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by Yendraliza Yendraliza

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Comparison of reproductive performance of Bali cows using different duration laser punctures and PGF2α hormone

Yendraliza[1*], Rahmi Febriyanti[1], Widia Setuti[1], Zumarni[1], Oksana[1], Rosmaina[1], Sudirman M Johan[2] and Chairusyuhur Arman[3]

[1]Faculty of Agriculture and Animal Science, Universitas Islam Negeri Sultan Syarif Kasim Riau, Jalan HR Soebrantas KM 15 Panam Pekanbaru, Riau, Indonesia.

[2]Faculty of Syariah dan Ilmu Hukum, Universitas Islam Negeri Sultan Syarif Kasim Riau, Jalan HR Soebrantas KM 15 Panam Pekanbaru, Riau, Indonesia

[3]Faculty of Animal Science, Mataram University, Jalan Majapahit 62 Mataram, West Nusa Tenggara, Indonesia.

*Corresponding author:yendraliza@uin-suska.ac.id

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Abstract

A study was conducted to determine the estrus response and pregnancy percentage of Bali cows after different laser-puncture treatments with PGF2α hormone. One hundred fifty Bali cows that had previously been pregnant were used in this study. They were divided into three groups based on body condition score of 3, 3.5, and 4, into 3 treatments: 1) injected with the PGF2α hormone, 2) laser puncture for 5 sec and 3) laser puncture for 10 sec. The laser used was a soft Helium-Neon 50 Hz of 220 volt and 50 watt which obtained 20 MW of power. The parameters measured were estrus percentage, estrus duration, onset of estrus, estrus intensity, total pregnancy, non-return rate, and services per conception (S/C). The data were collected for four days after the experiment and analyzed using Chi-Square Independence Test. The results revealed that the estrus response and pregnancy percentage of the Bali cows after synchronization with PGF2α hormone (Treatment 1, Control) and laser-puncture for 5 sec and 10 sec were significantly different ($p<0.05$). The use of laser-puncture in Bali cows had increased pregnancy rate from 60 to 80%, with S/C of 1.6 -2.0, decreased non return rate 20 to 40%, with estrus intensity of 1.6 to 2.0. The use of 5-sec laser puncture has produced the similar reproductive performance as synchronizing the cows with PGF2α hormone.

Keywords: Bali cattle, services per conception, pregnancy rate, estrus synchronization

Introduction

Bali cattle are local Indonesian cattle that have adapted with village farmers across Indonesia, and they are adaptable to marginal land conditions (Zulkharnaim et al., 2010). Bali cattle are well known for their high fertility rate (Purwantara et al., 2012) and high-quality meat with a low fat content (Warmadewi et al., 2017). The high fertility rate is not associated with rapid cattle reproduction, as indicated by the long calving interval. Therefore, estrus induction is required to shorten the calving interval. Estrus synchronization that is often done in cows uses PGF2α hormone. This hormone is not sold freely and its use also requires paramedics to handle it. Estrus induction can be applied by laser technology, i.e., laser (light amplification by stimulated emission of radiation) puncture, at acupuncture points

(Fattah, 2015). Laser-puncture is a non-aggressive treatment that it is painless and has a longer lasting effect (Kreisel and Weber, 2016). Lin et al. (2002) reported that acupuncture was effective in improving the function of reproductive organs after repeated breeding (77.7%) on dairy-cows in comparison with post-treatment with gonadotropin-releasing hormone (GnRH). The stimulation of certain organ points is related to reproductive function and can significantly affect release of sexual hormones, i.e., luteinizing hormone (LH), follicle-stimulating hormone (FSH), estradiol, and progesterone, in the plasma. Acupuncture is able to stimulate the ovarium in steroidogenesis and affects the functions of the paracrine and autocrine pathways. Laser technology can be applied to the biological manipulation of animal husbandry (Schoen, 2001). Laser puncture has been indicated to stimulate estrus in cows in Jembrana, Bali, Indonesia, obtaining an estrus percentage of 90.6% (Bintara, 2010). The estrus induction of Bligon goats using laser puncture obtained 85.7% estrus percentage (Fattah, 2015). Laser acupuncture has been effective in the enhancement of sperm quality (Behtaj and Weber, 2019) and decreasing Sertoli and Leydiq cells of the male Mojosari ducks (Adikara et al., 2017). Previous studies on puncture points in bulls and goats have found that they were similar, but the points were slightly different. However, there is a lack of research conducted on laser puncture applied to cattle. Therefore, the success of laser puncture in cattle needs to be further studied. This study was conducted to determine the estrus response and pregnancy percentage of Bali cows after laser puncture treatment.

