divided into two divisions: dairy production and beef production. However, in most of the developing countries, the same cows are used for both beef and milk production and therefore have a 'dual purpose' breeding aim (Galina and Geffroy, 2023).

Cows are known as a polyestrous animal species with estrous cycle throughout the year regardless the season. The estrous cyclicity continues except during pregnancy, about 6 weeks postpartum, and with certain pathological conditions (Perera, 2011). The average duration of estrus phase (period of sexual receptivity) is about 15 hours with a wide range of hours according to some factors including the breed, season, presence of bull, milk yield (Mičiaková *et al*., 2018; Pérez-Marín and Quintela, 2023). In cows, spontaneous ovulation takes place during the metestrus phase, about 12 hours after the end of estrus phase (Temesgen *et al*., 2022).

Metestrus phase is characterized as a period of hormonal transition from threshold estrogen (E2) levels during the estrus phase to maximum progesterone (P4) levels during the subsequent diestrus phase (Filant and Spencer, 2014). About 48 hours after the start of metestrus phase, cows and heifers may show a bright red sanguineous bloody discharge, coming mainly from the uterine blood vessels. And this phenomenon occurs irrespective to both service and conception (Dellinger *et al*., 2008). Metestrus bleeding occurs with higher incidence in heifers than in mature cows, likely due to rupture of the endometrial capillaries (Larson and Randle, 2008), as a process of diapedesis with denudation of the endometrial epithelium (Raven *et al*., 1948). The spiral shape of the endometrial capillaries in mature cows reduces the blood pressure effect (Sugiura *et al*., 2018). Despite, the fact that the appearance of metestrual bleeding discharges does not mean failure of conception, cows and heifers with metestrual bleeding have least chance to conceive (Giri and Yadav, 2000). Such failure of conception has been previously reported in both cows and heifers with metestrual bleeding (Kishore, 2006). Generally, the exact cause of conception failure or repeat breeding is not clear in many cases as unexplained infertility problem (Hassaneen *et al*., 2023). However, such infertility is likely due to either failure of fertilization or embryonic death. In addition, metestrual bleeding is an emerging predisposing of repeat breeding cows (Giri and Yadav, 2000).

Different managemental and therapeutic trials has been considered to improve the conception rate of repeat breeder cows and heifers with metestrus bleeding. Antimicrobial treatment was previously reported as the treatment of choice (Kutty, 2005), while hormonal based treatments such as gonadotrophin releasing hormone (GnRH) or P4 were considered as more effective alternatives to improve conception in such animals with metestrus bleeding (Marnikov and Vassilev, 1986; Kishore *et al*., 2005). Other studies reported the use of double insemination to improve conception rate in repeat breeding cows (Fleischmann, 1990; Sharma *et al*., 2002)

Therefore, the current study aimed to compare the efficiency of hormonal-based treatments including controlled internal drug release (CIDR), GnRH, PGF2α to other approaches such as double insemination or even no treatment (control) on conception rate of cows and heifers showing metestrual bleeding.

Materials and methods

Clinically healthy adult cows and heifers (n=128) were used in the study. The used animals included 89 heifers (1.80±0.26 years-old and body weight ranged from 230 to 320 kg) and 39 mature cows (6.6±1.08 years-old, number of parities of 2.8±0.73, and body weight ranged from 350 to 500 kg). Upon rectal and gynecological examinations, all used animals were free from any reproductive diseases including endometritis, cervicitis or vaginitis.

Ethical approval

The animals in the present study were handled in accordance with

ISSN: 2090-6277/2090-6269/

the standard protocol approved by Minia University, Faculty of Veterinary Medicine Ethics committee for Animal Use and Care (Number IRB-FVM-MU-2025-117).

Animals

All animals enrolled in this study (n=128) had a history of repeat breeding and showed met-estrual bleeding as fresh bloody vaginal discharges hanged from the ventral commissure of the vulva, soiling the tail or soiling the hind quarter of the animal (Fig. 1).

