Reproductive and cost assessment of a seasonal breeding program with *Bos indicus* in tropical Mexico

José F. Martínez1 MVZ; Carlos S. Galina1 Ph.D; Ivette Rubio2 Ph.D; Wendy L Balam2 MVZ; Manuel D Corro2* Ph.D.

1Universidad Nacional Autónoma de México, Facultad de Medicina Veterinaria y Zootecnia, Departamento de Reproducción, Ciudad Universitaria, Ciudad de México, México.
2Universidad Nacional Autónoma de México, Facultad de Medicina Veterinaria y Zootecnia, Centro de Enseñanza, Investigación y Extensión en Ganadería Tropical, Tlapacoyan, Veracruz, México.
*Correspondence: macorro@unam.mx

Received: November 2020; Accepted: March 2021; Published: April 2021.

ABSTRACT

**Objective.** To compare the reproductive performance of postpartum and open *Bos indicus* cows and to study the cost effectiveness of retaining non-pregnant animals after a short breeding season in tropical region of Mexico. **Material and Methods.** A total of 128 *Bos indicus* were included, 87 postpartum cows (PP) with ≤90 days after calving and 41 open cows (OC) with >90 days open. The study was divided into three phases: 1) Estrus synchronization followed by FTAI (day 0-10), 2) Estrus detection and AI (day 11-45) and 3) Natural mating (day 46-90). For the first phase, all animals were synchronized and AI at fixed time (day 10). Cows displaying overt signs of estrus (day 11-45) were AI. Open cows during the previous two phases were exposed to the bull. **Results.** Pregnancy in phase 1 was different (p<0.01) for PP and OC groups, 58.6% and 34.1%, respectively. Overall pregnancy percentage over the second service was 42.5% (p>0.05). No differences (p>0.05) were observed at phase 3, average 44.2%. By the end of the breeding season, the cost of OC, was 3 times more than PP cows. **Conclusions.** Pregnancy rate at first phase was higher in PP cows than OC cows. At the end of breeding season, a pregnancy rate of 80% was found. Incorporation of open cows from previous breeding season was more expensive than PP cows in all phases of the breeding program. Retaining an open cow for rebreeding one year or more could not be economically feasible.

Keywords: Artificial insemination; Bos indicus; cost analysis; postpartum (Source CAB)

RESUMEN

**Objetivo.** Comparar el desempeño reproductivo de vacas *Bos indicus* posparto y abiertas, así como evaluar el costo de retener vacas vacías al final de una temporada de empadre en el trópico mexicano. **Material y métodos.** Se incluyeron 128 vacas *Bos indicus*, 87 vacas posparto (PP) con ≤90 días posparto y 41 vacas abiertas (OC) con >90 días abiertos. El estudio se dividió en tres fases: 1) Sincronización de celos (día 0-10) e inseminación a tiempo fijo (FTAI), 2) Detección de celos e IA (día...
Para la primera fase, todos los animales fueron sincronizados inseminados FTAI (día 10). Las vacas que mostraban signos de estro (día 11-45) fueron inseminadas (IA). Las vacas abiertas durante las dos fases anteriores fueron expuestas al toro. **Resultados.** La tasa de gestación en la fase 1 fue 58.6 y 34.1% (p<0.01), para PP y OC, respectivamente. Durante la fase 2, el porcentaje de gestación fue 42.5% (p>0.05), mientras que en la fase 3, la tasa de preñez fue 44.2% (p>0.05). El costo de una vaca OC fue tres veces más que las vacas PP. **Conclusiones.** La tasa de preñez durante la primera etapa, de la estación reproductiva, fue mayor para vacas PP que vacas OC. Al final de la estación reproductiva la tasa de gestación fue 80 %. El costo beneficio de retener animales no preñados después de una corta temporada de empadre no es económicamente factible para una unidad de producción vaca-becerro.

**Palabras clave:** Análisis de costos; Bos indicus; inseminación artificial; posparto (Fuente CAB)

**INTRODUCTION**

Countries in the tropics have a growing interest in implementing Artificial Insemination (AI) programs using estrus detection followed by natural mating in combination with a short breeding season. A variety of factors may influence the success of AI, accentuated by the brief duration of estrus and a lack of full commitment to the program (1,2). Consequently, insemination by appointment, also known as fixed time AI (FTAI) has become a popular alternative. Several studies (3,4) have shown encouraging results in protocols incorporating various hormonal combinations with the added advantage that they can be used in both cycling and anestrous animals.