Materials and Methods

Location of study

The study was conducted in several villages in IndraGiri Hulu Regency, Riau Province, Indonesia. This district has the most Bali cattle in the Riau Province with an area of 8,198.26 km^2 which consists of 77,582 hectares of rubber plantations and 99,792 hectares of oil palm plantations. This regency is sovereign as a plantation area with a altitude of 50 to 100 m above sea level. It is characterized by temperatures around 21.9 °C to 33.4 °C and relative humidity of 51 to 98%.

Experimental animals

One hundred fifty Bali cows (previously pregnant and had given birth) were placed into individual pens and fed with cut native grass 3 times a day and water was provided ad libitum. Complementary feed additives, such as salt licks and concentrate, were also given. The cattle were divided into groups based on body condition score (BCS) of 3, 3.5, and 4, and three treatments: 1) induced by PGF2α (*Capriglandin*®) at a dosage of 2.5 ml, 2) laser puncture for 5 sec, and 3) laser puncture for 10 sec (Figures 1a, 1b and 1c). Biovet (2004) explained that there are six laser-puncture points. The first point is the estrus point (Ming-Meng/estrus), located on the dorsal area of the base of lumbar vertebrae II and III (as general point). Stimulation at this point improves hypophyse activity. The second is the ovarian point, which is located on the dorsal area between the processus transversus and lumbar vertebrae I-II-III. The stimulation of this point improves ovarium activity by enhancing follicle and reproductive hormone formation. The third point is the oviduct point, which is located on the dorsal area between processus transversus and lumbar vertebrae III-IV-V-VI. Stimulation at this point prevents

cystic ovaries and hypo functional ovaries. The next point is the cervical point, which is located at the high end of the os ileum. Stimulation at this point optimizes the cervix and improves the estrus duration by producing estral mucus. The fifth point, the hormonal point, is located on the dorsal base of the sacrococcygeal area. The stimulation of this point will improve prostaglandin hormones. The last point is the vaginal point, which is located on the vulva; two points are located on the right and middle left of the vulva. The laser-puncture was performed using a He-Ne laser gun (632.8 nm) with 50 Hz, 220 volts, 50 watts and transmitted with a cable length of 1.5 m. The second and third treatments were carried out for two days. Parameter measurements for the first treatment were carried out starting from the second day after PGF2α injection until fourth day of the experiment. Parameter measurements for the second and third treatments were conducted on days 3 and 4 after the first laser-puncture induction.

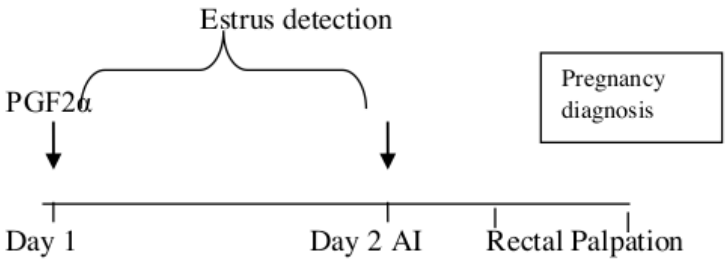

Figure 1a. First Treatment

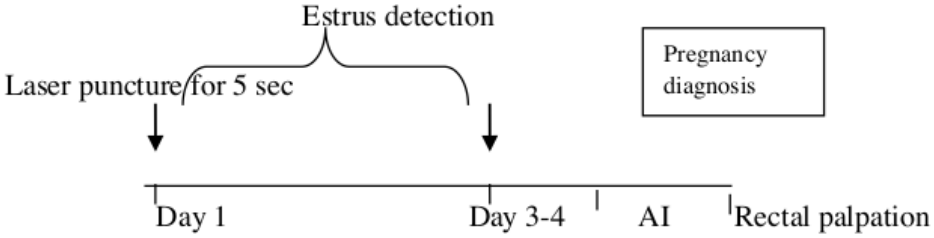

Figure 1b. Second Treatment

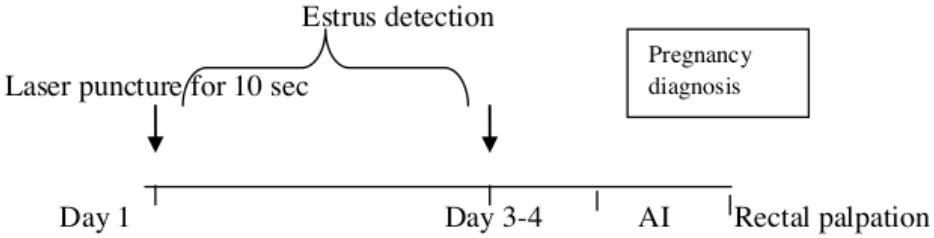

Figure 1c. Third Treatment

Procedure for laser-puncture

The cows were brought to a flat and shady place and the laser-puncture treatment was carried out by attaching the end of the tube for each point for 5 sec for the first treatment and 10 sec for the second treatment. The whole run required 110 sec and was carried out 3 times in one day. Similar procedure was repeated on day 2 until estrus appeared. To measure the quality of estrus arising from the treatment applied a teaser bull was used. All cows were observed continuously for discharge of vaginal mucus and standing to be mounted by another cow which were taken as indications of estrus, with at least 2 - 3 mounts performed.

