Animal Reproduction

AI and IATF

Dose-dependent effects of estradiol benzoate for resynchronization of ovulation at 14 days after timed artificial insemination in beef cows

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A dose of 1mg estradiol benzoate (EB) given at 14 days after timed artificial insemination (TAI) does not disturb the establishment of pregnancy in beef cattle. We aimed with this study to compare the effects of 1mg *vs.* 2mg EB given associated to progesterone (P4) at 14 days after TAI on follicle wave emergence (FWE) and pregnancy rates (PR) in *Bos indicus* beef cows. Nelore suckled cows (n=1,030) with a body condition score (BCS) between 2 and 4 (scale: 1-5) were subject to an estradiol/P4-based protocol and the day of TAI was considered D0. On D14, the cows received an 8-16 days-used intravaginal P4 device (Sincrogest®, Ourofino Saúde Animal) and were randomly assigned in two groups according to EB dose (Sincrodiol®,Ourofino): EB-1 (1mg; n=517) or EB-2 (2mg; n=513) groups. A subgroup of cows (n=18-19/group) was subjected to daily ultrasonography evaluations from D14 to D22 to evaluate follicular and corpus luteum (CL) dynamics. On D22, the P4 devices were removed and non-pregnant (NP) cows were identified based on determination of luteal size and blood perfusion to detect CL regression using Color Doppler ultrasonography. The NP cows received 1mg estradiol cypionate (SincroCP®,Ourofino), 0.5 mg sodium cloprostenol (Sincrocio®,Ourofino) and 300 IU eCG (Novormon®,Zoetis) on D22 and on D24 a second TAI was performed. Confirmatory diagnosis of pregnancy was performed between days 30 and 35 (D30-35) after first and second TAI. The data were evaluated by Fisher's exact test or logistic regression (GLIMMIX) of SAS, considering the effects of group, sire, BCS, farm, parity category, and their possible interactions. The proportion of cows with a synchronized FWE (from 3-5 days after EB treatment) was greater (P<0.05) in the EB-2 (89.5% [17/19]) than in the EB-1 group (44% [8/18]). As expected, a BCS effect (P<0.05) was observed on PR after first TAI, indicating greater PR in cows with BCS of 3 (50% [347/697]) compared to 2.5 (38% [117/310]). The PR at D22 and D30-35 after first TAI were greater (P<0.05) in the EB-1 (54.5% [281/516] and51% [264/515], respectively) than in EB-2 group (48% [243/510] and 42% [212/508], respectively). The rate of potential pregnancy loss between D22 and D30-35 was greater (P<0.05) in the EB-2 (13% [31/243]) than in EB-1 group (6% [17/281]). No difference (*P*>0.1) was observed in the PR of cows submitted to the second TAI between EB-1 (47% [106/226]) and EB-2 groups (42% [109/257]). Furthermore, cumulative PR (first and second TAI) was greater (P<0.05) in the EB-1 group (73% [370/508]) than in the EB-2 group (64% [322/502]). In conclusion, the use of 2mg EB at D14 after TAI improves the synchrony of the FWE but does not increase the PR at the second TAI and negatively impacts on the previous pregnancy. Therefore, the use of 1mg EB associated with the P4 device is preferred to avoid the risk of pregnancy loss in resynchronization protocols at D14 post-TAI in beef suckled cows. Acknowledgments: FAPESP(2015/10606-9, 2019/07805-0)

Animal Reproduction

AI and IATF

Evidence of negative relationship between reproductive performance and feed efficiency in Nelore (*Bos indicus*) heifers submitted to TAI

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The objective of this study was to evaluate phenotypic relationships between fertility traits and feed efficiency in Nelore *(Bos indicus)* heifers submitted to TAI. A total of 149 Nelore heifers [11.2±1.3 months of age, body weight (BW)= 249.6±23.3 kg and body condition score (BCS)= 2.8±0.03 (1-5 scale)] from HoRa (MS state, Brazil) were used. Individual feed intake was monitored using the Intergado efficiency automated feeding system (Intergado®, Brazil) for 90 days to estimate the residual feed intake (RFI). Heifers were synchronized to receive TAI 70 days after starting feedlot. At random day of the estrous cycle (without previous synchronization; D0), heifers received an intravaginal device with 0.6g P4 (Fertilcare 600®, MSD) associated with 2mg EB (Fertilcare Sincronização®, MSD). At the same time, BW and BCS were evaluated and the presence of CL was detected by US (DP-2200Vet, Mindray®). Also, longissimus Muscle Area (LMU) and subcutaneous rib fat thickness (RFAT) were estimated via US (SSD 500, Aloka®). On D8, device was removed and heifers received 0.5mg PGF (Ciosin®, MSD), 0.5mg of EC (Fertilcare Ovulação®, MSD) and 200IU of eCG (Folligon®, MSD). Heifers were painted with chalk on their tailheads, and removal of chalk on D10 was used as an indication of estrus. TAI was performed 48h after device removal. Pregnancy diagnosis was done by US 30 days after TAI, heifers not pregnant were assigned to a second TAI, following the same procedure already described. Statistical analysis was performed by GLIMMIX procedure of SAS® 9.4. At the end of feedlot, heifers were classified as low RFI (good feed efficiency; -1.21±0.16 kg DM/d, n=75) and high RFI (1.47±0.08 kg DM/d, n=74). On D0, heifers with low RFI or high RFI had similar average BCS (3.00±0.02 *vs.* 3.05±0.03; P=0.12), BW (323.8±3.0 *vs.* 325.4±2.7kg; P=0.69) and cyclicity rate [9.3% (7/75) *vs.* 14.9% (11/74); P=0.18]. Moreover, LMU did not differ among groups (Low RFI= 52.0±0.7 *vs.* High RFI= 51.3±0.7cm^2; P=0.42]. However, the RFAT was lower in heifers with good feed efficiency (Low RFI) when compared with High RFI (4.87±0.2 vs. 5.36±0.2 mm; P=0.01). Heifers with low RFI expressed less estrus than high RFI [68.0% (51/75) vs. 82.4% (43/74); P=0.01]. There was a tendency for lower pregnancy per AI (P/AI) in Low RFI heifers at 1st TAI [Low RFI= 50.7% (38/75) vs. High RFI= 59.5% (44/74); P=0.08] and at 2nd TAI [Low RFI= 19.4% (7/36) *vs.* High RFI= 36.7% (11/30); P=0.10]. Finally, the cumulative P/AI (1st+2nd TAI) was 13 percentage points lower for Low RFI heifers [Low RFI= 61.3% (46/75) *vs.* High RFI= 74.3% (55/74); P=0.02]. Although the RFI did not impact phenotypic characteristics as BCS, BW, LMU and cyclicity rate, heifers with high feed efficiency (Low RFI) had lower RFAT and cumulative P/AI compared with high RFI. The results showed a negative relationship between reproductive performance and feed efficiency in Nelore heifers, which must be considered within the framework of the entire beef enterprise.

Animal Reproduction

AI and IATF

Color Doppler ultrasonography on follicular dynamics in Holstein cows with high or low antral follicle count

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In order to better understand the relationship between antral follicle count (AFC) and the reproductive performance in dairy cattle, we compared the follicular dynamics and ovarian blood perfusion in Holstein cows (*Bos taurus taurus*) with low or high AFC. For this purpose, the number of antral follicles (≥ 2 mm of diameter) was determined in lactating Holstein cows (n = 80) with at least two lactations, a postpartum period ranging from 40 to 60 days, daily milk production between 45 and 60 liters, body condition score ranging from 2.75 to 3.0 (scale 1 to 5) and kept in a free-stall system. Then, we identified the low AFC group (≤ 15 follicles, n = 9) and high AFC group (≥ 25 follicles, n = 9). Both low and high AFC groups received the same conventional ovulation synchronization protocol, with an intravaginal progesterone device (Repro sync®, GlobalGen Vet Science, Jaboticabal, São Paulo, Brazil), 2 mg estradiol benzoate (Syncrogen®, GlobalGen Vet Science) and 25 µg GnRH synthetic analog - lycellelin (Tec- Relin®, Agener União Saúde Animal, Embu-Guaçu, São Paulo, Brazil) on a random day of the estrous cycle (D0). On the D7, 0.526 mg of sodium cloprostenol (synthetic prostaglandin analog - PGF2α, Induscio®, GlobalGen Vet Science) was applied. On D8, the progesterone device was removed, 0.526 mg of PGF2α and 1 mg of estradiol cypionate were applied (Cipion®, GlobalGen Vet Science). All cows were daily evaluated by the same technician with an ultrasound (SonoScape ™, Model S8, Domed, Valinhos, Brazil), in B and Doppler mode, during the days of the protocol and eleven days after ovulation, to evaluate the corpus luteum (CL). In addition, blood samples were collected from all animals 11 days after ovulation in duplicate to determine progesterone and estrogen concentrations by chemiluminescence method. In both groups, we compared the diameter of the preovulatory follicle (OF), the area of vascularization of the OF, the area of the CL, and the area of vascularization of the CL. Parametric data were analyzed by the T test or ANOVA and non-parametric by the Mann-Whitney test (P≤0.05). The animals with low AFC showed greater diameter of the OF (16.09 ± 0.35 vs 15.00 ± 0.40 mm; P = 0.05), greater vascularization area of the OF (17.36 ± 2.33 vs 8.16 ± 3.20 mm^2; P = 0.005) , greater area of CL (579.10 ± 16.90 vs 405.80 ± 21.20 mm^2; P = 0.0001) and the largest blood perfusion area of CL (97.16 ± 9.47 vs 68.30 ± 5.24 mm^2; P = 0.021) . Progesterone concentrations were also higher for low AFC animals when compared to high AFC (3.16 ± 0.26 vs 2.28 ± 0.26 ng/ml; P = 0.022). Estradiol concentrations did not differ between low and high AFC groups (31.66 ± 2.51 vs 28.86 ± 5.00 pg/ml; P = 0.628). We concluded that the low AFC Holstein cows showed larger ovulatory follicles and CLs and with higher blood perfusion when compared to the high AFC cows.

Animal Reproduction

ABSTRACTS: 34TH ANNUAL MEETING OF THE BRAZILIAN EMBRYO TECHNOLOGY SOCIETY (SBTE)

AI and IATF

Mineral supplementation with beta-carotene and vitamins and their effect on reproductive performance in TAI beef cows kept on pasture conditions

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The objective was to evaluate the effect of mineral supplementation with the addition of beta-carotene and vitamins (A, D, E and biotin) on the conception rate of the 1st TAI in grazing Nellore cows (*Bos indicus*).A total of 497 multiparous cows (5.68±0.11 parities) were homogeneously divided in 4 paddocks (*Brachiaria brizantha* spp) according to the body condition score (BCS=2.8 ±0.01;P<0001) and calving period [births at the beginning of the experimental period (BB; from September 15th to October 15th) and end of the experimental period (BE; from October 16th to October 31st)] in two experimental groups: Control (mineral supplementation; Fosbovi® Reprodução; n=251) and Vitamins (control+150mg beta-carotene+40.000IU Vit.A+5.000IUVit.D3+300mg Vit.E+20mg biotin/animal/day; n=246). Supplementation started 30 days before the 1st TAI and ended 30 days later, totaling 60 days of treatment, and was provided by DSM Produtos Nutritional Brazil S.A. Cows were synchronized with a P4/E2-based TAI protocol. Animals were rotated among paddocks every three days to avoid the pasture effect on the results.At the pregnancy diagnosis (30th and 77th days of gestation) the size of the fetus was also measured using the distances from the crown-rump and the thoracic diameter. Data were analyzed using the GLIMMIX procedure of SAS and the value P<0.05 was considered for effect and trend when P>0.05 and P<0.10. The fixed factor was the treatment, the random ones were calving period, farm, inseminator and bull. The Tukey test was used. The conception rate at the 1st TAI showed an increasing trend (P=0.08) for the vitamins group [control:56.6%(142/251) vs. treated:64.2%(158/246)].For estrus manifestation rate, there was an interaction between calving period*treatment (P=0.04), with vitamins increasing estrus detection in BB period. FD was influenced by the calving season (P<0001), with largest diameters in the BE period, with no treatment effect (P=0.14).Vitamins group presented the largest embryo crown-rump (P=0.002) and thoracic diameter (P<0001) lengths at 30th day of gestation. In addition, there was interaction between calving period*treatment for crown-rump length at 77th day of gestation (P=0.02), with embryos from vitamins group presenting longer length in BE period. When analyzed by repeated measure in time, the crown-rump length of the fetus at 30th and 77th days of gestation showed an interaction treatment*time*calving period (P=0.0010), however, this interaction was not observed for thoracic diameter (P=0.09).The BCS at TAI moment and at first pregnancy diagnosis was increased (P<0001) for the vitamins group, regardless of the calving period. Animals that gained BCS had a higher conception rate at the 1st TAI than animals that maintained or lost BCS (P=0.0042).The data from the present experiment support that treatment with beta-carotene and vitamins increases the conception rate of the 1st TAI, the development of the conceptus and the BCS of cows.