Regardless of the breeding program utilized in the farm, the economic success of beef herds depends on having good pregnancy rates in a short breeding season. As a result, a compact calving season is an essential component of reproductive management of suckled beef animals. Cows calving early in the season will have the advantage of a longer recovery period after calving, thus improving their chances for a gestation in the subsequent breeding season and their likelihood of avoiding culling (5). In different experiments (6,7) approximately 20% of cows failed to become pregnant in a short breeding season. Furthermore, Stagg et al (8) and Sinclair et al (9) reported that about 15% of cows failed to respond to the removal of the suckling/maternal calf bond which typically had prolonged postpartum anestrous intervals often described as deep anestrus. For this reason, the use of natural mating is commonly employed in cows which fail to become pregnant following an AI program (for review see Galina y Orihuela (2). On the other hand, the utilization of bulls for natural mating by itself is no guarantee of good pregnancy rates. Several researchers Chacón (10), Chenoweth and Mc Pherson (11) pointed out that about 15-35% of beef bulls would be classified as unsound or not adequate as a potential breeder, when assessed by Bull Breeding Soundness Evaluation (BBSE). Barth (12), indicated the success of a short breeding season using bulls in beef herds, depend among other factors by identifying potential sub fertile and infertile bulls.

The question then arises what are the economic issues involved in incorporating animals into an AI program that failed to become pregnant the previous year? Besides, how cost effective is the use of natural mating following an AI program? The objective of the present study was to compare the reproductive performance of postpartum and open cows in a 90-day breeding program and to study the cost effectiveness of retaining non-pregnant animals after a short breeding season.

**MATERIAL AND METHODS**

**Location.** The study was conducted at the Centre for Teaching, Research and Extension in Tropical Animal Husbandry belonging to the Faculty of Veterinary Medicine of the National Autonomous University of Mexico, located in the State of Veracruz, Mexico at 20° 04′N and 97° 03′W, with humid tropical climate, mean annual temperature of 24°C and mean annual rainfall of 1742 mm.

Ethical statement. The Animal Care Internal Committee of the Faculty of Veterinary Medicine and Zootechnics of the National Autonomous University of Mexico approved the methods used during the present research in accordance with The Code of Ethics of the World Medical Association (Declaration of Helsinki).
Animals. A total of 128 healthy multiparous Brahman cows diagnosed as non-pregnant by ultrasound were divided into two groups: postpartum cows (PP) with ≤90 days after calving (n=87) and open cows (n=41) (OC) with >90 days open. The average age and calving number were 5 ± 2 and 3 ± 2, respectively. All animals were kept under pasture conditions based on rotational Cynodon nlemfuensis (African star grass), Paspalum spp. y Axonopus pp supplemented with minerals and water ad libitum.

Experimental design. The study comprised three phases: 1) Estrus synchronization followed by FTAI (day 0-10), 2) Estrus detection and AI (day 11-45) and 3) Natural mating (day 46-90). For the first phase, all animals (n=128) were synchronized followed by FTAI (day 10). Then, cows displaying overt signs of estrus (day 11-45) were AI at detected estrus. Finally, all cows diagnosed open from the earlier period, were exposed to a 6-year-old bull of proven fertility (Figure 1).

Reproductive management. Estrus Synchronization and TAI. All cows were synchronized using the synchronization protocol based on the use of a device with 1.9 g of natural progesterone (CIDR 1900 Cattle Insert, Zoetis, Mexico) and the administration of 2 mg IM of estradiol benzoate (Benzoato de Estradiol, Zoetis, Mexico) on the day of insertion (day 0). At CIDR withdrawal (day 8) 400 UI IM of eCG were given (Novormon 5000, Zoetis, Mexico) together with 25 mg IM dinoprost trometamine (Lutalyse, Zoetis, Mexico). This treatment was followed by 1 mg estradiol benzoate (Benzoato de estradiol Zoetis, Zoetis, Mexico) at day 9. FTAI was performed at 56±2 h after CIDR withdrawal.

