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Reproductive Performance of Swamp Buffalo with Various Dosages of Gonadotropin-Releasing Hormone (GnRH)

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ABSTRACT

This study aims to determine the reproductive performance of Swamp buffalo with various dosage of gonadotropin-releasing hormone (GnRH). The study subjects were Swamp buffaloes (n=16) with an average age of about 5-6 years, a normal estrus cycle, non-pregnant, and had no history of reproductive disease. Estrus synchronization was performed with an injection of 5mL PGF2α hormone. GnRH was injected on the 8th day with various doses (A=without GnRH injection, B=200mg/2mL, C=300mg/3mL, and D=400mg/4mL). Variables observed were superovulation response, the success of artificial insemination, and speed and intensity of onset of estrus in Swamp buffalo. The results showed that the average superovulation response was 81.25%. The success rate of AI was on average 75% with the highest success rate (100%) with C treatment (300g GnRH + 5mL PGF2α). Swamp buffaloes that have been treated have onset of estrus ranging from 72-81hrs and high intensity of estrus. The administration of GnRH 300mg (group C) and 400mg (group D) gave the best superovulation response (100%) and a 100% pregnancy with the administration of 300mg of GnRH (group C). Administration of GnRH 200mg (group B) showed the fastest estrus onset and all treated buffaloes showed high estrus intensity with a score of +++.

Key words: Swamp Buffalo, GnRH, PGF2α, Reproductive Performance

INTRODUCTION

One of the diversity of livestock in Indonesia is buffalo which has many benefits. In addition to producing milk and meat, buffalo are widely used as carriers and plowing fields in Indonesia. Most of the buffaloes as much as 95% in Indonesia belong to the mud or swamp buffalo clump (Swamp buffalo), while the remaining 5% belong to the river buffalo clump (River buffalo) which is mostly kept in North Sumatra, Indonesia.

In general, buffaloes have symptoms of silent heat, which causes the productivity of buffalo to be hampered due to the length of sexual maturity and long calving intervals. Silent heat in buffalo causes buffaloes not bred on time because the breeder does not know the buffalo is in heat (Putro 1991; Nava-Trujillo et al. 2020; Anil et al. 2021). Reproduction in livestock is influenced by several factors, one of which is hormonal factors. Injection of the hormones like, progesterone and prostaglandin (PGF2α) in buffalo is one way to overcome the difficulty of estrus detection. Corpus luteum (CL) starts to regress the day after the PGF2α injection and diminished on the third day (Melia et al. 2013).

The population of buffalo in Lima Puluh Kota Regency from 2018 to 2020 was 12,327, 12,329, and 9,992 heads, respectively. Based on the biological and economic potential to be developed, the development and distribution of buffalo can be carried out in rural areas (Bettencourt et al. 2015). Buffalo that is often encountered and widely known in rural areas, is the type of mud buffalo (Swamp buffalo). Many of these mud buffaloes are kept with traditional rearing systems in Indonesia. Buffaloes have some unique characteristics such as delayed puberty, a low sign of heat, asymmetrical estrus cycle, silent heat, seasonal breeder, poor conception, and extended calving intervals (Rashid et al. 2019; Devkota et al. 2023).

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One of the efforts to develop and spread the buffalo population can be done through artificial insemination. The implementation of artificial insemination in buffaloes is hampered due to the difficulty of estrus detection as buffaloes have silent heat or calm estrus (Mohammed 2018). Extensive rearing systems that are often carried out by farmers and the habits of wallowing buffalo, make it difficult to detect estrus (Yendraliza et al. 2019). This study aims to determine the response of buffaloes after the administration of Gonadotropin Realizing Hormone (GnRH). This research is expected to be a source of information on the effect of giving various levels of hormones to buffaloes.

MATERIALS AND METHODS

Experimental Animals

This study used 16 Swamp buffaloes having an average body weight of 500kg and in their 2^{nd} lactation. The average age was around 5-6 years with a normal estrus cycle and no history of reproductive diseases.

Superovulation

Rectal palpation was done to ensure that the buffaloes to be treated were non-pregnant. Then 5mL PGF2α hormone was injected to synchronize buffalo estrus. GnRH injected on the 8th day with different doses (A=without GnRH injection, B=200mg/2mL, C=300mg/3mL, and D=400mg/4mL) with 2 doses i.e., in the morning and evening. Then 15mL PGF2α was injected intra-muscularly on 11th day.

Artificial Insemination (AI)

Each donor buffalo received AI 12 hours after showing signs of estrus. AI was carried out 3 times, in the morning and evening on Ist day and the morning on 2^{nd} day. Buffaloes were placed in clamped cages to facilitate the AI process and 1 straw dose was injected per donor.