Animal Reproduction

AI and IATF

Fertility of lactating Holstein cows initiating the FTAI protocol with estradiol benzoate or GnRH, and receiving or not GnRH 2 days later

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The experiment evaluated strategies to initiate the FTAI protocol (estradiol benzoate [EB] vs buserelin acetate [GnRH]), and the inclusion of GnRH 2 d later, using 943 Holstein cows (primiparous and multiparous, 43.8±0.3 Kg of milk/d). We hypothesized that GnRH on d0 would promote greater fertility, and GnRH on d2 would improve fertility, especially in cows receiving EB on d0. Cows were randomized into 4 groups using a 2x2 factorial (Groups: EBd0, EBd0-Gd2, Gd0, and Gd0-Gd2). On d0, cows received a 2g progesterone (P4) implant, and groups EBd0 received 2mg EB, whereas Gd0 groups received 10µg GnRH. Only in Gd2 groups, cows received 10µg GnRH on d2 of the protocols. The rest of the protocol was similar, with cows receiving 0.5mg cloprostenol (PGF) on d7, and a second PGF on d8, concomitant with P4 implant removal, and 1mg estradiol cypionate. FTAI was performed on d10 in all groups. Statistical analyses were performed using GLIMMIX of SAS 9.4 (P≤0.05). On d0, the proportion (% [n]) of cows with CL was similar for all groups (76.1% [799]). Ovulation after d0 was greater in groups with GnRH on d0 and d2 (EBd0 = 15.1 [179], EBd0-Gd2 = 29.1 [189], Gd0 = 44.2 [181]; Gd0-Gd2 = 55.4% [186]). EB treatment promoted greater luteolysis after d0 compared to GnRH (48.3 [271] vs 32.5% [286]). These differences in ovulation and luteolysis after d0 promoted an interaction between treatments on presence of CL on d7, with GnRH on d2 in cows that received EB on d0 increasing the number of cows with CL at PGF, and protocols with GnRH on d0, with or without GnRH on d2, inducing more cows to have CL on d7 (EBd0 = 56.6 [182], EBd0-Gd2 = 69.1 [191], Gd0 = 89.8 [187], Gd0Gd2 = 87.4% [190]). When creating categories of cows according to presence (Yes) or absence (No) of CL on d0 and d7, EBd0 groups had more cows in NoNo (14.8 [365] vs 3.8% [367]) and less cows in YesYes (52.1 [365] vs 70.3% [367]) compared to cows with GnRH on d0. The NoNo cows had lower pregnancy per AI (P/AI) than other classes (NoNo = 30.9% [68], NoYes = 45.3% [106], YesNo = 40.9% [110], YesYes = 44.2% [448]). Cows without CL on d0 that ovulated had greater P/AI than cows not ovulating after d0 (45.3 [106] vs 30.9% [68]), although in cows with CL on d0, ovulation did not influence P/AI (41.5 [159] vs 44.4% [399]). GnRH on d0 increased expression of estrus compared to EB (86.1 [445] vs 79.4% [442]), and cows in estrus had greater P/AI (44.7 [734] vs 27.5% [153]). There was an interaction between treatments in P/AI, in which GnRH on d2 improved fertility of cows receiving EB on d0 (EBd0 = 37.5 [232], EBd0-Gd2 = 44.3 [237], Gd0 = 42.8 [236], Gd0-Gd2 = 42.0% [238]). In conclusion, GnRH on d0 of the protocol promoted better fertility and optimized CL status compared to protocols initiated with EB, and GnRH on d2 of protocols initiating with EB improved ovarian dynamics and fertility in dairy cows. Acknowledgments: São Jorge Farm, FAPESP Grant # 2018/03798-7, CAPES, CNPq and GlobalGen vet science

Animal Reproduction

ABSTRACTS: 34TH ANNUAL MEETING OF THE BRAZILIAN EMBRYO TECHNOLOGY SOCIETY (SBTE)

AI and IATF

Use of automated monitoring device to detect heat in Holstein (Bos taurus) and Gir (*Bos indicus*) cows

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Precision dairy monitoring technologies have become increasingly popular for heat detection. The objectives of the studies were to validate the accuracy of an automated monitoring device collars (AMD, Ovalert tags, CRV, Arnhem, Netherlands) for heat detection in lactating Holstein cows (*Bos taurus*) and in Gir heifers (*Bos indicus*). In study 1, lactating dairy Holstein cows (n=91) received the AMD collars at D0 of the experiment. Cows were synchronized with intravaginal P4 device plus 2mg of estradiol benzoate (EB). On day 8, the P4 device was removed and all animals received PGF, no EB were applied at this time in this experiment. Ultrasound examinations (US) were done twice a day between day 8 and day 14 and on day 20 to evaluate follicular dynamics and ovulation. In study 2, Gir heifers (n=81) received the AMD plus intravaginal P4 device and 2mg of EB on day 0. On day 8, the P4 device was removed and all animals received PGF, 1mg of estradiol cypionate, 300 IU of eCG and a BIP. The US was done twice a day between day 8 to day 11 and on day 20 to evaluate follicular dynamics and ovulation. In both studies, the ovulation was used as a gold standard to calculate the test characteristics: sensitivity (SE), specificity (SP), accuracy (AC), negative predicted value (NPV) and positive predicted value (PPV). In study 1, 59.3% (59/91) of the cows were detected in estrus by CMA. The ovulation rate until d14 of the protocol was 81,3% (74/91). The average diameter of the ovulatory follicle was 16.3 ± 2.6mm. Fifty one animals were classified as TP (true positive - positive CMA and ovulation), 14 TN (true negative - no estrus on CMA and no ovulation), 3 FP (false positive - positive CMA and no ovulation) and 23 animals were classified as FN (false negative - negative on CMA, positive ovulation). The CMA SE was 68.9%, SP 82.4%, PPV 94.4%, NPV 37.8% and AC 71.4%. In study 2 95% (77/81) of the heifers were detected in estrus by the CMA. The ovulation rate until d11 of the protocol (d11) was 76.5% (62/81). The average diameter of the ovulatory follicle was 12.1 ± 1.55 mm. Fifty-nine animals were classified as TP, 1 TN, 18 FP and 3 animals FN. The CMA SE was 95.2%, the SP was 5.3%, the PPV was 76.6%, the NPV was 25% and the AC was 74.1%. In conclusion, indices reveal great sensitivity, specificity, PPV and accuracy for CMA in lactating Holstein cows, low NPV were observed and our hypothesis is that part of the cows had silent heat. In experiment 2, although the specificity and the NPV were low we still recommend and validate CMA use in *Bos indicus* heifers. When analyzing why some animals have demonstrated estrus by CMA without ovulation it is suggested that there was a failure in the synchronization of the new follicular wave during the FTET protocol and the estrus identified by CMA would come from the application of estradiol benzoate in D8 of protocol.The data showed that the heat can be predicted using an AMD in lactating Holstein cows and in Gir heifers.

Animal Reproduction

AI and IATF

Evaluation of pregnancy rates in milk buffaloes submitted to FTAI with ovsynch or P4/E2 and eCG based protocols with refrigerated or frozen semen during favorable or unfavorable breeding season

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The aim of this study was to compare the efficiency of chilled and frozen semen (diluted in Botu-Bov®) as a strategy to increase the pregnancy rate in the FTAI in buffaloes. For this, two studies were carried. In the first study, the ejaculates of 2 Murrah bulls obtained by collection with artificial vagina were used; the semen samples were fractionated in 2 aliquots; one diluted in Botu-Bov® commercial extender for refrigeration (BB-R, refrigerated group) and one diluted in the same extender for freezing (BB-F, frozen group). The samples were refrigerated at 5°C/24 hours at a rate of 0.25°C/minute and maintained at that temperature for 24 hours in the BB-R group. After four hours of equilibration at 5°C, the BB-F samples were frozen at 5 cm from the surface of the liquid nitrogen and subsequently immersed in the nitrogen. After 30 days postpartum, 90 buffaloes were submitted to the following synchronization scheme on a random day of estrus cycle: D0 (14:00hs), application of 20 µg im of GnRH (Cystorelin®), in D7 (14:00 hs) 0,530 mg im of PGF2α (Croniben®), in D9 (14:00 hs) 20 µg i.m of GnRH (Cystorelin®) and in D10 (6:00 hs in the morning) the AI was performed. In the second study, 8 bulls were collected and the process of experiment 1 was repeated. However, the 446 buffaloes were randomly distributed in four blocks according to the year: B1 = 143 (2014), B2 = 34 (B3 = 90 (2016) and B4 = 179 (2017), each block being subdivided into two for the AI with chilled semen and frozen semen of the same ejaculate and the same bull, maintaining a balance between cyclic and acyclic animals between groups. These animals were submitted to the following synchronization protocol: In the afternoon (T) D0 (14:00hs), animals received 2.0 mg im estradiol benzoate (BE, Estrogin®) and auricular implant CRESTAR® 3.0 mg P4), D9 (T) withdrawn from the implant and application of 400 IU im eCG (Folligon® 5000) + 0.530 mg $PGF_2\alpha$ Cloprostenol im (Sincrocio®). In D10 (T) 1.0 mg im of BE (Estrogin®) was applied in the morning (M) D12 (8:00hs) AI. All females were inseminated with $50x10^6$ total SPTZ/dose, and the dose corrections were performed according to motility after 24 hours of refrigeration and post-thaw. Ultrasonography (Aloka-SSD 500, 5 MHz probe) was performed 30 days after the AI for pregnancy diagnosis in both studies. The pregnancy rate for the first study was 57.8^a and 31.1^b% for chilled and frozen semen ($p<0.05$), respectively. For the second study, total pregnancy rates of 48.2^a% and 34.6^b% were found for chilled and frozen semen, respectively. Difference that was maintained for all the evaluated years, being found rates of 45.1ª and 33.3b% (2014); 50.0^a and 37.5^b% (2015), 55.3^a% and 30.2^b% (2016) and 46.6^a% and 37.1^b% (2017). In both studies significant statistical differences were obtained ($P<0.05$), being 26.6 and 13.6% for study 1 and 2, respectively. It is concluded that dairy buffaloes presented higher pregnancy rates for refrigerated semen.