Estrus detection + IA. Estrus detection was carried out in two periods (06:00 a 08:00 a.m. y 16:00 a 18:00 p.m.). A cow resulted positive when she was seen to accept mounting (13). AI was performed on the system AM-PM.

Ultrasound examinations. Two days before each phase of the breeding program, the presence of a corpus luteum (PCL) was evaluated, with ultrasound (Aloka SSD 500, Tokyo, Japan) using a 7 MHz transductor. Blood samples were taken in the coccygeal vein or artery. Progesterone was measured using an ELISA kit (DRG® Progesterone ELISA, Germany). The presence of a viable corpus luteum was determined when values were above 1ng mL (14). A value of 1, indicated the absence of a corpus luteum and progesterone levels of < 1 ng mL and 2; if a visible corpus luteum was present and progesterone levels were > 1ng mL.

Figure 1. Schematic diagram of the phases of breeding season in postpartum and open cows.

Phase 1: Estrus synchronization protocol performed in both groups at the start of the breeding season with controlled internal drug release (CIDR) with 1.9 g of natural progesterone and 2 mg of estradiol benzoate (EB). At CIDR withdrawal (day 8) 400 UI of equine chorionic gonadotropin (eCG) were given together with 25 mg synthetic prostaglandin (Pgf2α). This was followed by 1 mg EB at day 9. Fixed-time artificial insemination (FTAI) was performed at 56±2 h after CIDR withdrawal. Phase 2: Estrus detection (ED) and artificial insemination (AI) performed at day 11 to day 45. Phase 3: Bull introduced with the females at day 46 and remained until the end of the breeding season at day 90.

Body condition score (BCS). Body condition score was graded on the scale 1 (thin) to 9 (obese) according to the methodology proposed by Nicholson & Butterworth, (15).
Dorsal back fat evaluation (DBF). It was assessed at beginning of the breeding season, using ultrasonic device (Aloka SSD 500, Tokyo, Japan) with a convex transducer of 3.5 MHz frequency. The DBF was measured in the thurl area located midway between the tuber coxae (hooks) and the tuber ischiae (pins), 2–3 cm above the greater trochanter of the femur following immobilization of the animal (16).

Cost analysis. The methodology developed from Torres-Aburto et al (17) was adapted to establish the cost of the calving interval per cow a day ($0.98 USD), opportunity cost per day for an open cow which did not wean a calf ($0.96 USD). The cost for FTAI program per cow was calculated at $35.61 USD, which consist of hormones, materials (AI disposables), semen and labor (phase 1). For phase 2 (35 days), the expenses included labor cost per cow per day ($0.22 USD) and one dose of semen ($7.73 USD). For phase 3 (45 days) charges were labor cost, cost of 6-year bull maintenance per day ($1.85 USD). Finally, the total cost at each phase and the cost of keeping an open cow for a year was calculated.

Statistical analysis. Two-way contingency tables were displayed for pregnancy rates in the three phases for the PP and OC groups. Additionally, a Spearman coefficient correlation test was performed on phase 1 and 3 for age, number of calving, body condition score, presence of corpus luteum, pregnancy and days open. All statistical analyses were performed on IBM SPSS 22 and GraphPad Prism 8 statistical packages. p<0.05 were statistically significant.

RESULTS

Overall pregnancy in phase 1 was 50.8 % with a significant difference (p=0.0093) between groups (PP = 58.6%; 51/87 and OC = 34.1%; 14/41) (Table 1).

At the beginning of phase 1, there was a significant difference in BCS, PP cows were thinner (p<0.0001) than OC, 3.6±1.13 and 5.4±1 respectively. In addition, the percentage of cows with a corpus luteum was also different (p<0.05), for PP and OC group, 43.7% and 85.4%, correspondingly. In contrast, PP group showed a moderate positive correlation between body condition and presence of corpus luteum (r: 0.31, CI 95%, p=0.004). Furthermore, a weak negative correlation between days open and presence of corpus luteum. As well as low positive correlation with pregnancy (r: -0.23, IC 95%, p=0.030 y r: 0.22, IC 95%, p=0.044), respectively (Figure 2).