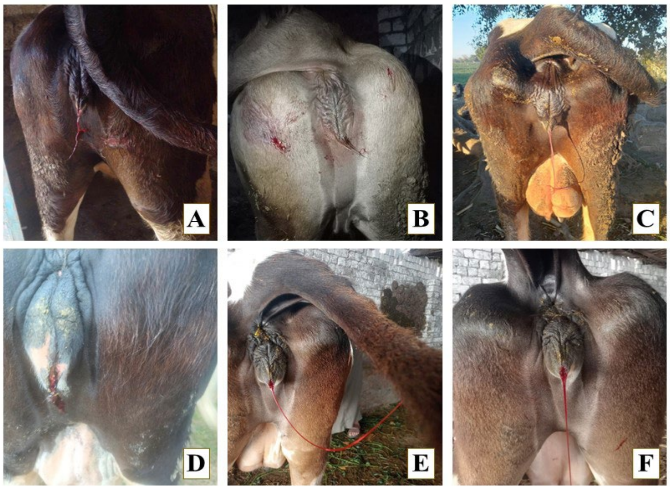

Fig. 1. Representative images of the metestrual/post-estrual bleeding in cows and heifers with metrorrhagia. Showing external genitalia contaminated with bloody discharges in a heifer (A), soiling of the hind quarter with post-estrual bleeding (B), light-red post-estrual bleeding hanged from the ventral commissure of the vulva (C), contaminated ventral commissure of the vulva with bloody discharges in mature cow (D), freshly red bloody discharges hanged between the tail and the vulva (E), or hanged from the ventral commissure (F).

Ultrasonography

Transrectal ultrasonography were performed using a real-time B-mode echo camera (Chison ECO 2 VET, manufactured by Chison Medical Technologies Co., Ltd., a company) connected to a 6-8 MHz linear array transrectal transducer. Ovarian structures including the dominant/mature graffian follicle (MGF) and the corpus luteum (CL) were measured. Ultrasonographic images were taken to the uterine horns, as well.

Blood sampling

Five-ml blood samples (day 0 (day of estrus), day 3, day 6, and day 18) were collected from the jugular veins into 5-ml plain vacutainer tubes to obtain serum. The blood samples were centrifuged at 3000 rpm for 20 minutes, and the harvested sera were aliquoted and stored at -20°C until hormonal assay for determination of estradiol (E2) and progesterone (P4) concentrations.

.

Hormonal assay

Determination of serum progesterone (P4) concentrations

Progesterone concentrations were determined using a commercially available P4 Enzyme Immunoassay kit (FERTIGNIX-PROG-EASIA). The range of the standards used was 0–11.5 ng/ml. The inter- and intra-run precision had a coefficient of variation of 2.9 and 4.8%, respectively. Values above 1 ng/ml (3.18 nmol/l) were considered indicative of luteal activity.

Determination of serum estradiol (E2) concentrations

Estradiol concentrations were determined using commercial E2 ELISA kits (Human Gesellschaft fur Biochemica und Diagnostica, Wiesbaden – Germany). The coefficient of variance of intra- and inter-assay were 5.2 and 9.3%, and the sensitivity of the assay was 3 pg/ml.

Experimental design

After transrectal ultrasonographic examinations, all heifers and cows were randomly categorized into four groups and subjected to one of the following treatments

Control group (Ctrl; n = 15)

All animals included in this group received no treatment and were assigned as control group.

Double Insemination group (n = 15)

All animals allocated to this group received two inseminations. The first insemination was performed during estrus, and the second at the day of bleeding. This method is one of the managemental treatments that is commonly used by local farmers in Egypt for treatment of repeat breeding and to improve conception rate in cows and heifers with metrorrhagia.

Prostaglandin (PGF2α)-injected group (n = 30)

All animals allocated to this group received intramuscular injection of 2 ml synthetic PGF2α analogue (estrumate; 500 µg cloprostenol sodium, MSD Animal health) after showing the met-estrual bleeding by one week or at day 9 or day 10 (Day 0 is day of estrus). Insemination was performed the induced heat following the PGF2α injection.