Animal Reproduction

ABSTRACTS: 34TH ANNUAL MEETING OF THE BRAZILIAN EMBRYO TECHNOLOGY SOCIETY (SBTE)

AI and IATF

Effect of increasing PGF2alfa dose during TAI protocol based on P4 and Estradiol Benzoate on conception of lactating dairy cows

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Lack of complete luteolysis has been shown to reduce fertility in timed AI protocols that are used in dairy cows and heifers, using either commercially available PGF2α analogue, dinoprost tromethamine or cloprostenol sodium. The objective was to compare the dose of 3 mL versus 2 mL of Ciosin, during TAI protocol based on P4 and Estradiol Benzoate on conception of lactating dairy cows. The experiment was conducted at a commercial dairy farm in Minas Gerais State, Brazil, from April to November 2019. The cows were housed in a composted bedding pack barn, and milked 3 times daily. Cows was fed *ad libitum* a TMR based on corn silage as forages, with concentrates composed of corn and soybean meal, and added minerals and vitamins, which was balanced to meet or exceed the nutritional requirements of lactating dairy cows. Were analyzed 289 inseminations events in lactating Holstein cows, producing 32kg of milk/day. Cows were blocked by parity (primiparous and multiparous); all cows that were past the voluntary waiting period (30 DIM) and not pregnant was used and randomized into the study, without regard to whether they had been previously used in the study. Within each block, cows were randomly assigned to receive 1 of 2 treatments: Control Group (n = 136) – application of 2 mL of Ciosin at Day -2; and Treatment Group (n = 153) - application of 3 mL of Ciosin at Day -2 of the following TAI protocol: Day -10 - insertion of a progesterone slow-release intravaginal device containing 1.0 g of progesterone and an IM injection of 2.0 mL of estradiol benzoate; Day -2 - an IM injection of 2 or 3 mL of Ciosin and 2 mL of eCG; Day 0 - TAI. In all groups, the ovaries were evaluated by transrectal ultrasonography (Mindray with a 5.0 - 7.5-MHz linear-array transducer) on d −10 and 7 to determine presence of a CL (independent of its diameter) and large follicles. The synchronization rate was determinate by presence of CL on day 7. Pregnancy success (P/AI) was calculated by dividing the number of pregnant cows at the pregnancy diagnosis at 32 and 60 days after TAI by the total number of cows that received TAI. The binomial variables (presence of CL on day 7 after TAI and P/AI on day 32 and 60) was analyzed by logistic regression on MINITAB program, including in the model effects of treatment, parity, presence of CL on day -10, and their interactions. No effects (P>0,05) were detected on percentage of cows with CL on day 7 after TAI (88.24 vs. 90.95%) and P/AI on 32 days (23.53 vs. 28.10%) and 60 days (20.59 vs. 21.57%) after TAI for cows that received 2 or 3 mL of Ciosin, respectively. Also, was not detected effects of the interactions between CL presence on day -10 and treatment, neither the effects of the interaction between parity and treatment. It is concluded that the 3mL dose of Ciosin do not increase the percentage of cows with CL on day 7 after TAI, neither increase P/AI in cows synchronized with protocol based on progesterone and estradiol.

Animal Reproduction

AI and IATF

Evaluation of treatment response to benzoate or 17ß-estradiol (associated with P4) at the beginning of TAI protocol

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The objective of this study was to evaluate the pregnancy rate of cows that received 17ß-estradiol associated with progesterone (P4) as treatment at beginning of the ovulation synchronization protocol to TAI. On a random day of estrous cycle (D0), primiparous (n=400) of Nelore breed (Bos indicus) with 43±7.1 days postpartum, body condition score (BCS) and an average weight of 2.9±0.4 (1-5) and 414±40 Kg, respectively, were randomly distributed in two different treatments: Group 17ß+P4 (5.5mg 17ß-estradiol associated with 50mg of P4; n=204) and Group BE (2 mg BE; n=196) im. The cows received an intravaginal device with 0.96g of P4 (Progestar®, Boehringer Ingelheim, Campinas, Brazil) and were evaluated by transrectal ultrasonography to measure dominant follicle (DF) diameter and record the presence of CL for randomization of groups. On Day 8.5 (D8.5) the device was removed and cows received 1mg estradiol benzoate (Estrovulinn®, Boehringer Ingelheim, Campinas, Brazil), 0.530mg sodium cloprostenol (Cioprostinn®, Boehringer Ingelheim, Campinas, Brazil) and 300IU of eCG (Folligon®, MSD Animal Health, Sao Paulo, Brazil) im. Also, tail chalk was applied on on sacrococcygeal region for estrus detection. On day 10 (D10), 48h after device removal, cows were checked for occurrence of estrus and measurement of DF, and subsequently, artificially inseminated. The pregnancy diagnosis was realized 31 days after TAI. Statistical analyzes were performed using GLIMMIX procedure of SAS®. There was no difference (P=0.32) in pregnancy rate according to experimental groups (17ß+P4= 54.41% [111/204] vs. BE= 57.65% [113/196]). No interaction was observed between pregnancy*estrous (P=0.95) and pregnancy*BCS (P=0.16). However, there was a tendency for interactions pregnant*diameterDF (DF<11mm: 17ß+P4= 29.41% [10/34] vs. BE= 46.88% [15/32]; P>0.05 and FD≥11mm: 17ß+P4= 70.45% [31/44] vs. BE= 57.14% [28/49]; P=0.058) and pregnancy*weight (<400 Kg: 17ßP4= 57.14% [40/70] vs. BE= 46.15% [30/65]; P=0,07 and ≥400 Kg: 17β+P4= 52,5% [63/120] vs. BE= 63,2% [74/117]; P>0.05). It was concluded that animals with higher DF (≥11 mm) and lower weight (<400 Kg) showed a tendency towards higher pregnancy rate when treated with 17ß-estradiol associated with P4 at the beginning of TAI protocol.Aknowledgments: Boehringer Ingelheim Animal Health.

Animal Reproduction

ABSTRACTS: 34TH ANNUAL MEETING OF THE BRAZILIAN EMBRYO TECHNOLOGY SOCIETY (SBTE)

AI and IATF

Effects of some factors on gestation period in the girolando herd of pesagro-rio (1990-2019)

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In dairy cattle, service and gestation periods, pregnancy rates, service per conception, age at first parturition and calving interval are the parameters used to assess reproductive efficiency. Among them, the gestation period (GP) has its importance relegated to a secondary plan, in most of the farms due to its small variability. However, when analyzing fetal losses, there are high rates of stillbirths and abortions, affecting the birth rate in some dairy farms. This event highlights the need for a schedule of calving predictions for the herd. For this, technical knowledge of the gestation period of the exploited breed is fundamental for the management of births, in order to provide adequate assistance to dystocic births and avoid losses due to mortality in the peripartum. In order to evaluate the effects of some variables on the gestation period (GP), 1990 to 2019, were used the records of the Girolando herd in Geraleite software, The PESAGRO-RIO/CEPAO bovine herd is formed by crosses between the Holstein (H) and Gir (G) breeds, to form the Girolando race. The reproductive system adopted has been traditional artificial insemination with observation of estrus and FTAI, with frozen semen, in two annual seasons, in order to meet the research projects.The data related to the GP (day) of Girolando cows, according to (1) calving year, (2) sex, (3) maternal blood group, (4) paternal race, and (5) calving order, were submitted to descriptive analyzes, one-way anova and Bonferroni test at 0.05% (Bioestat Software). The results of GP (mean±standard deviation in day/Calving number) were: 1) Calving year ($p>0.05$): General average from 1990 to 2019 = 282.9±6.5/2104); 2) Sex of the calf ($p>0.05$): a) Female 282.5±6.5/982 and b) Male 283.5±6/1122; 3) Maternal blood group ($p>0.05$): a) 01 to 20% of Holstein = 282.7±6.3/115; b) 21 to 40% of Holstein = 282.4±6.5/436; c) 41 to 60 % of Holstein = 282.3±6.6/442; d) 61 to 80% of Holstein = 283.4±7.5/592; and e) 81 to 100% of Holstein = 284.5±6.9/519); 4) Paternal breed ($p>0.05$): a) Holstein = 282.2±6.1/1118; b) 5/8 Holstein = 284.4±7.1/65; c) G = 285.3±6.9/596; and d) Unidentified breed = 282.7±8.9/325; 5) Birth order ($p>0.05$): 1^{st} = 282.5±6.4/602; 2^{nd} = 283.1±6.6/505; 3^{rd} = 282.8±6.5/378; 4^{th} = 283.9±6.8/265; 5^{th} = 284.0±6.5/178; 6^{th} = 284.8±7.01/103; 7^{th} = 282.9±7.0/46; 8^{th} = 283.0±4.9/19; and 9^{th} = 282.8±6.8/8. The results did not show significant effects the factors on the gestation periods. Thus, the general average of 282.9±6.5 days can be considered an estimated calving prediction for the Girolando herd, aiming to assist in the management and adequate care of dystocic calving, reducing mortality in the peripartum. Keywords: reproductive evaluation, dairy herd, gestation length

Animal Reproduction

AI and IATF

Treatment with levamisole phosphate (Biopersol®) at the onset of TAI protocol improves reproductive and productive performance of Nelore females

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The aim of this study was to evaluate the effect of using levamisole phosphate at the onset of a TAI protocol on reproductive and productive outcomes of beef females. A total of 655 Nelore females (551 cows with 30-55 DPP and 94 heifers aging 18-24m old), with average BW 412.3±2.8kg and BCS 2.91±0.02 (1-5 point scale) from a farm in MT State, Brazil were used. At random days of the estrous cycle (D0), cows and heifers were evaluated by ultrasonography for uterine condition and cyclicity (CL). Cows received a Cronipres® Mono Dose intravaginal device with 1g P4 (Biogénesis Bagó, Curitiba, Brazil), 2mg estradiol benzoate (EB; Bioestrogen®, Biogénesis Bagó), 150µg D-Cloprostenol (PGF, Croniben®, Biogénesis Bagó) IM and were homogenously allocated to receive or not 2.363g (10mL) of levamisole phosphate (Biopersol®, Biogénesis Bagó) SC. On D8, device was removed and 150µg D-Cloprostenol, 300IU eCG (Ecegon®, Biogénesis Bagó) and 1mg estradiol cypionate (EC; Croni-Cip®, Biogénesis Bagó) IM were given. TAI was done 48h after device removal, concomitant with 10.5µg buserelin acetate (GnRH; Gonaxal®; Biogénesis Bagó) IM. Heifers were treated with similar protocol, except for the doses of EB (1mg), PGF (75µg), eCG (200IU), EC (0.5mg), and 1.890g (8mL) of levamisole phosphate. Pregnancy diagnosis was done 30d after TAI in all females and 60d after TAI in a subset. Fecal samples were collected from 82 cows on D0 for counts of eggs per gram (EPG) and oocysts per gram (OoPG) of feces. Follicular dynamics was assessed on D8 and D10 in 277 cows. Data was analyzed using SAS. Same EPG (27.5±7.2 vs 33.3±9.0; P=0.53) and OoPG (40.0±10.8 vs 65.5±25.3; P=0.67) were found for control and treated cows before treatment. On D0, 30.7% of cows and 88.3% of heifers had a CL. Any effect of treatment was found for luteolysis rate between D0-D8 [64.3% (18/28) vs 78.1% (25/32); P=0.24], diameter of the dominant follicle on D8 [12.9±0.3 vs 12.3±0.2; P=0.21] and D10 [14.2±0.3 vs 13.8±0.2; P=0.36], follicular growth [0.86±0.07 vs 0.84±0.06mm/d; P=0.29], estrus occurrence D8-D10 [71.2% (232/326) vs 72.2% (236/327); P=0.36], P/AI [52.9% (173/327) vs 55.5% (181/326); P=0.53], and pregnancy loss 30-60d [6.3% (5/79) vs 4.6% (4/87); P=0.56; control vs treated cows, respectively]. However, lesser tendency of occurrence of ovulation between D0-D8 [6.5% (9/139) vs 2.2% (3/138); P=0.09] and presence of CL on D8 [15.8% (22/139) vs 8.7% (12/138); P=0.07] was found for cows treated with levamisole. Mostly, the cumulative pregnancy rate (TAI+ bull) 60d after TAI was greater for treated [82.5% (113/137)] vs control [72.9% (102/140); P=0.04] females, and the BW tended to be greater for treated than control ones 30d after TAI (442.7±4.3 vs 431.3±3.8kg; P=0.06) even though they were similar on D0 (413.3±2.8 vs 410.2±2.8kg;P=0.39). Thus, the use of levamisole phosphate on D0 of a TAI protocol improved cumulative pregnancy rate (TAI+ bull) and BW of Nelore cattle. Credits: Farm São José.