Table 1. Comparison between pregnancy rates at the three phases for PP and OC groups.

| Phase | Postpartum cows | Open Cows |
|-------|-----------------|-----------|
|       | Pregnant PR %   | Pregnant PR % |
| 1     | 58.6 a          | 34.1 b     |
| 2     | 45.8 a          | 39.1 a     |
| 3     | 40.0 a          | 50.0 a     |
|       | N               | N          |
| 1     | 87              | 41         |
| 2     | 24              | 23         |
| 3     | 25              | 18         |
| Overall PR % | 50.8 | 42.55 |

* (PR) overall pregnancy rate equals the total proportion of pregnant cows (OC+PP) at the end of each phase; (a, b) Different letters within columns represent significant differences (p<0.05).

Figure 2. Correlation between characteristics in the postpartum cows at phase 1. Number of calving (NC), Body condition (BCS), Presence of corpus luteum (PCL), Pregnancy (PG) and Days open (DO).

During this phase, non-pregnant cows in PP group, had fewer (p<0.001) open days (68.58±15.17 d) than pregnant cows (74.98±13.37 d). Conversely, pregnant and empty cows from group OC, did not show significant statistical differences with any of the characteristics (p>0.05).
During phase 2, the percentage of heat detection between groups was different \( (p<0.05) \), 66.7\% (24/36) and 85.2\% (23/27) for PP and OC, respectively. The pregnancy proportion from the second service was 42.5\% with no statistical difference \( (p>0.05) \) (Table 1).

A total of forty-three cows remained for phase 3 (Table 1). The pregnancy rate was 44.2\% \( (p>0.05) \) without statistical differences between groups. However, BCS was significantly lower \( (p<0.0001) \), in PP cows \( (7.4 \pm 0.86) \) than OC \( (7.4 \pm 0.86) \). Almost 90\% \( (p>0.05) \) of cows had a CL at this time. A weak positive correlation \( (r: 0.46, IC 95\%, p=0.019) \) was found in the PP group, between the body condition score and presence of a corpus luteum (Figure 3).

Non-pregnant cows in the PP group had fewer \( (p<0.05) \) days open than the pregnant cows 110.2 ± 11.89 and 141.5 ± 10.48 days, respectively. Conversely, pregnant and empty cows from group OC did not show significant statistical differences with respect to any of the parameters \( (p>0.05) \).

The cost analysis aimed to discern the value of a non-pregnant cow by the end of reproductive program. The initial cost was $59.40 USD for PP cow. At this moment, the opportunity cost was not charged to PP cows. On the other hand, the initial expense by keeping open cows from last year breeding season (opportunity and maintenance costs) was $615.12 USD.

During first phase of breeding season, a PP cow remaining open represents a total cost of $200.19 USD. This amount increased to $278.68 even up to $360.90 USD if not pregnant during the breeding season. Instead, on OC group, an open cow by the end of phase 1 had a cost of $626.13 USD. At the phase three, the cost determined for a non-pregnant cow was $711.9 USD. The total cost for keeping an open cow one year was $1,327.02 USD (Table 2).

The correlation between characteristics in the postpartum cows at phase 3. Number of calving (NC), Body condition (BCS), Presence of corpus luteum (PCL), Dorsal back fat (DBF) and Days open (DO).

![Figure 3](https://example.com/figure3.png)

**Table 2.** Cost analysis (USD) of a non-pregnant cow after FTAI program and natural service in Bos indicus cows postpartum (PP) or barren (OC).

| Initial Cost* | Phase 1 (10 days) | Phase 2 (11-45 days) | Phase 3 (46-90 days) |
|---------------|-------------------|----------------------|----------------------|
| OC | PP | OC | PP | OC | PP | OC | PP |
| Opportunity cost | 350.50 | 0.0 | 268.00 | 96.00 | 302.00 | 129.60 | 345.60 | 172.80 |
| Cow maintenance | 264.60 | 59.40 | 277.2 | 69.30 | 308.70 | 133.65 | 356.45 | 178.20 |
| FTAI /cow | 35.61 | 35.61 | |
| Labor | 7.70 | 7.70 | 9.90 | 9.90 |
| Semen dose | 7.73 | 7.73 | |
| Bull maintenance | | | |
| Total cost | 615.12 | 59.40 | 580.81 | 200.91 | 626.13 | 278.68 | 711.90 | 360.90 |