Gonadotropin-releasing hormone (GnRH)-injected group (n = 28)

All animals allocated to this group received intramuscular injection of 2 ml GnRH analogue (ovarelin; 100 µg gonadorelin diacetate, Bayer) at day 0.

Controlled-Internal drug releasing device (CIDR) group (n = 40)

In this group (n.= 40), a flexible T-shaped P4-containing CIDR device (1.38g P4) was gently inserted into the cow's vagina. The device was removed after 7 days. And PGF2α analogue was injected one day before the removal of CIDR. Then, two days after the CIDR removal, GnRH was injected followed by artificial insemination about 4 hours after GnRH injection.

Statistical analysis

Data was arranged using Microsoft excel 2010. All values were expressed as mean ± standard deviation (SD). Data was analyzed using SPSS program (Version 25) for windows. Results were considered statistically significant at the $P<0.05$ level.

Results

Cows in both PGF2α-treated group and CIDR-treated group did not show metestrual bleeding after the induced post-treatment heat/estrus.

Conception rate (%) in Control (Ctrl), Double Insemination, PGF2α, GnRH, and CIDR group

The conception rate (%) was significantly higher in the CIDR group (P value< 0.001) in comparison to the PGF2α, GnRH, Double Insemination and control group (Table 1).

Diameter of dominant follicle (cm; Mean ±SD) in Control (Ctrl), Double Insemination, PGF2α, GnRH, and CIDR group

No difference in the diameter of the dominant follicle (MGF; cm) was reported (P value= 0.735) between all treated groups. The diameter of the dominant follicle (cm; Mean ±SD) was 1.56±0.47, 1.58±0.50, 1.54±0.39, 1.49±0.29, 1.62±0.36, and in the Ctrl, Double Insemination, PGF2α, GnRH, and CIDR, respectively (Table 1, and Fig. 2).

Diameter of corpus luteum (cm; Mean ±SD) in Control (Ctrl), Double Insemination, PGF2α, GnRH, and CIDR group

No difference in the diameter of the corpus luteum (CL; cm) was reported (P value =0.075) between all treated groups. The diameter of the CL (cm; Mean ±SD) was 2.33±0.31, 2.35±0.30, 2.54±0.20, 2.43±0.31, and 2.49±0.29 in the Ctrl, Double Insemination, PGF2α, GnRH, and CIDR, respectively (Table 1, and Fig. 2).

Estrogen concentrations (E2 ng/ml; Mean ±SD) at day 0, day 3, day 6, and day 18 in Control (Ctrl), Double Insemination, PGF2α, GnRH, and CIDR group

This study showed that there was no significance difference (P value = 0.944) in the concentrations of E2 (ng/ml; Mean ±SD) between all the treated groups at day 0 with E2 levels (ng/ml; Mean ±SD) of 20.23±0.06, 20.23±0.07, 20.23±0.02, 20.23±0.05, and 20.24±0.08 in the Ctrl, Double Insemination, PGF2α, GnRH, and CIDR, respectively (Table 2). Concentrations of E2 (ng/ml; Mean ±SD) at day 3, day 6, and day 18 were significantly higher in the CIDR group (P value <0.001) compared to the Ctrl, Double insemination, PGF2α, and GnRH group (Table 2).

Progesterone concentrations (P4 ng/ml; Mean ±SD) at day 0, day 3, day 6, and day 18 in Control (Ctrl), Double Insemination, PGF2α, GnRH, and CIDR group

Our results showed that there was no significance difference (P value = 0.604) in the concentrations of P4 (ng/ml; Mean ±SD) between all the treated groups at day 0 with P4 levels (ng/ml; Mean ±SD) of 0.43±0.01, 0.43±0.01, 0.43±0.09, 0.42±0.02, and 0.43±0.04 in the Ctrl, Double Insemination, PGF2α, GnRH, and CIDR, respectively (Table 3). Progesterone level at day 3 (ng/ml; Mean ±SD) tended to be higher in the CIDR group (P value = 0.056) compared to the Ctrl, Double Insemination, PGF2α, and GnRH group (Table 3). Moreover, the levels of P4 (ng/ml; Mean ±SD) in the CIDR group were significantly higher at day 6 (P value = 0.002), and day 18 (P value = 0.012) compared to the Ctrl, Double insemination, PGF2α, and GnRH group (Table 3).