Animal Reproduction

AI and IATF

Serum profile of different doses of injectable progesterone in Bos indicus cows submitted to a timed artificial insemination protocol

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The objective was to evaluate the serum profile of different doses of injectable progesterone (P4i; Sincrogest Injetável®, Ourofino, Brazil) in *Bos indicus* cows submitted to a timed artificial insemination (TAI) protocol. In this study, 53 suckled multiparous Nelore cows with a body condition score (BCS) of 2.75 ± 0.2 (scale from 1 to 5) and body weight of 423 ± 47 Kg were used. Cows were divided into five experimental groups with different doses of P4i (P4i60mg, n = 9; P4i105mg, n = 11; P4i150mg, n = 12; P4i195mg, n = 11 and P4i240mg, n = 10). All cows received a dose of PGF two days before TAI protocol was initiated (D-2). On D0, 2mg of estradiol benzoate i.m. (Sincrodiol®, Ourofino, Brazil) and different doses of P4i i.m. (60mg, 105mg, 150mg, 195mg and 240mg) were administered. Eight days later (D8), 1mg of estradiol cypionate i.m. (SincroCP®, Ourofino, Brazil), 300UI of eCG i.m. (SincroeCG®, Ourofino, Brazil) and 500 µg of Cloprostenol i.m. (Sincrocio®, Ourofino, Brazil) were administered. Daily blood samples (D0 to D12) were collected to determine the serum progesterone (P4) profile. The P4 concentration peak occurred one day after treatments (D1) for different doses of P4i administered (P4i60mg = 0.7±0.2 ng/mL, P4i105mg = 1.6±0.2 ng/mL, P4i150mg = 2.5±0.4 ng/mL, P4i195mg = 2.9±0.4 ng/mL and P4i240mg = 3.0±0.4 ng/mL). After the P4 peak, there was a gradual decrease in the progesterone concentration for all P4i doses. The P4i105mg group had P4 concentration below 1ng / mL on D2 (0.8 ± 0.2 ng / mL) and the other groups on D3 (P4i150mg = 0.5 ± 0.1 ng / mL; P4i195mg = 0, 6 ± 0.1 ng / mL and P4i240mg = 0.7 ± 0.1 ng / mL). All groups, except P4i 240mg, had concentrations of P4 below 0.5ng / mL in D6. However, all groups remained below 1ng / mL until the end of the study (D12). The concentrations obtained in groups P4i60mg, P4i105mg, P4i150mg, P4i195mg and P4i250mg were on D8 (0.09±0.08ng/mL, 0.20±0.06ng/mL, 0.41±0.10ng/mL, 0.50±0.05ng/mL, 0.86±0.11ng/mL) and D10 (0.10±0.02ng/mL, 0.11±0.02ng/mL, 0.37±0.15ng/mL, 0.29±0.07ng/mL, 0.37±0.08ng/mL). In conclusion, the peak and the decrease in serum P4 concentrations were dose dependent and occurred on D1 after the administration of treatments.Support: Ourofino Saúde Animal, Capes

Animal Reproduction

AI and IATF

Increasing 12 h of intravaginal device permanence allows greater estrus synchrony but lower pregnancy rates in artificially inseminated acyclic dairy goats

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This study assessed in dairy goats the effect of hormonal induction treatments varying only 12 h in progestogen device permanence on estrus synchrony and fertility. In both Experiments, goats received 60 mg of medroxyprogesterone acetate (MAP) intravaginal devices for 6 (G6) or 6.5 d (G6.5), and 200 IU of eCG i.m. and 30 µg of d-cloprostenol were i.m. applied at 24 h or 36 h before device removal, respectively. In Experiment 1 (n = 24), data related to sexual behavior and ovarian ultrasonography were recorded and, in Experiment 2, (n = 83) fertility was assessed after Flexible Time Artificial Insemination (FxTAI). Parametric variables were subjected to one-way ANOVA and nonparametric variables were analyzed by the chi-square test; P value <0.05 was considered as significant. Estrus response rate was similar (P>0.05) between G6 (92%) and G6.5 (100%). The interval from device removal to estrus was shorter (P<0.05) in G6.5 (34.7 ± 1.8 vs. 44.4 ± 3.3 h). At the time of removal of the device, animals with follicles larger than 6 mm represented 54% (13/24) of the goats, being 31% (4/13) and 69% (9/13) from G6 and G6.5, respectively (P = 0.057). In comparison with goats with follicles larger than 6 mm, those animals with follicles smaller than 6 mm had longer (P <0.05): follicular wave emergence after sponge removal (78.8 ± 4.7 vs. 62.6 ± 3.3 h), interval from device removal to estrus (43.8 ± 2.5 vs. 35.9 ± 2.9 h) and to ovulation (81.1 ± 4.8 vs. 67.1 ± 3.4 h) and follicular growth rate per day (0.9 ± 0.1 vs. 0.6 ± 0.1 mm / day). The association of G6.5 with the number of goats presenting follicles larger than 6 mm affected the ovulatory follicle and/or the formation and maintenance of the corpus luteum (CL), resulting in lower fertility. The largest ovulated follicle, the second largest and the interval from device removal to ovulation were similar (P> 0.05) between groups. According to the doppler ultrasound exams, luteal dysfunction [partial (only 1 CL) or total regression] increased progressively from day 3 (100.0 vs. 100.0%) to 7 (100.0 vs. 83.3%), 10 (100.0 vs. 75%), 13 (100.0 vs. 66.6%), 17 (63.6 vs. 41.6%) and 21 (50.0 vs. 33.3%) to G6 and G6.5 treated goats, respectively. This dysfunction was related to the CL presence and blood perfusion. The G6.5-goats had greater estrus synchrony, but a lower (P<0.05) conception rate (53 vs. 83%). In conclusion, increasing the time of intravaginal device permanence from 6 to 6.5 d resulted in greater estrus synchrony, but lower fertility in dairy goats subjected to estrus induction treatment at the non-breeding season. The G6 treatment resulted in high conception rate and can be recommended to support FxTAI programs in dairy goats in the non-breeding season. Keywords: AI; Anestrus; Caprine; Corpora Lutea; Follicular Dynamics; Ultrasound. Promotion Institutions: CNPq; Projects 310166/2012-8 and 479826 2013-7, Fapemig; Project CVZ-PPM 00201-17 and EMBRAPA Project 20.19.01.004.00.00.

Animal Reproduction

AI and IATF

Conception rates may be reduced in cows receiving PGF2α at the moment of AI when high estrus expression is displayed

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Estrus behavior at timed-AI is related to higher ovulation rates and increased fertility (Pereira et al.,2016). PGF2α assists the mechanisms involved in the ovulatory process (Pfeifer et al.,2014) which could benefit cows with reduced estrus expression. However, it is not known whether the treatment of PGF could interfere in the pregnancy rates of animals with high estrus expression The objective of this study was to evaluate the effect of PGF2α treatment at the moment of AI on conception rate (CR) of animals displaying diferentt grades of estrus behavior. Data from timed-AI outcomes of 621 multiparous Nelore cows separated into four lot of animals of the same farm (CEUA UFMG 348/2018) were collected. On Day 0 (D0), the animals received an intravaginal progesterone (P4) implant (Sincrogest®) and 2mg of estradiol benzoate (Sincrodiol®). On D8, 12.5mg of Dinoprost (Lutalyse®), 1mg of estradiol cipionate (SincroCP®) and 300UI of eCG (Sincro eCG®) were injected and P4 implant was removed. In addition, estrus detection device (Estrotec®) was applied on the base of the tail. Timed-AI was performed 48 h after P4 implant removal (D10), by two experienced AI technicians, using semen from two bulls equally distributed among the lots. Moreover, on D10 (moment of AI), the animals randomly received 12.5mg i.m. of Lutalyse® (PGF group) or did not receive any treatment (control group). Expression of estrus was recorded with a scale 1 to 4 (grade1= ≤25% removal of gray protective paint; grade2=≤50% removal of gray paint; grade3=≤75% removal; grade4= >75% removal) (Pohler et al.,2016). Animals presenting estrus expression 1 and 2 were considered with low heat expression and animals with grades 3 and 4 were considered presenting high estrus. CR was analyzed by logistic regression, considering P>0.05 and <0.10. Total CR was 52%, being higher (P=0.001) for cows expressing evident estrus (CR=59%; n=379) than for cows that expressed lower evidency of estrus behavior (CR=44%, n=242). No effect of lot (P=0.332), bull (P=0.413) and AI technician (P=0.166) were observed but interaction between lot and PGF2α treatment was detected (0.004). An tendency (P=0.093) for interaction between estrus expression and PGF2α treatment was observed [No estrus+PGF= 42% (n=118) A; No estrus+No PGF= 47% (n=124) A; Estrus+PGF= 53% (n=197) AB; Estrus+No PGF= 63% (n=182) B] suggesting that PGF2α at AI moment can cause some detrimental effects in CR of cows presenting high estrus expression. It was concluded that animals expressing more evident estrus behavior present higher conception rate but the application of PGF2α at the time of AI may reduce the physiological advantages of these animals. Fertility of cows with low estrus expression was not benefited by the application of PGF2α at the moment of AI. Acknowledgments: Agropecuária Sta.Bárbara. References: Pereira et al.,J.DairySci.99:2237-47,2016; Pfeifer et al., Theriogenology 81:689–95,2014; Pohler et al.,Theriogenology 85:1652-59,2016.