* Initial cost for open cows (OC) considers 270 days and 60 days for postpartum cows (PP)
DISCUSSION

The pregnancy rate at FTAI (50.4%) was comparable to data from Sá Filho et al (3), Baruselli et al (4), Ayres et al (18), and Sales et al (19), these researchers have worked with Bos indicus cattle using hormone treatments based on progesterone (P4) releasing devices along with estradiol benzoate (EB), PgF2α, and eCG treatment at the time of device withdrawal. Our results pointed out that almost 60% of postpartum cows and one third of OC cows included in the FTAI program would have calve all together during first week of calving season. The addition of open cows from the previous year to FTAI program, cut in half the percentage of pregnant cows during breeding season.

At phase 1, PP cows had reduced BCS and lower proportion with corpus luteum than OC. In this scenario, PP cows had a better pregnancy rate than OC cows on a FTAI program. One explanation could be that PP cows had better response to FTAI treatment. The reason for this disparity could be the use of eCG (20) but more demand for research is in need.

On the other hand, when comparing only pregnant cows on PP group. Most of them had a functional corpus luteum as well as better BCS. This data agrees to Diskin and Kenny (5), who mentioned that BCS at the beginning of the breeding season, could be more important than BCS at calving.

As pointed out by Ibendahl et al (21), the OC would have gained more weight prior to the reproductive program than the postpartum cows. In this latter group, body condition score and days open directly influenced the presence of a corpus luteum and pregnancy. In addition, pregnant cows from the PP group in phase 1 had more days (74 days) since calving than PP non-pregnant cows (68.5 days). These was probably the reason as ovarian activity increased in those cows who started breeding season later in the postpartum period. These data agreed with the review of Stevenson et al (22) pointed out that the percentage of cows cycling increasing was in accord of BCS. The highest percentage of cows with ovarian activity occurred at 81 to 90 days. Nevertheless, this reflection could only be applied to PP cows.

At phase 2, almost 75% of cows diagnosed open at phase 1 were detected in estrus. In similar studies where cows were exposed to a FTAI and the non-pregnant followed up with an AI program at detected estrus, Larson et al (23), Sá Filho et al (3) and Rodrigues et al (24) reported estrous detection rates of 42%, 25.4% y 40%, respectively. In our study 66.6% and 85.2% of the PP y OC, respectively were detected in estrus. A possible explanation for this difference with previous studies, could be that a larger proportion of open cows were cycling and likely to be displaying estrus. On the other hand, it could be the consequence of the time dedicated to estrus detection (2).

The percentage of cows pregnant in phase 2 was like previous data (22,23) with values from 36 to 45.5%. It is worth noting that despite a larger number of cows detected in estrus in the OC group, there were no significant differences in pregnancy rates when compared to the PP group. The most obvious explanation could be erroneous estrus detection as already suggested by others (5,24). Nonetheless, the possibility exists that these animals have a fertility problem despite cycling.

There is ample information to assume that only 50% of cows calve per year in a natural mating program (25,26). During phase 3, the percentage of cows pregnant after natural mating was 44.19%, as found in previous studies (27,28,29). In recent studies, Washaya et al (30) reported 56.7% of pregnancies in a period of three months. The above suggests that variability in the performance of bulls may be responsible for limiting the possibility of increasing pregnancy rates (12,31). However, the bull is at a disadvantage when used after an AI program, as he is likely to have to deal with the cows that have problems conceiving. Several reports have highlighted that no matter what reproductive program is used, a constant feature is that around 20% of the cows fail to become pregnant in a short breeding season (32,33). The percentage of open cows at the end of a breeding season was similar in the present study. The combination of FTAI, ED and NS during a short breeding season, seeks to have the greatest number of pregnancies in a compact calving season.