Discussion

The current study clinically compared the efficiency of hormonal based treatments and double insemination to improve the conception rate in both the cows and heifers with metestrual bleeding. The results of our study clarified that the use of hormonal treatments such as CIDR, GnRH, and PGF2α were effective to improve the conception rate in the

Table 1. Conception rate (%), follicular diameter (cm; Mean ±SD) and diameter of the corpus luteum (CL) (cm; Mean ±SD) in different treated groups; Control (Ctrl), Double Insemination, PGF2α, GnRH, and CIDR group.

Group/ Parameter	Conception rate (%)	Follicular Diameter (cm; Mean ±SD)	Diameter of CL (cm; Mean ±SD)
Control (Ctrl; n=15)	13.3	1.56 ± 0.47	2.33 ± 0.31
Double Insemination (n=15)	33.3	1.58 ± 0.50	2.35 ± 0.30
PGF2α (n = 30)	66.6	1.54 ± 0.39	2.54 ± 0.20
GnRH (n = 28)	71	1.49 ± 0.29	2.43 ± 0.31
CIDR (n = 40)	87.5	1.62 ± 0.36	2.49 ± 0.29
P value	<0.001	0.74	0.08

P value <0.05 is statistically significant, SD: standard deviation.

Table 2. Estrogen concentrations (E2 ng/ml; Mean ±SD) at day 0, day 3, day 6, and day 18 in different treated groups; Control (Ctrl), Double Insemination, PGF2α, GnRH, and CIDR group.

Group/ Parameter	E2 Day 0 (ng/ml)	E2 Day 3 (ng/ml)	E2 Day 6 (ng/ml)	E2 Day 18 (ng/ml)
Control (Ctrl; n=15)	20.23±0.06	10.61±0.17	13.64±0.59	17.15±0.84
Double Insemination (n=15)	20.23±0.07	10.62±0.16	13.67±0.60	17.19±0.83
PGF2α (n = 30)	20.23±0.02	11.34±0.30	14.26±0.45	17.43±0.82
GnRH (n = 28)	20.23±0.05	11.40±0.47	14.54±0.70	17.74±0.85
CIDR (n = 40)	20.24±0.08	11.65±0.52	14.70±0.66	18.00±0.76
P value	0.94	<0.001	<0.001	<0.001

P value <0.05 is statistically significant, SD: standard deviation.

Table 3. Progesterone concentrations (P4 ng/ml; Mean ±SD) at day 0, day 3, day 6, and day 18 in different treated groups; Control (Ctrl), Double Insemination, PGF2α, GnRH, and CIDR group.

Group/ Parameter	P4 Day 0 (ng/ml)	P4 Day 3 (ng/ml)	P4 Day 6 (ng/ml)	P4 Day 18 (ng/ml)
Control (Ctrl; n=15)	0.43±0.01	1.71±0.32	5.11±0.45	8.24±0.74
Double Insemination (n=15)	0.43±0.01	1.71±0.31	5.12±0.45	8.27±0.72
PGF2α (n = 30)	0.43±0.09	1.86±0.38	5.25±0.57	8.49±1.00
GnRH (n = 28)	0.42±0.02	1.92±0.36	5.45±0.44	8.70±0.71
CIDR (n = 40)	0.43±0.04	2.00±0.48	5.61±0.55	8.94±0.74
P value	0.60	0.06	0.00	0.01

P value <0.05 is statistically significant, SD: standard deviation.