Animal Reproduction

AI and IATF

The application of PGF2α at the moment of timed-AI did not increase the success of pregnancy in cows inseminated with sexed semen

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Beef cows inseminated with sexed semen generally present unsatisfactory results at timed-AI (Sá Filho et al., 2010). Sexed sperm presents higher capacitation (Lu et al., 2004) and thus cannot long remain waiting for ovulation occurrence. Because the application of PGF2a seems to improve the mechanisms of ovulatory process (Pfeifer et al., 2014) we hypothesized that PGF2a at the moment of AI may improve the fertility of cows inseminated with sexed sperm. The study aimed to assess the effect of PGF2a administration at the moment of AI on conception rates (CR) of cows submitted to timed-AI inseminated with conventional (frozen-thawed semen) or sexed semen (X-sorted frozen-thawed semen). A total of 355 multiparous (40 days postpartum) Nelore cows were inseminated with the same timed-AI protocol, in a farm in the state of Pará, Brazil. On Day 0 (D0), the animals received an intravaginal progesterone (P4) implant (CIDR®, Zoetis, São Paulo, Brazil) and 2mg of estradiol benzoate (Gonadiol®, Zoetis). On D7, 12.5mg of Dinoprost (Lutalyse®, Zoetis) was applied. On D9, 12.5mg of Dinoprost (Lutalyse®), 1mg of estradiol cipionate (ECP®, Zoetis) and 300UI of eCG (Novormon®, Zoetis) were injected and P4 implant was removed. The timed-AI was performed 48 h after P4 implant removal (D11), by two experienced technitians being intercalated every 20 animals. Conventional (CS) and sexed semen (SS) from two different Nelore bulls were used. For every 5 inseminated cows, the type of semen was alternated (CS ou SS) and for every 10 inseminated animals the bull was switched. Moreover, at D11 (moment of AI), the animals randomly received 12.5mg i.m. of Lutalyse® (PGF group) or 2.5mL i.m. of saline solution (control group). At 30 days after timed-AI, pregnancy diagnosis was assessed by ultrasonography. TheCR was analyzed by logistic regression, considering P>0.05. No effect of AI technitian (P=0.130) and no bull effect were detected (P=0.535). No interaction of bullXsemen was observed [Bull1/CS=51.1% (n=94), Bull1/SS=29.2% (n=89), Bull2/CS=57.0% (n=86), Bull2/SS=31.4% (n=86); P=0.0429]. Additionally, no effect of treatment was observed (control=40.5%, n=173; PGF=44.0%, n=182; P=0.523) but an effect of semen was detected where CR of animals inseminated with CS (53.8%, n=180) was higher (P<0.001) than CR of animals inseminated with SS (30.3%; n=175). No interaction between treatment and semen was observed (P=0.804). For CS, the following CRs were observed: Control=54.8% (n=73) and PGF=53.3% (n=107), (P=0.861). For SS the following CRs were observed: Control = 30.0% (n=100) and PGF = 31% (n=75), (P=0.939). It was concluded that sexed semen presented lower fertility and the application of PGF2α at the moment of timed-AI did not increase pregnancy rates for this type of semen. References: Lu et al., Theriogenology62:819-30, 2004; Pfeifer et al., Theriogenology 81:689–95,2014; Sá Filho et al. Theriogenology 74:1636–42, 2010. Acknowledgments: Agropecuária Sta.Bárbara and Zoetis.

Animal Reproduction

AI and IATF

Comparison of the preovulatory follicular dynamics induced by kisspeptin or eCG in ewes

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The production of eCG is under debate due to animal welfare concerns. The neuropeptide kisspeptin (Kp) may be a viable alternative as it increases the secretion of GnRH, FSH and LH, but has the limitation of the short half-life. The aim of this study was to compare the preovulatory follicular development induced by C6, a Kp analogue with a longer half-life, and eCG in ewes. The study was performed in February (mid-summer) with 30 cyclic Corriedale multiparous ewes (2-4 years old). The estrus was synchronized using intravaginal devices (IVD) with medroxyprogesterone acetate (60 mg) for 14 days. The experimental groups were: 1) eCG group (n = 10) received 300 IU eCG (Novormon, Syntex, Buenos Aires, Argentina) at IVD withdrawal; 2) Group C6 (n = 10) received 15 nmol of C6 24 h after removal of the IVD; and 3) control group (n = 10), were only treated with the IVD. The ovaries were scanned with ultrasound every 12 h, from the withdrawal of the IVD to ovulation. In each examination, the number and diameter of the ovarian follicles were recorded. Estrus behavior was controlled with androgenized wethers every 12 h during the same period. The number of corpora lutea (CL) and their functionality were classified according to vascularization using B-mode and Doppler ultrasound (Cosentino et al., Theriogenology, 121:104-11, 2018) 11 days after IVD removal. More eCG and control than C6 ewes came into estrus (10/10 and 7/10 vs 2/10; P<0.0001). The time from of IVD withdrawal to ovulation did not differ between groups (control: 57.0±2.84; eCG: 60.1±2.69; C6: 52.1±2.84; P = 0.14). However, the ovulations were better synchronized in C6 than eCG and control ewes (SD: 4.1, 10.7, and 8.3, respectively, P = 0.016). The frequency of ewes with more than 1 CL was greater in C6 (7/10) than eCG (7/9) and control (2/10) (P = 0.002). The ovulatory follicles of eCG and C6 ewes were smaller than those of control ewes (6.4 ± 0.2 mm and 7.0 ± 0.6 vs 7.1 ± 0.8; P = 0.05). However, there were no differences in the quality of the corpus luteum, assessed in B-mode (P = 0.94), nor in vascularization of the CL, Doppler mode (P = 0.43). In conclusion, C6 can be used to synchronize ovulation in ewes as stimulates similar follicular development as eCG and synchronizes ovulation better.

Animal Reproduction

AI and IATF

Association between sperm hyperactivation and estrus on fertility in timed AI postpartum cows

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The objective of this study was to evaluate the effect of sperm hyperactivation on fertility of cows that displayed estrus or not in timed AI postpartum beef cows. In Experiment 1, 24 Nelore cows were included in a estradiol- progesterone based protocol ([2 mg BE (Gonadiol®, Zoetis)+ CIDR® (Zoetis) on D0 / 2 mL PGF (Lutalyse®, Zoetis) + 0,6 mg ECP (E.C.P.®, Zoetis) – CIDR® on D8 / TAI 48 h] and a estrus detector device (Estrotect®) were placed in the sacrocaudal region. After CIDR removal, the dominant follicle was monitored by ultrasonography every 12 h until the ovulation. After pregnancy detection, 12 non-pregnant cows were resynchronized with the same protocol. Therefore, cows were separated according to the moment of estrus detection in 2 groups: 1) Estrus 48h (n =19), cows that display estrus, and No Estrus (n =17), cows that did not display estrus until 48 h after CIDR removal. Cows that were detected in estrus before TAI ovulated earlier (P<0,02) than cows not detected in estrus (73.3±13.1 vs. 95.5±37.5h). In Experiment 2, 417 postpartum Nelore cows were subjected to the same TAI protocol of the Experiment 1. For TAI procedure, 2 batches from each of the 2 bulls were used. Batches were analyzed by CASA and were classified, according to the curvilinear velocity (VCL), amplitude of lateral head displacement (ALH), and linearity (LIN), as follows: Hyper-activated (H+; LIN < 50.6%, ALH > 7 µm and VCL > 149 µm/s), and Non-Hyper-activated (H-; LIN > 55.5%, ALH < 6.3 µm and VCL < 142.5 µm/s) as described previously (Pfeifer et al. 2019): H+ and H- semen from each of the chosen bulls were distributed homogeneously into the estrus group of cows (Estrus at 48 h, E; and Non Estrus, NE). Thus, after distribution, the groups were: EH+ (n=132), EH- (n=132), NEH+ (n=98), and NEH- (n=55). There were no effects of sperm kinematics (P = 0.19) and interaction between sperm kinematics and estrus (P = 0.42) in the pregnancy per AI (P/AI). However, there was an effect of estrus expression (P<0.01). The EH+ (60.6%, 80/132) and EH- (68.2% 90/132) groups had a greater P/AI (P<0.02) in comparison to the NEH+ groups (50.0 %, 49/98) and NEH- (49.1%, 27/55). Cows that displayed estrus within 48 hours after P4 removal ovulate earlier and are more likely to become pregnant than cows that did not display estrus. However, sperm kinematics did not affect the fertility of beef cows regardless of the estrus occurrence. Acknowledgments: EMBRAPA, CAPES and Seleon Biotechnology.Keywords: estrus detection, sperm kinematics, moment of ovulation.

Animal Reproduction

AI and IATF

The use of progestogens preceding the early pregnancy diagnosis and its effects on caprine corpus luteum

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The corpus luteum (CL) is a temporary endocrine gland of highly importance in the pregnancy maintenance, especially for goats, since ewes and cows may sustain the late pregnancy without CL. Therefore, studies in which exogenous P4 administration is used for resynchronization protocols and its concerns on goat's CL at early pregnancy are needed. The aim of this study was to evaluate the effect of exogenous P4 from day 16 to 21, in goats with unknown pregnancy status, to predict its effect and use it to avoid ovulation on a subsequent early resynchronization protocol. In this sense, intravaginal sponges containing 60 mg of medroxyprogesterone acetate (Progespon; Schering Plough, SP, Brazil) were used for 6 days in 54 does. One day before sponge withdrawal, 200 IU of equine chorionic gonadotropin (eCG, Folligon, MSD, São Paulo, Brasil) and 0.24 mg of cloprostenol sodium (Estron, Agner União, São Paulo, Brazil) were administered intramuscularly (i.m.). The does received 0.025 mg of lecirelin (GnRH – Gestran Plus, Tecnopec, São Paulo, Brasil) 34h after sponge withdrawal and artificially inseminated with commercial frozen semen 18h after it. From 16th to 21st day of the following estrus cycle, the does received a new P4 device and had blood samples daily collected. At 21st day, all goats were early diagnosed for non-pregnancy by ultrasonographical luteal blood flow assessment and confirmed at Day 30 by the presence of conceptus (Sonoscape S6, Shenzhen, China – 7.5 MHz rectal transducer). Serum P4 values from 26 animals (G_{NPSP} – Group of non-pregnant does with second sponge – n=8; G_{NPNSP} – Group of non-pregnant does without second sponge – n=6; G_{PSP} – Group of pregnant does with second sponge – n=5; G_{PNSP} – Group of pregnant does without second sponge – n=7) was determined by radioimmunoassay using commercial kits (MP Biomedicals, LLC, Diagnostics Division, Orangeburg, NY, USA). P4 values were analyzed ($p<0,05$) separately for goats pregnant or not pregnant using a mixed model procedure including the group (treated or not treated – Friedman test), time (Student's T-test), and their interaction as main effects in the model (Mann-Whitney test). No P4 differences were found between groups (GNPSP – 3.1 ± 2.8; 1.7 ± 1.8; 0.4 ± 1.0; and 0.0 ± 0,0 vs. GNPNSP – 4.4 ± 1.8; 3.0 ± 2.2; 0.8 ± 0.8; and 0.0 ± 0.0 or GPSP – 4.2 ± 1.0; 3.4 ± 0.6; 3.3 ± 1.6; 3.2 ± 0.9; 3.6 ± 1.2; 3.5 ± 1.3; 2.7 ± 1.3 vs. GPNSP – 4.4 ± 1.6; 3.6 ± 1.5; 3.7 ± 1.5; 3.8 ± 1.4; 3.2 ± 1.2; 3.1 ± 1.2; 3.6 ± 1.1; D16. D17. D18. D19; D20; D21; D24 respectively) or for the interaction of group and time. From the 14 pregnant diagnosed does at D21, 13 were confirmed at D24 (93%), and 12 at D30 (86%), both from group without second sponge (22% of pregnancy – 21/54). In conclusion, there was no effect of a second P4 device on luteolysis or early pregnancy from the first estrous cycle after FTAI, and, therefore, a second P4 device can be used for resynchronization protocols in goats. AcknowledgmentsFAPERJ; CAPES

Animal Reproduction

AI and IATF

Effect of the delay in the artificial insemination in cows with less ovulatory competence