Furthermore, it is quite possible that a large percentage of open cows at the end of the breeding period may be anestrous. More research is needed on this subject. Regardless of the causes of this shortcoming.
Because of nature of beef cattle production system, it requires an economic analysis. Reproduction efficiency is related to profitability in a beef farm. The cost analysis approach in this experiment was towards the value of keeping an open cow to the forthcoming breeding season. In effect, as observed in table 2, an open cow from the previous breeding program, cost 10 times more at the beginning of the next breeding season (phase 1). The reason to keep an open cow for the next breeding season, could be related to a management decision. Ibendahl and Anderson (34) explained other factors for deciding to keep an open cow. The economic price of the replacement heifers, plus the value of the weaned calf and maintenance cost of the cow are examples for decision making. On the other hand, Torres-Aburto et al (17) studying cost of commercial cow-calf units in the tropics of south east Mexico. They concluded that increasing calving interval, conduits towards a decrease in calves per year, reducing the profitability and stability of the production unit. In this experiment the total cost of keeping an open cow from a previous breeding season reached a total cost of $1327.02 USD which it was 3 times the cost of an open PP cow.

The pregnancy rate at first phase was higher in PP cows than OC cows. No differences in pregnancy rate were found in phases two and three. At the end of breeding season, a pregnancy rate of 80% was found. The incorporation of open cows from the previous breeding program was more expensive than PP cows in all phases of the breeding program. This could be not economically feasible for a cow-calf enterprise to retain an open cow for rebreeding one year or more.

**Conflict of interest**

The authors declare no conflict of interest.

**Acknowledgements**

The authors are grateful for the assistance to the administrative staff of the Centre for Teaching, Research, and Extension in Tropical Animal Husbandry belonging to the Faculty of Veterinary Medicine of the National Autonomous University of Mexico for their assistance in the housing and care of animals.

**Financial support statement**

This research received partial support from Programa de Apoyos a Proyectos de Investigación e Innovación Tecnológica (PAPIIT) IN216820, Universidad Nacional Autónoma de México.

**REFERENCES**

1. Orihuela A. Some factors affecting the behavioural manifestation of oestrus in cattle: a review. Appl Anim Behav Sci. 2000; 70(1):1–16. [https://doi.org/10.1016/S0168-1591(00)00139-8](https://doi.org/10.1016/S0168-1591(00)00139-8)

2. Galina CS, Orihuela A. The detection of estrus in cattle raised under tropical conditions: What we know and what we need to know. Horm Behav. 2007; 52(1):32–38. [https://linkinghub.elsevier.com/retrieve/pii/S0018506X07000724](https://linkinghub.elsevier.com/retrieve/pii/S0018506X07000724)

3. Sá Filho MF, Penteado L, Reis EL, Reis TANPS, Galvão KN, Baruselli PS. Timed artificial insemination early in the breeding season improves the reproductive performance of suckled beef cows. Theriogenology. 2013; 79(4):625–632. [http://dx.doi.org/10.1016/j.theriogenology.2012.11.016](http://dx.doi.org/10.1016/j.theriogenology.2012.11.016)

4. Baruselli PS, Ferreira RM, Sá Filho MF, Bó GA. Review: Using artificial insemination v. natural service in beef herds. animal. 2018; 12(1):45–52. [https://www.cambridge.org/core/product/identifier/S175173111800054X/type/journal_article](https://www.cambridge.org/core/product/identifier/S175173111800054X/type/journal_article)
5. Diskin MG, Kenny DA. Managing the reproductive performance of beef cows. Theriogenology. 2016; 86(1):379–87. http://dx.doi.org/10.1016/j.theriogenology.2016.04.052

6. Pérez-Torres L, Rubio I, Corro M, Cohen A, Orihuela A, Galina CS, et al. A pre-synchronization program at early postpartum might increase the chances of Bos indicus cows cycling prior to 50 days regardless of the length of calf separation. J Reprod Dev. 2015; 61(3):199–203. https://www.jstage.jst.go.jp/article/jrd/61/3/61_2014-114/_article

7. Díaz BR, Galina CS, Rubio I, Corro M, Pablos JL, Orihuela A. Monitoring changes in back fat thickness and its effect on the restoration of ovarian activity and fertility in Bos indicus cows. Reprod Domest Anim. 2018; 53(2):495–501. https://dx.doi.org/10.1111/rda.13136