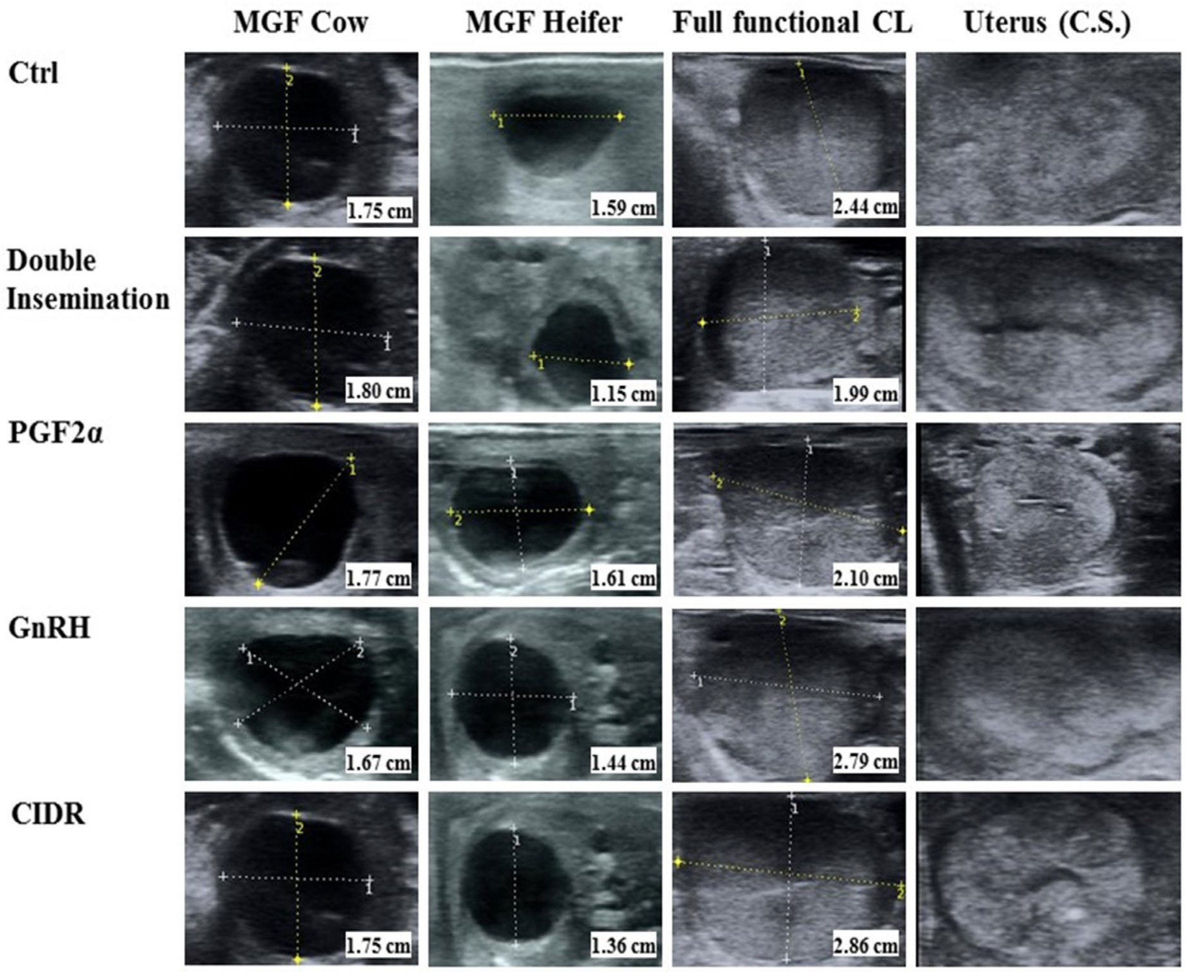

Fig. 2. Representative ultrasonographic images of the mature graffian follicle in cows (MGF; left panel), MGF in heifers (left-middle panel), full functional CL (right-middle panel), and cross section of the uterus (right panel) in different treated groups; Control (Ctrl), Double Insemination, PGF2α, GnRH, and CIDR group.