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The objective of the study was to evaluate the effect of the 24-hour delay in the artificial insemination procedure in females with low ovulatory potential in FTAI. In this study, lactating Nelore cows (Bos indicus; n = 236), with 430-640 kg body weight, were submitted to a TAI protocol with two moments of insemination (Block 0 = cows with follicles ≥13 mm; Block 1 = cows with follicles ≤12.9). On Day 0, cows were given a progesterone device (1.9g Progesterone; CIDR®, São Paulo, Brazil) plus 2 mg of estradiol benzoate (Bioestrogen®, Curitiba, Brazil) i.m, to synchronize follicular waves. On Day 8 they received 12.5 mg of Dinoprost i.m. (PGF2α-analogue; Lutalyse® Pfizer, Cravinhos, Brazil), 1 mg Estradiol Cypionate (ECP® Pfizer, Cravinhos, Brazil), and 300 IU of Equine Chorionic Gonadotropin (Novormon®, Syntex, Buenos Aires, Argentina) and had the CIDR device removed. On Day 10 (07:00 a.m.), the diameter of the pre ovulatory follicle (POF) was assessed by ultrasonography, and cows were randomized into Control (n = 121) and Block (n = 115) Groups. Control-Group cows were inseminated 48 h after CIDR removal, and Block-group cows were inseminated at two time points according to the diameter of the POF as described by Pfeifer et al. (Animal Reproduction Science, 163: 89-96, 2015): B0 (POF ≥ 13 mm, TAI at 08:00 am on Day 10, n = 52), and B1 (POF ≤12.9 mm, TAI at 08:00 am on Day 11, n = 63). Further ultrasound examination was performed 30 days after AI to detect pregnancy. The pregnancy per artificial insemination (P / AI) did not differ between the Control and Block groups (P = 0.8). Cows in the block group had P / AI of 56% (64/115) vs. 54% (66/121) of the control group. Cows from block group inseminated at 48 h after CIDR removal had higher P / AI than cows inseminated 24h later (69% vs. 44%; P <0.01). There was no difference in P / AI between females in the B1 group (POF ≤12,9 mm) and females in the Control group who had follicles ≤12.9 mm (44% vs. 47%; P = 0.8). In conclusion, the results of the present study demonstrate that the delay of artificial insemination in 24 hours in females with follicles ≤12.9 mm did not increase the pregnancy rate in timed AI cows.Keywords: Follicular diameter. Fertility. Ovulation. Ultrasonography.Acknowledgements: This research project was supported by CNPq (Project 407307/2016-8).

Animal Reproduction

AI and IATF

Effect of long-acting progesterone use at early diestrus on pregnancy maintenance in beef and dairy recipient cattle

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We tested in the present study the hypothesis that supplementation with long-acting P4 (iP4) at different times of the initial diestrus improves the pregnancy rates in dairy and beef recipients submitted to timed-embryo transfer (TET). Recipients in good body condition score (BCS) had their estrous cycle synchronized with E2/P4 based protocol in three experiments (Exp. 1 to 3). On Day -10, all animals received an intravaginal P4 device (1g) and an im administration of 2mg estradiol benzoato. After 8 days, the P4 devices were removed and 0.530mg PGF2α, 300IU eCG and 2mg estradiol cypionate were im administered. In Exp. 1, dairy heifers (n=76) and lactating dairy cows (n=104) were randomly assigned to 2 experimental groups: C group (n= 89) and iP4D4 Group (n= 91). For Exp. 2 and 3, suckled beef recipients were used. In Exp. 2, recipients were assigned in two experimental groups: C group (n= 147) and iP4D7 group (n= 144); whereas, in Exp. 3 recipients were randomly assigned in three experimental groups: C group (n= 85), iP4D4 group (n= 86) and iP4D7 group (n= 81). Recipients in the iP4D4 and iP4D7 groups received an im administration with 150mg long-acting iP4 (Sincrogest®, Ourofino, Cravinhos, Brazil), respectively, on Days 4 or 7 (day of ET). Nine days after P4 device withdrawal (Day 7), all recipients were evaluated by transrectal ultrassonography (US) and those bearing an well developd CL received an fresh or vitrified IVP embryo from a commercial laboratory. In Exp. 2 and 3, the CL area was also determined by US at the time of TET. The pregnancy diagnosis was performed by US on 30 days of pregnancy in Exp. 1 and Exp. 2, and on 30 and 60 days in Exp. 3. The data were analyzed using a logistic regression model by PROC GLIMMIX (SAS). In Exp. 1, pregnancy rate did not differ (P>0.1) between the C group (38.2% [34/89]) and iP4D4 group (49.5% [45/91]); however, a parity effect indicated a greater (P<0.05) pregnancy rate in heifers (57.9% [44/76]) than cows (30.8% [32/104]). In Exp. 2, the pregnancy rate was greater (P<0.05) in the iP4D7 group (45.0% [65/144]) than in the C group (34.0% [50/147]). Also, a tendecny for greater (P=0.08) pregnancy rate was observed for recipients with small CL (≤2.75 cm²) that were treated with iP4 on Day 7 than the control recipients (46.4% [32/69] vs. 32.6% [28/86]). In Exp. 3, no signifficant effects (P>0.1) of treatment group or CL size were detected on pregnancy rates at days 30 and 60. Pregnancy rates in the C, iP4D4 and iP4D7 groups were, respectively, 49.4% (42/85), 53.5% (46/86) and 56.8% (46/81). In conclusion, the benefitial effects of long-acting iP4 supplementation at early diestrus on pregnancy maintanence may vary according to the experimental conditions, but its use at the moment of FTET is an interesting alternative to enhance fertility of beef recipients, specially in those with a small CL. Acknowledgments: FAPESP (2012/04004-8) and Ourofino Saúde Animal and Tecplan companies

Animal Reproduction

AI and IATF

Body condition score index (BCSi): a new approach to evaluate the effect of BCS on fertility of timed AI beef cows

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Visual evaluation of body condition score (BCS) is a categorical variable that is widely accepted as an important tool for subjective quantification of endogenous energy reserves in cattle. The objectives of this study was to develop an index to estimate the relationship between BCS and fertility. In this study, 2321 lactating Nelore cows were enrolled from 29 cohort of cows enrolled in TAI programs in Mato Grosso do Sul and Rondonia states, Brazil. All females were evaluated according to BCS (scale 1 to 5) and were included in a estradiol-progesterone based timed artificial insemination (TAI) protocol. All cows were marked with chalk and estrus score was evaluated (scale 1-3) at TAI. The fertility of each TAI protocol was categorized according to the pregnancy per AI (P/AI) in low fertility group (<45%, LFG), moderate (45 – 60%, MFG) and high (>60%, HFG). To calculate the BCSi for each group, it was necessary to evaluate whether the BCS of each cow is in accordance with the target BCS (3 to 4) for beef cows in the breeding season. Therefore, three variables were calculated: 1) Difference between real BCS and the target BCS (DBCS) by the formula, ; 2) DBCS was transformed in a percentage value, called level of BCS adequate (LBA), by the formula, ; and 3) The adequate BCS rate (ABR), in which is considered an binomial variable and, therefore, cows with BCS on target received 100% of ABR and cows, which BCS was not on target, received 0% of ABR. Finally, the average between LBA and ABR resulted in the BCSi. Anova was used to compare BCSi, BCS, and estrus score among groups of fertility. Tukey's test was used to compare means among groups. There was difference in the BCSi among fertility groups ($P<0.05$). The HFG had greater BCSi (85.7%), than MFG (81.9%) and LFG groups (76.7%). HFG and MFG had greater ($P<0.05$) BCS than LFG (3.14, 3.08, and 2.98, respectively). Finally, cows from HFG displayed more estrus ($P<0.05$) than cows from LFG (2.3 and 2.2). The estrus score of cows from MFG did not differ from the other groups (2.21). These results demonstrate that the BCSi represents a novel approach to deal with the BCS categorical variable and that is associated with fertility of TAI programs.

Animal Reproduction

AI and IATF

Adjustments in fixed-time AI to benefit Bos indicus sires with lower fertility

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This study aimed to investigate strategies to improve pregnancy per AI (P/AI) of sires with known lower (L) field fertility. The strategies consisted of adjusting the timing of ovulation GnRH-induced, or by inseminating simultaneously with two straws of semen during fixed-time AI (FTAI) protocols. The main hypothesis was that anticipating treatment with GnRH in relation to FTAI, but not doubling the number of straws would improve P/AI of L sires. Therefore, 1167 Nelore cows (BCS=2.9±0.4) were submitted to a protocol with 1g progesterone (P4) implant and 2mg estradiol benzoate (EB) on D0 and P4 removal, 1mg estradiol cypionate (EC), 0.5mg cloprostenol (PGF) and 300IU eCG on D8. Cows had their tailhead painted on D8 for estrus detection and were randomized to receive 10µg buserelin acetate (GnRH) either at 32h (G32) or 48h (G48) after D8. Additionally, cows were inseminated with a single straw of higher fertility sires (H1) or with one (L1) or two (L2) straws of L sires on D10. Therefore, six groups [n] were created in a 2x3 factorial arrangement (G32-H1 [212], G32-L1 [194], G32-L2 [179], G48-H1 [185], G48-L1 [201], and G48-L2 [196]). Pregnancy diagnosis was performed on D40 after AI. Semen was from Alta Genetics (Uberaba, Brazil), and hormones were from Biogénesis Bagó (GnRH; Curitiba, Brazil) and Ourofino (Cravinhos, Brazil). Statistical analyses were performed by GLIMMIX procedure of SAS 9.4. Expression of estrus on D9.5 did not differ between G32 and G48 (16.3% [583] vs. 16.3% [583]; P=0.62). However, less cows from G32 compared to G48 were detected in estrus on D10 (77.2% [583] vs. 81.3% [571]; P=0.09). Independent of treatment, P/AI was greater in cows detected in estrus (57.9% [914] vs. 48.2% [240]; P=0.01). There was no interaction between time of GnRH treatment, number of straws and sire fertility on P/AI (P=0.84). In addition, P/AI was similar between G32 and G48 (56.4% [585] vs. 55.7% [582], respectively; P=0.81) and there was no effect of sire, or number of straws on P/AI (H1 = 57.4% [397], L1 = 57.3% [375], and L2 = 53.4% [395]; P>0.10). P/AI was greater in cows with higher (>2.75) than lower (≤2.75) BCS (58.4% [671] vs. 52.8% [496], respectively; P=0.05), and was lower when cows with BCS ≤2.75 were inseminated with L sires (L1 and L2), compared to H sires (50.3% [332] vs. 58.2% [165]; P=0.10). In conclusion, although the sires evaluated had been previously classified as with L or H field fertility, there was no difference between them regardless of treatment, except for cows with lower BCS, in which neither anticipating GnRH treatment, nor doubling the number of straws increased P/AI when L sires were used. It is speculated that GnRH treatment either before or at the time of FTAI may have restored P/AI of lower fertility sires, however, controlled studies (with vs. without GnRH) must be performed to confirm this assumption.Acknowledgements: FAPESP Grant # 2018/03798-7, Alta Genetics, Biogénesis Bagó, and Produzir Agropecuária.