Research Parameters

The variables in this study were the response rate of buffaloes after giving various doses of GnRH as seen from the number of CLs present on ovary. Donor buffaloes were considered to respond if they have a CL of more than one. Then the success of artificial insemination, the speed of onset of estrus, and the intensity of estrus in buffalo were studied.

Data Analysis

Data thus collected for various parameters were subjected to statistical analysis Chi-square using Minitab statistical program.

RESULTS AND DISCUSSION

Superovulation Response

A total of 13 out of 16 (81.25%) donor buffaloes showed a positive response to superovulation, while 3 donor buffaloes didn't respond to the treatment (Table 1). Groups C (300g GnRH+5mL PGF2α) and D (400g GnRH+5mL PGF2α) treatments showed the highest superovulation response with a superovulation response rate of 100%. In group B, superovulation response was 75%. The results showed non-significantly (P=0.799) differences in superovulation response between buffaloes given different treatments. According to Afriani et al. (2014), follicle maturation takes place effectively stimulated by the injection of GnRH. A low dose of GnRH leads to luteolysis without ovulation while a high dose of GnRH renders ovulation and a lot of CLs are formed (Noakes et al. 2001).

Afriani et al. (2020) stated that buffalo given different doses of follicular stimulating hormone (FSH) had a super-ovulatory response with a response rate of 56.25%. Different levels of super-ovulatory response caused by internal factors (genetic, nutrition, and reproductive health) and external factors (use of FSH preparations, seasonal dose, and field management) (Silva et al. 2009). Superovulation response can decrease due to inadequate livestock nutrition and reproductive organ disorders (Mikkola et al. 2019). However, According to Presicce et al. (2022), the season did not affect pregnancy rates when the results of the two treatments (two protocols for synchronization of ovulation: (i) Ovsynch (OV) and (ii) progesterone) were pooled together with regard to the first synchronization protocol, followed by AI.

AI Success Rate

Artificial insemination in buffalo gave different results for each treatment. The average pregnancy rate in this study was 75%. The success results of AI of buffalo given 200, 300 and 400mg GnRH and 5mL of PGF2α showed 75, 100 and 75%, respectively with non-significant (P=0.855) difference (Table 2). Treatment C (300mg GnRH+5mL PGF2α) showed the highest (100%) pregnancy rate. Meanwhile, the lowest pregnancy rate was 50% in the A treatment (control) which was not given GnRH and PGF2α. Differences in pregnancy rates at each dose level of GnRH can be caused by various possibilities such as different livestock conditions, semen quality and handling, and also artificial insemination time. In this regard, Irikura et al. (2003) stated that semen deposition during AI affects pregnancy rates. Factors that affect the condition of buffalo that are not pregnant are individual buffaloes, estrus condition, quality of spermatozoa after thawing, and time of insemination (Berdugo et al. 2007).

Table 1: Percentage of donor response to superovulation

Treatments	Donor	Donor Response	Donor Not Responding	
			No.	Response Rate (%)
A (Control)	4	2	2	50
B (200mg GnRH + 5mL PGF2α)	4	3	1	75
C (300mg GnRH + 5mL PGF2α)	4	4	0	100
D (400mg GnRH + 5mL PGF2α)	4	4	0	100
Total	16	13	3	81.25

Chi-square value=1.009, df=3, P-Value=0.799

The 100% pregnancy rate in C treatment (300mg GnRH+5mL PGF2α) probably resulted in 100% clear signs of estrus. The appearance of a clear sign of lust can make it easier to predict the exact time of AI (Baruselli et al. 2003). Irikura et al. (2003) stated that the administration of GnRH combined with PGF2α in heifers resulted in a 100% pregnancy rate indicating 100% estrus. The combination of GnRH and PG2α gives a clear appearance of estrus and progesterone levels (Roza et al. 2019).