Parameter measurements

The variables measured were 1) estrus response (%) - the number of cows in estrus divided by the number of cows in the treatment group multiplied by 100, 2) the speed of estrus occurrence (estrus onset, hours) - starting from the third day after treatment until the appearance of estrus, 3) duration of estrus (hours) – taken from the time of estrus symptom appearance until the symptom disappeared, 4) service per conception (S/C) - the number of pregnant animals from the number of times mated, 5) non-return rate (%) - the number of animals that were not subsequently rebred after 21 days post mating and 6) pregnancy rate (%) - the number of cows pregnant divided by the number of cows mated.

Data analysis

Data for all treatments were analyzed for independence using Chi- Square Test (Steel et al. 1991).

Results and Discussion

Estrus intensity

The results on estrus intensity of the Bali cows are presented in Table 1. The estrus intensity of Bali cows treated with 5-sec laser puncture was greater than those cows treated with 10-sec laser puncture. The difference in estrus intensity was due to the different response of livestock to laser puncture. This was because the stimulation at the laser puncture points had activated cells that were associated with the reproductive system and each individual's physiological response was also different (Herdis, 2010). The estrus quality was determined by the estrus intensity value and was defined by the visible estrus signs. This is consistent with Jainudeen and Hafez (2016b) that the estrus intensity value is a cumulative value determined by the appearance of the vulva, abundance of mucus and estrus behavior. In addition, Ferraz et al. (2017) stated that estrus intensity is influenced by estrogen concentration, which generates positive feedback and stimulates luteinizing hormone release. Estrogen is produced by cells that form the follicular wall. Larger follicles produce higher estrogen concentrations. Lyimo et al. (2000) stated that the estrus intensity of cattle was influenced by the estradiol concentration in the body. The estrus intensity of Bali cattle is different from the intensity of cattle in China (62.7%) (Luo and Gu, 2009), the intensity of Bali cattle in Kalimantan (Malik et al., 2018) and Bali cattle in Lampung (Handayani et al., 2014). The difference in estrus intensity between livestock breeds is caused by the differences in the species used in this study, therefore resulting in different responses (Zuo et al., 2016).

Table 1. Percentage of estrus, estrus intensity, onset of estrus, estrus duration, non-return rate, number of services per conception (S/C) and pregnancy rate of Bali cows synchronized and treated with laser puncture

Treatments	n	Estrus %	Estrus intensity	Onset estrus (h)	Duration estrus (h)	Non return rate (%)	S/C	Pregnancy rate (%)
Hormone PGF2α	50	100	1.8[b]	52.2±14.7[c]	13.4±1.6	40[b]	1.67[b]	(30/50) 60[a]
5-sec laser puncture	50	100	1.6[a]	61.6±7.83[a]	13±0.7	20[a]	1.25[a]	(40/50) 80[b]
10-sec laser puncture	50	100	2.0[c]	56±11.73[b]	13.6±1.14	20[a]	1.25[a]	(40/50) 80[b]

[ab] Means within columns with different subscripts differ significantly (P<0.05)

Percentage of estrus

The percentage of estrus in the Bali cows synchronized by PGF2α induction and laser-puncture showed 100% success. The estrus percentage was not significantly different between cows induced by PGF2α and those induced by laser-puncture, which indicates that prostaglandin hormone and laser puncture are effective in inducing estrus. The percentage of Bali cows in estrus in Riau was similar to that of Bali cows in Nusa Tenggara Barat (Mardiansyah et al., 2016) and Bali cattle in Lampung (Handayani et al., 2014). The successful estrus reactions in the Bali cows showed the good fertility of the cows for the experiment. This finding corresponds with the results reported by Arifiantini and Purwantara (2010) that the fertility of Bali cattle was approximately 80%. The estrus percentage of Bali cows treated with laser-puncture in Riau was different from the estrus percentage of Boerawa goats and Peanut goats treated by laser puncture in Bulopoddo of 80-90% (Fattah, 2015) and buffalo in the Jembrana area of 90.6% (Guntoro and Yasa, 2002). The difference in the percentage of estrus was caused by the different responses of each herd of cattle to laser puncture (Schoen, 2001). Furthermore, Lin et al. (2003) stated that the nurture factor greatly determines the effectiveness of laser puncture.