Animal Reproduction

AI and IATF

Estradiol valerate is a great alternative to estradiol benzoate to promote the synchronization of ovulation and timed artificial insemination in suckled *Bos indicus* beef cows

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The objective was to evaluate the effect of administration of Estradiol Valerate at the beginning of the timed artificial insemination (TAI) protocol on ovarian follicular growth and pregnancy rate of suckled Bos indicus cows. Nelore cows (n=899), at 30-50 days postpartum and body condition score of 2.76±0.01 (scale of 1-5) were used. On a random day of the estrous cycle (D0), cows were allocated in 2X3 factorial design with a protocol of the Estradiol Valerate/TAI54h (EV group, 5mg;Valerol®, MSD, Brazil, n=400) or protocol of the Estradiol Benzoate/TAI48h (EB group, 2mg;Fertilcare Sincronizacao®, MSD , Brazil, n=499) and received intravaginal progesterone device (P4; FertilCare® 1200 with 1.2 g of P4, MSD, Brazil) new device (New, n=298) or previously used for 8 days (1X, n=309) or 16 days (2X, n=292)]. On D9, the P4 device was removed and all cows received 300IU of eCG (Folligon, MSD, Brazil). However, only cows of the EB group received 1mg of estradiol cypionate (Fertilcare Ovulacao®, MSD, Brazil) and 265µg of Cloprostenol (Ciosin®, MSD, Brazil). TAI was performed 48h after the removal P4 device in the EB group and 54h in the EV group. In a subset of cows (n=216), ultrasound exams were performed to evaluate the diameter of the largest follicle (D9 and TAI), follicular growth and early ovulation rate. Pregnancy diagnosis was 30d after TAI. Moreover, concurrent with removal of the P4 device, the tail-head of was marked with chalk in a subgroup of cows (n=452). Statistical analyses were performed by procedure LOGISTIC and GLIMMIX of SAS. There was no interaction between protocol groups and types of intravaginal P4 device on the follicular diameter at TAI (P=0.60), follicular growth (P=0.47), early ovulation rate (P=0.19), estrus occurrence (P=0.45) and P/AI (P=0.30). However, the follicular diameter on the D9 was greater in cows that received EB (EB - 10.6±0.3mm and EV - 9.4±0.4mm; P=0.003). In addition, no difference was observed between new or used of P4 device on the follicular diameter on D9 (P=0.59), follicular growth (P=0.93) and early ovulation rate (P=0.37). However, there was difference between previously used P4 device for 16 days and new P4 device on the follicular diameter at TAI (New - 12.0±0.4bmm, 1X - 12.6±0.4abmm e 2X - 13.2±0.4amm; P=0.06). Estrus occurrence was similar between estradiol groups (P=0.12) and uses of P4 device (P=0.91). The P/AI was similar between estradiol groups [EB - 50.3% (251/499) and EV - 51.8% (207/400)] and among different uses of P4 device [New - 48.7% (145/298), 1X - 48.8% (154/309) and 2X - 54.5% (159/292); P=0.47]. Moreover, cows that displayed estrus showed greater P/AI (P=0.03).In conclusion, despite lower follicular diameters at the P4 device removal, the use of the estradiol valerate at the beginning of the synchronization protocol and TAI 54h promotes similar occurrence of estrus and P/IA than the use of estradiol benzoate protocol and TAI 48h, regardless of the use of the P4 device.

Animal Reproduction

AI and IATF

Retrospective study of pregnancy loss in Nelore cows submitted to fixed-time AI protocols with or without GnRH at the time of AI

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This retrospective study evaluated pregnancy loss (PL) of Nelore cows submitted to fixed-time AI (FTAI) protocols with or without GnRH at the time of AI. We hypothesized that GnRH treatment at AI would not increase PL. Data from 2 breeding seasons (BS) were used, and in each one, experiments that included FTAI protocols with and without GnRH at AI were performed. In the first BS, cows were randomly assigned to 1 of 4 groups in a 2x2 factorial arrangement. On D0, cows either received 16.8 µg GnRH or 2 mg estradiol benzoate (EB), and at AI cows were treated with 8.4 µg GnRH or not treated, resulting in four groups. Simultaneously with the treatments on D0, cows received a 1 g intravaginal progesterone (P4) implant for 7 d. Also, on D7, cows received 0.5 mg cloprostenol (PGF) and 0.5 mg estradiol cypionate (EC). Cows treated with GnRH on D0 received 300 IU eCG and an additional PGF on D6, whereas cows treated with EB received the eCG treatment on D7. FTAI was performed on D9. PL in BS 1 was considered between AI and parturition. During BS 2, cows were allocated to 1 of 8 protocols in a 2x2x2 factorial arrangement. On D0, cows received a 1 g P4 implant and 2 mg EB and were treated or not with PGF. On D7, P4 implants were removed, all cows received PGF, 300 IU eCG, and 0.5 or 1.0 mg EC. On D9, at the time of AI, cows were treated or not with 8.4 µg GnRH, resulting in 8 groups. In the BS 2, PL was considered between 30 and 60 d after AI. Statistical analyses were performed using PROC GLIMMIX of SAS 9.3 ($P \leq 0.05$). Data regarding BCS and estrus expression at the end of the protocol were collected in both BS. The 2 BS were analyzed separately, and there were no interactions between GnRH at AI and FTAI protocol, parity, BCS and estrus expression. Moreover, there was no effect of FTAI protocol, and the variables also did not interact among them. There was no effect of parity on PL. Primiparous and multiparous had similar PL in BS 1 (9.1 [8/88] vs. 6.0% [17/263]) and BS 2 (5.8 [7/121] vs. 4.6% [22/474]). Cows with BCS ≥ 3 had similar PL than thinner cows in both BS 1 (5.5 [13/236] vs. 8.9% [12/135])] and BS 2 (5.3 [14/262] vs. 4.5% [15/333]). Expression of estrus also had no effect on PL. Cows that expressed estrus had similar PL than cows not displaying estrus in both BS 1 (6.0 [20/331] vs. 12.5% [5/40]) and BS 2 (5.1% [22/436] vs. 4.4 [7/159]). The main result proposed in the study was the effect of GnRH at AI on PL, and cows receiving GnRH had similar PL compared to those not treated in both BS 1 [6.2 (14/194) vs. 7.3% (13/177)] and BS 2 (4.6 [14/304] vs. 5.2% [15/291]). In summary, in this retrospective study, the use of GnRH treatment at the time of AI in Nelore cattle did not increase PL. More studies should be performed to precisely evaluate factors that affect PL in early pregnancy or between AI and parturition in Nelore beef cattle.Acknowledgements: Figueira Farm, FAPESP Grant # 2018/03789-7, CNPq, GlobalGen vet science.

Animal Reproduction

AI and IATF

Supplementation of injectable progesterone on the beginning of timed artificial insemination protocol in high-producing lactating Holstein dairy cows

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The objective was to evaluate the effect of supplementation of injectable progesterone on the beginning of timed artificial insemination protocol on pregnancy rate of high-producing lactating Holstein dairy cows. Holstein dairy cows (n=469; 255 primiparous and 244 multiparous) with milk production of 35.1±0.4 kg/day, at 147.8±5.1 days in milk, 2.9±0.1 inseminations and allocated in 3 farms were enrolled in this experiment. On a random day of the estrous cycle (D0), cows received a new intravaginal progesterone (P4) device (Sincrogest®, Ourofino, Brazil) or previously used P4 device for 8 days, 2mg of estradiol benzoate (Sincrodiol®, Ourofino, Brazil) and 100µg of buserelin (Sincroforte®, Ourofino, Brazil). At that time, cows were homogenously assigned to one of the treatments groups (Control; n=223 and P4i; n=246). Cows received no injectable progesterone (Control group) or 300mg of injectable progesterone (P4i group; Sincrogest injetavel®, Ourofino, Brazil). Seven days later (D7), cows received 500µg of Cloprostenol sodium (PGF; Sincrocio®, Ouro Fino, Brazil). On D9, the P4 device was removed and 500mg of PGF and 1mg of estradiol cypionate intramuscularly (SincroCP®, Ouro Fino, Brazil) were administered. The TAI was performed 48h after the removal of P4 device. Ultrasound examinations were performed on D0 to available presence of corpus luteum and 30 days after the TAI to evaluate pregnancy rate. Statistical analyses were performed by SAS. There were no interaction (P=0.05) between treatment and other variables (farm, CL on D0, progesterone device use, number of inseminations and parity). The pregnancy rate was lower in P4i group [Control 35.9% (80/223) and P4i 24.8% (61/246); P=0.01]. In conclusion, the supplementation of 300mg of injectable progesterone on beginning of timed artificial insemination protocol decreases the fertility of high-producing lactating Holstein dairy cows submitted to TAI.Acknowledgements: Ouro Fino Saude Animal, FAPEMIG, CAPES, CNPq

Animal Reproduction

AI and IATF

Does estrus synchronization based on estrogen-induced pseudopregnancy affect embryonic production and development in gilts?

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The development of estrus synchronization protocols with lower costs and simple application would benefit the swine production system. In pigs, estrogen stimulation from conceptuses plays a fundamental role for the establishment of pregnancy. The administration of exogenous estrogen during the pregnancy recognition (d 12) can be used to extend luteal phase, resulting in pseudopregnancy. The administration of PGF2α in a pseudo pregnant female will result in the expression of estrus within a given period. Therefore, we have investigated the effect of a single dose of the estradiol cypionate (ECP) to induce pseudopregnancy, followed by the administration of sodium cloprostenol to induce luteolysis, on the embryonic parameters of gilts. On d 12 of the estrous cycle (d 0 = last day of standing heat), 24 gilts (MS115), 220 ± 5 d old, 145 ± 3 kg of body weight and 3 estrous cycle were randomly assigned to two experimental groups: non-treated (NT, n = 9) and treated (CYP, n = 15) with a single dose of 10 mg of ECP i.m (SincroCP®, Ourofino Saúde Animal, SP, Brazil). Estrus detection started on d 17 and were conducted twice a day using a mature boar and back-pressure test. NT gilts were inseminated at onset of natural heat. On d 28 pseudopregnant gilts were treated twice (i.e., 800 and 1400 h) with 263 µg of sodium cloprostenol i.m (Sincrocio®**,** Ourofino Saúde Animal, SP, Brazil**).** Gilts were inseminated (AI) with refrigerated semen (3 × 10^9 sperm cells; >80% motility), from the same boar, at first sign of standing heat and every 24 h until the end of estrus. Five days after the last AI, gilts were euthanized and the reproductive tracts were obtained. Embryos were collected by uterine horn flushing as described by Marques et al, (2019) (Technical Report, Embrapa, 570:1-5, 2019) and classified according the IETS. Number of corpora lutea, total number of embryos and recovery rate of groups (NT and CYP) were compared by the t-test; all other variables were analyzed by the Wilcoxon test using SAS® software, significance was considered if P<0.05. Gilts from both groups had similar number of ovulations (NT, 12.5 ± 0.5; CYP, 14.0 ± 0.90; P = 0.17). No difference was observed on embryo recovery rate (NT, 77.5% ± 7.4; CYP, 87.7% ± 3.5; P = 0.18) and total number of embryos per gilt (NT, 9.75 ± 1.01; CYP, 12.53 ± 0.93; P = 0.06). For the analysis of embryo development, the percentage of non-fertilized oocytes (NT, 12.5% ± 12.39, CYP, 7.69% ± 7.69; P = 0.38); percentage of fragmented embryos (NT, 0.96% ± 2.71; CYP, 5.00% ± 1.57; P = 0.10) and the percentage of viable embryos (NT, 86.53% ± 11.07; CYP, 84.19% ± 7.78; P = 0.18) did not differed among groups. In conclusion, estrus synchronization of gilts treated with ECP to induce pseudopregnancy followed by the administration of sodium cloprostenol to induce luteolysis did not show differences on the ovulation rate, embryo production and their development when compared with the physiological estrus.