treated animals with conception rates of 87.5 %, 71.0 %, and 66.6% respectively. And the use of CIDR- PGF2α- GnRH with timed insemination after the GnRH injection would be more superior than other treatments. It was previously reported that the P4-treated animal, at day 3, showed higher conception rate in case of repeat breeding with metestrual bleeding (Sharma, 2008). Similar results of higher conception rate have been reported in PGF2α-treated repeat breeder animals with history of metestrual bleeding. Interestingly, our study reported cessation of metestrual bleeding in the induced estrus after CIDR and PGF2α treatments, as previously reported (Chako, 2003; Juhani, 2003).

The current study showed a lower conception rate in double insemination group (33.3%) and untreated control group (13.3%). Lower conception rate (34.2%) was previously reported without any treatment in cows and heifers with metestrual bleeding (Sharma, 2008). Much lower conception rates in untreated repeat breeder cows with metestrual bleeding were previously reported with conception rates of 25.80%, 20.68% (Quayam and Austin, 1983; Kishore, 2006). Moreover, negligible conception rate was reported untreated repeat breeder cows with metestrual bleeding (Kutty, 2005). On the other hand, double insemination would be effective only in those cases where signs estrus persisted for about 24 hours after insemination (Fleischmann, 1990; Sharma *et al*., 2002). However, in the present study, double insemination did not improve the conception rate. This is likely because of normal estrus phase duration in the treated animals. A combined protocol of GnRH analogue injection with double insemination was effective to improve pregnancy rate in repeat breeder dairy cows (Hasan *et al*., 2021).

The current study measured the MGF and CL as the most significant ovarian structures (Macmillan *et al*., 2018). Although, no differences were detected neither in the size and contour of the ovaries, ovarian structures, nor the uterine horns in all treated groups. The CIDR treated group showed higher levels of the measured sex steroid hormones especially at day 6 and day 18. Absence of differences in the measured diameters of dominant follicle and CL between different treated groups was reported (Lewis *et al*., 1990).

Higher P4 concentrations detected in the present study at day 6 and day 18 is a possible explanation of the higher conception rate reported in the same treated group reported by our study. It should be noted that, an association between metestrual bleeding and deficiency of P4 had been previously reported (Duchens *et al*., 1995; Muller, 1999). Moreover, maintenance of appropriate P4 levels is critical to support pregnancy. Thus, inadequate P4 levels, especially during early gestational period cause early embryonic death and repeat breeding (Lamming *et al*., 1989). In addition, P4 concentration during estrous cycle prior to insemination is a possible indicator of subsequent fertility of cow (Fonesca *et al*., 1983).

The high conception rate detected in the GnRH treated group, in the present study, would be likely because of the increase in the P4 concentrations especially at day 6 and day 18. It was suggested that GnRH injection can enhance the function of an existing CL (Robertson *et al*., 1993) and thus increasing the P4 concentrations (Hansel *et al*., 1991). In line with these findings, the repeat breeding heifers had less luteal tissue, as previously reported (Albhin, 1991). Moreover, it has been suggested that GnRH may act through its anti-luteolytic mechanism not only its luetotrophic mechanism to maintain high P4 concentrations particularly during the critical period of maternal recognition (Saratsis *et al*., 1998).

Conclusion

This study concluded that hormonal treatment using PGF2α, GnRH, and CIDR would be clinically effective to improve the conception rate in cows and heifers with metestrual bleeding with superior efficacy of CIDR protocol for 7 days with PGF2α at day 6 and GnRH after CIDR removal by 48 hours. Such higher conception rate is likely related to the maintenance of higher P4 levels in the treated cows and heifers.

Acknowledgments

Authors would like to appreciate their deep thanks to staff members of Department of Theriogenology, Faculty of Veterinary Medicine, Minia University, Egypt for their help and support.

Conflict of interest

The authors have no conflict of interest to declare.