Duration of estrus

The duration of estrus in Bali cattle injected with PGF2α was not significantly different from the duration of estrus induced of laser puncture cows, 13.4 ± 1.67 h for hormone induction, 13 ± 0.71 h for 5-sec laser puncture and 13.6 ± 1.14 h for 10-sec laser puncture (Table 1). The duration of estrus is influenced by the ability of the animal's body to respond to the laser stimuli at the laser puncture points to induce activity of the reproductive organs. Lin et al. (2002) stated that laser puncture treatment can activate cells associated with reproduction, which results in the balance of cells and can lead to estrus. The estrus period of Bali cattle in Riau that were treated with laser-puncture was different from that of Garut sheep of 27.44-27.50 h (Herdis, 2010) and buffalo in Jembrana of 24.30 h (Guntoro and Yasa, 2002). This difference is caused by the difference in livestock species, resulting in different physiological response to laser puncture (Hafez and Hafez, 2016).

Onset of estrus

The onset of estrus in this study was significantly different between Bali cows induced by PGF2α and laser-punctured. The difference in the onset of estrus of Bali cows in Riau using the PGF2α hormone and laser-puncture was due to the different levels of hormones in the bodies of the livestock used, and each body's response is different (Hafez and Hafez, 2016). The 5- and 10-sec laser puncture treatments also showed that the onset estrus was significantly different of 52.2 ± 14.72%, 61.6 ± 7.83%, and 56 ± 11.73 h, respectively. This was due to the cell activation caused by the stimulation of laser puncture points through intercellular bridges in the form of polarization changes, electrical charges, and the occurrence of influx, which stimulate rRNA and mRNA for protein synthesis. This stimulation would result in extra- and intra-cellular activities in the organ (Hafez et al., 2016).

Non-return rate

The non-return rate of Bali cows induced by PGF2α hormone was different from that of Bali-cows treated by laser puncture (Table 1). The laser puncture duration at the reproductive point of Bali cows did not show a difference in the non-return rate. The non-return rate of Bali cows in this experiment was lower than that of Bali cattle in Nusa Tenggara Barat of 60-80% and 80-86.6%, respectively (Mardiansyah et al., 2016). In this regard, Yusuf et al. (2015) stated that the success of insemination is determined by several factors, such as semen deposition, quality of the semen, handling of the semen and the skill of the inseminators. The difference in the non-return rate between hormone-treated Bali cows and laser-puncture treated Bali-cows was due to the difference in the estrus response among the Bali cattle.

Number of services per conception

The number of services per conception (S/C) of the Bali cows was significantly lower at 1.25 for laser-puncture groups compared to 1.67 for hormone group (Table 1). Jainudeen and Hafez (2016a) stated that normal S/C values were approximately 1.6 - 2.0. The S/C values in this study were within the normal range. This result might vary due to the body's response to prostaglandins, which also differs. The PGF2α hormone must be injected at a specific time with an exact dose to obtain the maximum effects (Hafez et al., 2016). Saili et al. (2017) stated that individual responses would affect hormone synchronization. Furthermore, the use of the PGF2α hormone at the beginning or end of the luteal phase would reduce the effectiveness of the hormone. The 5- and 10-sec laser puncture showed that the S/C values were not significantly different. This result corresponds to the findings of Hafez and Hafez (2016) that S/C was affected by the condition and prolificacy of the animals. The S/C of Bali-cows in this study of 1.25 - 1.67 was not different from those cited in Nusa Tenggara Barat at 1.3 - 1.56 (Mardiansyah et al., 2016). It is likely that S/C values are influenced by inseminator's skill, handling procedure, and estus status of the animals.

Percentage of pregnancy

The percentage of pregnancy of Bali cows treated with laser-puncture and PGF2α hormone was significantly different (Table 1), higher for laser-punctured cows compared than cows that received only the PGF2α hormone. This difference was due to the variance of S/C values of Bali cows of the two groups. Percentage of pregnancy found in this study was higher that the dairy cows in China of 80% and 77.7%, respectively (Lin et al., 2002) but lower than those in West Nusa

Tenggara Province, Indonesia of 91.9% - 94.2% (Pribadi et al., 2015). Thus suggesting that laser-puncture could be a simple and effective method to improve pregnancy rate in cows (Zuo et al., 2016).

Conclusion

The use of laser puncture in Bali cows indicates increases in pregnancy rate up to 80% and estrus intensity to 1.6 – 2.0 and a decrease in the number of services per conception to 1.6 and non-return rate to 20%. The use of 5-second laser-puncture produces similar reproductive performance as that of the synchronizing hormone, PGF2α.

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