Onset of Estrus

The onset of estrus in this study after injection of GnRH with different doses and combined with PGF2α (5mL) ranged from 72-81 hours (Fig. 1). The results of statistical analysis showed that the administration of GnRH had non-significant (P>0.05) effect on the rate of onset of estrus. The fastest estrus occurred with the treatment B (200mg GnRH+5mL PGF2α) which is 72.5h while the longest estrus seen in control group (without treatment). The injection of one dose of GnRH in buffalo can improve the estrus cycle and the injection of two doses of GnRH when combined with PGF2α usually accelerate the onset of estrus in buffalo (Berber et al. 2002). Gunawan et al. (2020) reported that the combined use of the GnRH+PGF2α in Swamp buffalo resulted in an estrus rate of 53.67h. In line with the findings of Yendraliza et al. (2012) who stated that the rate of postpartum buffalo estrus in Kampar Regency with a dose of GnRH which synchronized with 12.5mg of PGF2α in a row in hours was 52±6, 53.88±5.1, 27.8±2.5, 28.8±0.5 and 30±1.9. While Afriani et al. (2020) conducted a study of giving different doses of the FSH (16, 18, 20, and 22mL) combined with 500mg GnRH in buffalo to produce the fastest estrus, namely the injection of 16mL of FSH. The synchronization of estrus and ovulation are the key strategies that have been attempted to improve fertility in buffaloes (Ahmad and Arshad 2020).

The onset of estrus in livestock can be influenced by many factors, one of which is the rearing system. The traditional rearing system with a grazing system that does not meet the need for feed, naturally will disrupt estrus cycle in livestock. This is following the opinion of Reith and Hoy (2018), who stated that the factors that affect the timing of the onset of estrus in livestock include poor feed conditions and traditional rearing systems that are lacking feed. One of the important factors that can affect reproductive function is the nutrition (Nurfathaya et al. 2019). A combined strategy of improvement in environment, nutrition and management is pre-requisite for hormonal manipulation in order to improve productivity of buffaloes during the summer season (Vijayalakshmy et al. 2020; Abulaiti et al. 2022; Gallab et al. 2022).

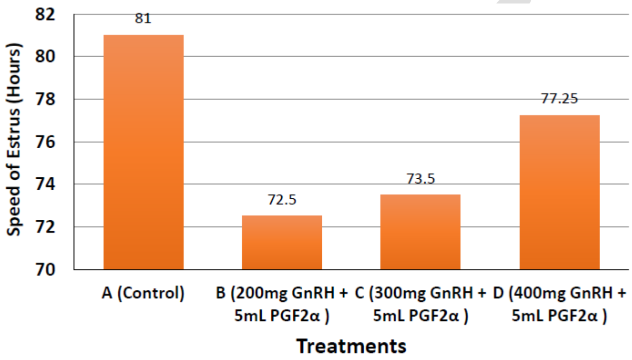

Fig. 1: The average percentage onset of estrus in buffalo with the treatment of GnRH and PGF2α.

Intensity of Estrus

The results in the present study showed that treated with GnRH in combination with PGF2α buffalo had higher estrus intensity compared to buffaloes not received GnRH (Table 3). This high intensity of estrus is characterized by swollen vulva, mucus discharge, red color, warmth, and silence when climbed by other livestock which are all symptoms of estrus (Hafid et al. 2021). In the present study P0 (without treatment) buffaloes showed a lower intensity of estrus. This is in line with the research results of Yendraliza et al. (2012), they stated that the combination of the hormone GnRH with PGF2α produces the highest intensity of estrus. Following the opinion of Rhodes et al. (2003), estrus in buffalo is much clearer due to the use of GnRH along with PGF2α.

Table 2: Artificial insemination results in Swamp buffaloes treated with various doses of GnRH

Treatments	Donor	Number of Pregnancies	Pregnancy Rate (%)
A (Kontrol)	4	2	50
B (200mg GnRH + 5mL PGF2α)	4	3	75
C (300mg GnRH + 5mL PGF2α)	4	4	100
D (400mg GnRH + 5mL PGF2α)	4	3	75
Total	16	12	75

Chi-square value=0.778, df=3, P-Value=0.855.

Table 3: The intensity of estrus in buffalo.

Treatments	Score	Amount	Intensity of Estrus	Description
A (Control)	++	2	Medium	The vulva is swollen, red, and warm.
B (200mg GnRH + 5mL PGF2α)	+++	3	High	The vulva is swollen, red, warm, mucus out, and silent when climbed.
C (300 mg GnRH + 5mL PGF2α)	+++	3	High	The vulva is swollen, red, warm, mucus out, and silent when climbed.
D (400 mg GnRH + 5mL PGF2α)	+++	3	High	The vulva is swollen, red, warm, mucus out, and silent when climbed.

Conclusion

It was concluded that the administration of GnRH 300mg (group C) and 400mg (group D) gave the best superovulation response (100%) and a 100% pregnancy rate at the administration of 300mg of GnRH (group C). Administration of GnRH 200mg (group B) showed the fastest onset of estrus and all treated buffaloes showed high estrus intensity with a score of +++.

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Author's Contribution

All authors contributed equally to this work.

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