References

Albhin, A., 1991. Standing oestrus, ovarian function and early pregnancy in virgin and repeat breeder heifer. J. Vet. Med. 38, 212-221.
Chako, L., 2003. Metoestrual bleeding and its effect on fertility in natural and induced oestrus in cattle. M.V.Sc. thesis. Faculty of Veterinary and Animal Sciences Kerala Agricultural University, Thrrissur, India.
Consentini, C.E., Wiltbank, M.C., Sartori, R., 2021. Factors that optimize reproductive efficiency in dairy herds with an emphasis on timed artificial insemination programs. Animals (Basel). 11, 301.
Dellinger, A.B., Bobbett, J.J., Olivier, D.F., Ellett, C.D., 2008. Measuring teachers' self-efficacy beliefs: Development and use of the TEBS-Self. Teaching and Teacher Edu. 24, 751–766.
Duchens, M., Maciel, M., Gustafsson, H., Forsberg, M., Rodriguez, M., Edqvist, L., 1995. Influence of periestrus superabasal progesterone level on cycle length, oestrous behaviour and ovulation in heifers. Anim. Reprod. Sci. 37, 95-108.
Filant, J., Spencer, T.E., 2014. Uterine glands: biological roles in conceptus implantation, uterine receptivity, and decidualization. Intern. J. Develop. Biol. 58, 107.
Fleischmann, K., 1990. Double insemination is better. It improves the conception rate of cows with prolonged estrus or delayed ovulation. Tierzuchter. 42, 346-347.
Fonesca, F.A., Britt, J.H., McDaniel, B.T., Wilk, J.C., Rakes, A.H., 1983. Reproductive tracts of Holstein and Jerseys: Effect of age, milk yield and clinical abnormalities on involution of cervix and uterus, ovulation, oestrous cycles, detection of estrus, conception rate and days open. J. Dairy Sci. 66, 1128-1149.
Galina, C.S., Geffroy, M., 2023. Dual-Purpose Cattle Raised in Tropical Conditions: What Are Their Shortcomings in Sound Productive and Reproductive Function? Animals 13, 2224.
Giri, S.C. Yadav, B.P.S., 2000. Management for optimizing production from livestock. XVIth Annual Concention and National Symposium on Reproduction. Nov. 6-8. pp. 91-92
Hansel, W., Alila, H., Dowd, J., Milvae, R.A., 1991. Differential origin and control mechanisms in small and large bovine luteal cells. J. Reprod. Fertil. Supp. 43, 77-89.
Hasan, I., Hasan M.M., Mohanta, R.C., Miah, M.A., Rahman, M., Rahman, M.S., Juyena, N.S. 2021. Double-timed artificial insemination along with gonadorelin acetate injection improved the fertility in repeat breeder cows. Intern. J. Natur. Sci. 11, 29-39.
Hassaneen, A.S., Rawy, M.S., Yamanokuchi, E., Elgendy, O., Kawano, T., Wakitani, S., Kitahara, G., Osawa, T., 2023. Use of platelet lysate for in-vitro embryo production and treatment of repeat breeding in cows. Theriogenology 210, 199-206.
Juhani, T., 2003. Ovarian function in dairy cattle after gonadotropin-releasing hormone treatments during periestrus. Academic dissertation. Fac. Vet. Med., Univ. Helsinki, Helsinki, Finland.
Kishore, N., 2006. Studies on clinical efficacy of some therapeutic regimen(s) on the conception rate in repeat breeder cows showing metestrual bleeding. M.V.Scs thesis, CSK Himachal Pradesh Krishi Vishvavidyalya, Palampur.
Kishore, N., Singh, M., Kumar, P., Vasishta, N.K., 2005. Effect of progesterone administration on conception rate in cattle showing metestrual bleeding. XXI Annual Convention and National Symposium on Recent Trends and Innovations in Animal Reproduction. 23-25, 2005. pp. 112.