8. Stagg K, Spicer LJ, Sreenan JM, Roche JF, Diskin MG. Effect of Calf Isolation on Follicular Wave Dynamics, Gonadotropin and Metabolic Hormone Changes, and Interval to First Ovulation in Beef Cows Fed Either of Two Energy Levels Postpartum. Biol Reprod. 1998; 59(4):777–783. https://doi.org/10.1095/biolreprod59.4.777

9. Sinclair KD, Molle G, Revilla R, Roche JF, Quintans G, Marongiu L, et al. Ovulation of the first dominant follicle arising after day 21 post partum in suckling beef cows. Anim Sci. 2002; 75(1):115–126. https://www.cambridge.org/core/product/identifier/S13577298000052899/type/journal_article

10. Chacón J. Assessment of Sperm Morphology in Zebu Bulls, under Field Conditions in the Tropics. Reprod Domest Anim. 2001; 36(2):91–99. http://doi.wiley.com/10.1046/j.1439-0531.2001.00253.x

11. Chenoweth PJ, McPherson FJ. Bull breeding soundness, semen evaluation and cattle productivity. Anim Reprod Sci. 2016; 169:32–36. http://dx.doi.org/10.1016/j.anireprosci.2016.03.001

12. Barth AD. Review: The use of bull breeding soundness evaluation to identify subfertile and infertile bulls. Animal. 2018; 12(s1):s158–s164. http://dx.doi.org/10.1017/S1751731118000538

13. Orihuela A, Galina C, Escobar J, Riquelme E. Estrous behavior following prostaglandin F2α injection in Zebu cattle under continuous observation. Theriogenology. 1983; 19(6):795–809. https://linkinghub.elsevier.com/retrieve/pii/0093691X9390323W

14. Bisinotto RS, Chebel RC, Santos JEP. Follicular wave of the ovulatory follicle and not cyclic status influences fertility of dairy cows. J Dairy Sci. 2010; 93(8):3578–3587. http://dx.doi.org/10.3168/jds.2010-3047

15. Nicholson MJ, Butterworth MH. A guide to condition scoring of Zebu Cattle. Addis Ababa: International Livestock Centre for Africa; 1986.

16. Schröder UJ, Staufenbiel R. Invited review: Methods to determine body fat reserves in the dairy cow with special regard to ultrasonographic measurement of backfat thickness. J Dairy Sci. 2006; 89(1):1–14. https://doi.org/10.3168/jds.S0022-0302(06)72064-1

17. Torres-Aburto VF, Domínguez-Mancera B, Vázquez-Luna D, Espinosa Ortiz VE. Cost of the calving interval in tropical bovine production in southeastern Mexico. Agro Product. 2020; 13(7):45–51 https://doi.org/10.32854/agrop.vi.1651

18. Ayres H, Martins CM, Ferreira RM, Mello JE, Domínguez JH, Souza AH, et al. Effect of timing of estradiol benzoate administration upon synchronization of ovulation in suckling Nelore cows (Bos indicus) treated with a progesterone-releasing intravaginal device. Anim Reprod Sci. 2008; 109(1–4):77–87. https://linkinghub.elsevier.com/retrieve/pii/S0378432007003971
19. Sales JNS, Carvalho JBP, Crepaldi GA, Cipriano RS, Jacomini JO, Maio JRG, et al. Effects of two estradiol esters (benzoate and cypionate) on the induction of synchronized ovulations in Bos indicus cows submitted to a timed artificial insemination protocol. Theriogenology. 2012; 78(3):510–516. http://dx.doi.org/10.1016/j.theriogenology.2012.02.031

20. Baruselli P., Reis E., Marques M., Nasser L., Bô G. The use of hormonal treatments to improve reproductive performance of anestrous beef cattle in tropical climates. Anim Reprod Sci. 2004; 82–83:479–486. https://linkinghub.elsevier.com/retrieve/pii/S037843200400079X

21. Ibendahl GA, Anderson JD, Anderson LH. Deciding when to replace an open beef cow. Agr Finance Rev. 2004; 64(1):61-74. https://doi.org/10.1108/00214660480001154