Kutty, C.I., 2005. Investigations on metestrual bleeding in repeat breeding crossbred cattle bred through artificial insemination. XXI Annual Convention and National Symposium on Recent Trends and Innovations in Animal Reproduction. pp. 74.
Lamming, G.E., Darwash, A.O., Back, H.L., 1989. Corpus luteum function in dairy cows and embryo mortality. J. Reprod. Fertil. Suppl. 37, 245-252.
Larson, R.L., Randle, R.F., 2008. The bovine estrous cycle and synchronization of estrus. Colleage of Veterinary Medicine K-State University. Retrived on 2025 March 2nd, https://www.vet.k-state.edu/academics/student-faculty-handbook/studentorgs/aadpDocs/Estrous_Cycle_physiology1.pdf
Lewis, G.S., Coldwell, D.W., Rexroad, J.R., 1990. Effect of gonadotropin hormone and human chorionic gonadotropin on pregnancy rate in dairy cattle. J. Dairy Sci. 73, 66-72.
Macmillan, K., Kastelic, J.P., Colazo, M.G., 2018. Update on multiple ovulations in dairy cattle. Animals 8, 62.
Marnikov, T., Vesilev, G., 1986. Experiment to increase the conception rate in cows with disordered ovulation and spontaneous estrus. Veterinarno-Meditsinki Nauki. 23, 72-80.
Mičiaková, M., Strapák, P., Szencziová, I., Strapáková, E., 2018. Several methods of estrus detection in cattle dams: a review. Acta Universitatis Agriculturae et Silviculturae Mendelianae Brunensis 66.
Muller, K., 1990. Post Oestrual bleeding in cattle. Tierarztliche Umschau. 54, 690-691.
Perera, B., 2011. Reproductive cycles of buffalo. Anim. Reprod. Sci. 124, 194-199.
Pérez-Marín, C.C., Quintela, L.A., 2023. Current insights in the repeat breeder cow syndrome. Animals 13, 2187.
Quayam, S.A. Austin, A.J., 1983. Influence of metoestrous bleeding on conception rate in cattle. Cheiron. 12, 281-282.
Raven, R.W., Weber, F.P., Price, L.W., 1948. The necrobiotic nodules of rheumatoid arthritis: Case in which the scalp, abdominal wall (involving striped muscle), larynx, pericardium (involving myocardium), pleurae (involving lungs), and peritoneum were affected. Annals Rheum. Dis. 7, 63.
Robertson, L., Salaheddine, M., Harvey, M., 1993. Ultrasound monitoring of ovarian changes in cattle after injection of Buserelin 11-13 days post insemination. AVTRW 1993. 1.
Saratsis, P., Schmidt, A., Dampoulou, B., Ypsilantis, P., Brozos, C., Demertzis, A., 1998. Effect of Buserelin on corpus luteum activity and the fertility of dairy cows. Bulletin Hellenic Vet. Med. Soc. 49, 34-38.
Sharma, A., Singh, M.M., Vasishta, N.K., Sood, P., 2002. Effect of gonadotropin releasing hormone administration on conception rate following artificial insemination in cattle. Proceeding of XVIII Annual Convention and National Symposium on Reproductive Technologies for Augmentation of Fertility in Livestock. pp. 14-16.
Sharma, V., 2008. Studies on the effect of supplementation of certain hormones on conception in cows showing metestrual bleeding. M.V.Cs. thesis, CSK Himachal Pradesh Krishi Vishva Vidyalaya Palampur, India.
Sugiura, T., Akiyoshi, S., Inoue, F., Yanagawa, Y., Moriyoshi, M., Tajima, M., Katagiri, S., 2018. Relationship between bovine endometrial thickness and plasma progesterone and estradiol concentrations in natural and induced estrus. J. Reprod. Develop. 64, 135–143.
Temesgen, M.Y., Assen, A.A., Gizaw, T.T., Minalu, B.A., Mersha, A.Y., 2022. Factors affecting calving to conception interval (days open) in dairy cows located at Dessie and Kombolcha towns, Ethiopia. PloS One 17, e0264029.