22. Stevenson JS, Johnson SK, Milliken GA. Incidence of Postpartum Anestrus in Suckled Beef Cattle: Treatments to Induce Estrus, Ovulation, and Conception. Prof Anim Sci. 2003; 19(2):124–134. http://dx.doi.org/10.15232/S1080-7446(15)31391-7

23. Larson D, Musgrave JA, Funston RN. Effect of Estrus Synchronization with a Single Injection of Prostaglandin During Natural Service Mating Effect of Estrus Synchronization with a Single Injection of Prostaglandin During Natural Service Mating. Nebraska Beef Cattle Reports. 2009; 527:9-10. https://digitalcommons.unl.edu/animalscinbcr/527

24. Rodrigues WB, Jara JDP, Borges JC, Fialho de Oliveira LO, Gomes de Abreu UP, Anache NA, et al. Efficiency of mating, artificial insemination or resynchronisation at different times after first timed artificial insemination in postpartum Nellore cows to produce crossbred calves. Anim Prod Sci. 2018; 59(2):225-231. http://www.publish.csiro.au/?paper=AN17466

25. Palmer MA, Olmos G, Boyle LA, Mee JF. Estrus detection and estrus characteristics in housed and pastured Holstein-Friesian cows. Theriogenology. 2010; 74(2):255–264. http://dx.doi.org/10.1016/j.theriogenology.2010.02.009

26. Silva-Mena C, Aké-López R, Delgado-León R. Sexual behavior and pregnancy rate of Bos indicus bulls. Theriogenology. 2000; 53(4):991–1002. https://linkinghub.elsevier.com/retrieve/pii/S0093691X00002454

27. Molina R, Bolaños I, Galina C., Pérez E, Paniagua G, Estrada S. Sexual behaviour of Zebu bulls in the humid tropics of Costa Rica: single versus multiple-sire groups. Anim Reprod Sci. 2000; 69(3–4):139–148. https://linkinghub.elsevier.com/retrieve/pii/S037843200002062

28. Molina R, Galina CS, Camacho J, Maquivar M, Diaz GS, Estrada S, et al. Effect of alternating bulls as a management tool to improve the reproductive performance of suckled Zebu cows in the humid tropics of Costa Rica. Anim Reprod Sci. 2002; 69(3–4):159–173. https://linkinghub.elsevier.com/retrieve/pii/S0378432001001798

29. Molina R, Galina CS, Diaz MS, Galicia L, Estrada S. Evaluation of a bull rotating system using natural mating: Effect on the reproductive performance of zebu cows. Agrociencia. 2003; 37(1):1–10. http://www.colpos.mx/agrocien/Bimestral/2003/ene-feb/art-1.pdf

30. Washaya S, Tavirimirwa B, Dube S, Sisito G, Tambo G, Ncube S, et al. Reproductive efficiency in naturally serviced and artificially inseminated beef cows. Trop Anim Health Prod. 2019; 51(7):1963–1968. http://link.springer.com/10.1007/s11250-019-01889-z

31. Petherick JC. A review of some factors affecting the expression of libido in beef cattle, and individual bull and herd fertility. Appl Anim Behav Sci. 2005; 90(3–4):185–205. https://linkinghub.elsevier.com/retrieve/pii/S0168159104001959

32. Pessoa GA, Martini AP, Sá Filho MF, Batistella Rubin MI. Resynchronization improves reproductive efficiency of suckled Bos taurus beef cows subjected to spring-summer or autumn-winter breeding season in South Brazil. Theriogenology. 2018; 122:14–22. https://doi.org/10.1016/j.theriogenology.2018.08.021
33. Ferreira RM, Conti TL, Gonçalves RL, Souto LA, Sales JNS, Sá Filho MF, et al. Synchronization treatments previous to natural breeding anticipate and improve the pregnancy rate of postpartum primiparous beef cows. Theriogenology. 2018; 114:206–211. https://doi.org/10.1016/j.theriogenology.2017.11.022

34. Ibendahl G, John Anderson. Open Cow Replacement Decisions: an Application of Asset Replacement Theory. Western Agricultural Economics Association Annual Meetings. 2001. https://ageconsearch.umn.edu/record/36184