Animal Reproduction

ABSTRACTS: 34TH ANNUAL MEETING OF THE BRAZILIAN EMBRYO TECHNOLOGY SOCIETY (SBTE)

AI and IATF

Pregnancy losses in properties using timed artificial insemination in Mato Grosso do Sul

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The objective was to identify the factors that influence gestational losses in Zebu dams after timed artificial insemination program (TAI). The trial was conducted in three farms at MS state, two in the municipality of Miranda and one in Dois Irmãos do Buriti, with 14,525 Zebu cows and heifers, from three breeding seasons (years 2016, 2017 and 2018), in categories: precocious (14-16 months of age, n= 305), nulliparous (n= 2,245), non-lactating (n= 139), primiparous (n= 3,993), secondiparous (n= 1,147) and multiparous (n= 6,356), the last three being enrolled at TAI protocol after a minimum of 30 days postpartum. On the first day of the TAI protocol, the animals received 2 mg (IM) of estradiol benzoate (Gonadiol®; Zoetis, São Paulo, SP, Brazil), intravaginal progesterone device (CIDR ®, 1.9 g of progesterone; Zoetis) and body condition scored (BCS, 1-5). On D9, the device was removed, the sacro-caudal region was painted with a chalk and 0.6 mg (IM) of estradiol cypionate applied (ECP®, Zoetis), and also received 12.5 mg (IM) of PGF2α (Lutalyse®; Zoetis) and 300 IU (IM) of ECG (Novormon®; Zoetis). On day 11, 48 hours after implant removal, were inseminated, with semen from 14 Angus and 75 Nellore bulls of known fertility, and received at the time of insemination a note according to the removal of the chalk, for evaluation of estrus (ESCT) being: 1 - without removing the chalk; 2 - partial ink removal; 3 - total ink removal. After TAI, cows were mated with Nelore Bulls in a bull:cow ratio of the 1:40 for 90 days. The pregnancy diagnosis was performed 30 days after TAI with ultrasound and repeated at the end of the breeding season, with an average of 150 days of pregnancy, and animals pregnant in the first diagnosis, and empty at the second, or with lower gestational age relative to the date of the TAI, were classified in category pregnacy loss. The data were analyzed by PROC GLIMMIX from SAS (SAS / STAT® 9.2). There was a difference in the P/AI according to the category ($p < 0.001$), bull ($p < 0.001$), bull breed ($p < 0.001$), BCS ($p = 0.005$), month of calving ($p < 0.001$) and ESCT ($p < 0.001$). Regarding gestational losses, there was no difference ($p = 0.61$) according to the category (precocious= 9.55%; nulliparous= 7.21%; non-lactating= 9.46%; primiparous= 8.60%; secondiparous= 10.80%; multiparous= 7.20%) as well as for the bull breed (Nellore= 7.90%; Angus= 8.09%; $p = 0.6884$). There was a difference for the BCS ($p = 0.008$), the estrus expression score (ESCT 1= 8.88% [a], ESCT 2= 9.44% [a] and ESCT 3= 7.60%[b]; $p = 0.0479$), bull (range from 1.2% to 21.21%; $p = 0.009$) and for the calving month ($p < 0.001$). It is concluded that the body condition score at the beginning of the FTAI, heat expression, bull and the month of calving of the cows are the factors that had the greatest impact on pregnancy losses. This work is the basis for new studies to identify factors that impact gestational losses on beef cattle.

Animal Reproduction

AI and IATF

Association of controlled light program, cloprostenol and gonadotropins for synchronous estrus induction and fixed time artificial insemination in dairy goats during the non-breeding season: preliminary results

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Animal preparation prior to artificial insemination (AI) in goats usually involves a combination of intravaginal devices containing progesterone/progestagens plus PGF2-α analogues and gonadotropins, being eCG the most used. The use of intravaginal devices soaked in synthetic progesterone analogues requires milk discard for 60 days. Progesterone soaked intravaginal devices, as synthetic ones, are associated with more animal handling and discomfort. In addition, it leads to environmental impacts because it generates more residues. On the other hand, eCG has presented continuous restriction due to animal welfare issues associated with its production (Vilanova et al., Animals, 9:1053, 2019) and it has been substituted by hCG for this purpose. This study tested the efficiency of estrus induction by light program followed by estrus synchronization protocol with two doses of cloprostenol 7.5 days apart and gonadotropins to provide suitable condition able to support fixed time AI (FTAI) during the non-breeding season in dairy goats. Goats (n=18) were subjected to controlled light program using 16 h light and 8 h darkness during 60 days from June 30 (Day-0) to August 29. At Day-130 (6 am) and 137.5 (6 pm), goats received i.m. 37.5 µg d-cloprostenol (Prolise®, Syntex, Buenos Aires, Argentina) plus 250 IU hCG (Vetecor® 5000; Hertape Calier, São Paulo, Brazil) at second cloprostenol dose. Embrapa AI technique (Fonseca et al., Reproductive Biology, 17:268–273, 2017) with frozen-thawed semen was performed 63 to 64 h after second cloprostenol dose. Two goats whose mucus was considered inadequate were not inseminated. Immediately after AI, goats received alternatively 50 µg of gonadorelin (Gestran®, Tecnopec, São Paulo, Brazil) injected into the vagina with insulin syringe without needle (GnRH, n=8) or nothing (control, n=8). After 60 days of AI, pregnancy was checked by transrectal ultrasonography. Non-parametric data was evaluated by Fisher Exact Test with 5% minimal level significance. Pregnancy rate was similar (P>0.05) to GnRH treated (37.5% or 3/8) or not treated goats (75.0% or 6/8). One goat from GnRH group showed embryonic loss followed by hydrometra. Overall pregnancy rate was 56.2%. The results of this study showed the possibility of using a more natural form to induce asynchronous estrus during the non-breeding season (light program), associated with an estrus synchronization protocol using only cloprostenol and hCG as gonadotropins. It was possible to provide synchronous estrus conditions able to support FTAI in goats during the non-breeding season, resulting in good pregnancy rates. The use of GnRH analogues at AI time did not result in additional pregnancies. Financial Support: CNPQ (Project 314952/2018-7), Fapemig (Project CVZ-PPM 00201-17) and EMBRAPA (Project 20.19.01.004.00.03.001).

Animal Reproduction

AI and IATF

Administration of GnRH at AI increases pregnancy rate of suckled Nelore cows that received iP4 prior to ovulation synchronization and without estrus demonstration

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The aim of this study was to evaluate the effect of using GnRH at AI on pregnancy per AI of Nelore (*bos indicus*) cows that did not show estrus during TAI protocol. A total of 794 suckled Nelore cows, at 30-60 days postpartum (DPP) with average BCS 2.50±0.02 (1-5 point scale) from Couto Magalhaes farm (MT state, Brazil) were used. All the cows received 150mg of P4i (Sincrogest Injetável®, Ouro Fino, Brazil) 10 days before TAI protocol (D-10). On D0, cows received 2mg of estradiol benzoate (Sincrodiol®, Ouro Fino, Brazil) and a progesterone intravaginal device (Sincrogest®, Ouro Fino, Brazil). On D8, the progesterone device was removed and cows received 500µg of Cloprostenol (Sincrocio®, Ouro Fino, Brazil), 300IU of eCG (SincroeCG®, Ouro Fino, Brazil) and 1mg of estradiol cypionate (Sincrocp®, Ouro Fino, Brazil). Also, on D8 cows were painted with chalk on their tailheads, and removal of chalk on D10 was used as an indication of estrus. TAI was performed 48h after device removal, concomitant with estrus determination. At that time, cows that did not show (n=434) estrus were homogenously allocated to receive or not buserelin acetate (GnRH): Control [No treatment, n=214] and GnRH [Treatment with 10µg GnRH (Sincroforte®, Ourofino, Brazil), n=220]. Semen of bulls and inseminator were equally distributed between groups. Pregnancy diagnosis was performed by US 30 days after TAI. Statistical analysis was performed by GLIMMIX procedure of SAS®. Cows showing estrus had greater BCS than those without estrus demonstration (2.63±0.02 vs 2.26±0.02; P<0.001). The pregnancy rate was higher for GnRH group [Control: 52.8% (113/214) vs. GnRH: 60.9% (134/220); P=0.05]. In conclusion, the administration of GnRH at AI increases pregnancy rate of suckled Nelore cows that did not showed estrus during TAI and received an iP4 treatment 10 days prior to the beginning of the protocol. Thus, it can be used as a tool to optimize TAI outcomes.

Animal Reproduction

AI and IATF

Effect of time of permanence (7 vs. 8 days) of intravaginal progesterone devices on follicular dynamics and pregnancy rate of Nelore (*Bos indicus*) heifers

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The present study evaluated the permanence of the intravaginal progesterone device (7 or 8 days) on follicular dynamic and pregnancy rate of Nelore heifers submitted to TAI. The experiment was carried out at Nelore Paranã Farm (Iaciaria, GO, Brazil). A total of 780 Nelore heifers [26.8±0.3 months of age, 360.6±1.8 kg and 3.3±0.01 (1-5 scale) of BCS] were distributed into two experimental groups: 8DayP4 (8 days of device permanence; n= 404) and group 7DayP4 (7 days of device permanence; n= 376). Heifers from group 8DayP4 received intravaginal device with 0.6g P4 (P4D; Fertilcare 600®, MSD, Brazil) associated with 2mg estradiol benzoate (EB; Fertilcare Sincronização®, MSD, Brazil) and 0.25mg Sodic Cloprostenol (PGF; Ciosin®, MSD, Brazil) on D0. After 8 days (D8), P4D was removed and heifers received 0.25mg PGF (Ciosin®, MSD, Brazil), 0.5mg of estradiol cypioante (EC; Fertilcare Ovulação®, MSD, São Paulo) and 200IU of eCG (Folligon®, MSD, Brazil). At the same time, heifers were painted with chalk on their tailheads, and removal of chalk on D10 was used as an indication of estrus. Heifers from group 7DayP4 received the same P4D, BE and PGF doses, but on D1. After 7 days (D8), P4D was removed and followed by the same treatment as group 8DayP4. All heifers were inseminated on the same day (D10) and received 0.1mg Gonadorelin (Fertagyl®, MSD, Brazil). Pregnancy diagnosis was done by US 30 days after TAI. Moreover, a subgroup of heifers (8DayP4=78 and 7DayP4=85) were evaluated by US (Mindray® DP-2200Vet) in order to measure the diameter of the dominant follicle (DF) on D8 and D10. Data were analyzed by the GLIMMIX procedure of SAS®. The diameter of DF at P4D removal was larger in 8DayP4 heifers (8DayP4= 8.3±0.2mm vs. 7DayP4= 7.3±0.3mm; P=0.003). However, the DF at TAI was similar between the groups (P=0.20). In addition, early ovulation rate (between P4D and TAI) was higher in 8DayP4 than 7DayP4 group [16.7% (13/78) vs. 4.7% (4/85); P=0.02] and heifers that ovulated earlier had a tendency (P=0.07) for lower pregnancy rate [29.4% (5/17) *vs.* 45.8% (66/144)]. There was a tendency to increase estrus expression in 8DaysP4 group [8DayP4= 54.5% (220/404) vs. 7DayP4= 48.4% (182/376); P=0.07]. The pregnancy rate did not differ among groups [8DayP4= 44.3% (179/404) vs. 7DayP4= 47.3% (179/376); P=0.20]. Additionally, heifers were classified according to age [Old heifers=35.5±0.2 months (n=390) *vs.* Young heifers=19.1±0.2 months (n=390)] and it was found a tendency for the interaction age*group [8DayP4*Old heifers= 40.2%^{b} (82/204); 8DayP4*Young heifers= 48.5%^{a} (97/200); 7DayP4*Old heifers= 48.4%^{a} (90/186) and 7DayP4*Young heifers= 46.8%^{a} (89/190); P=0.07]. In summary, the protocol with P4D for 8 days showed a larger DF on D8 and a higher early ovulation rate. Heifers with early ovulation decreased P/AI. Despite no difference in pregnancy rate between the groups, older heifers with 8 days of device permanence may have lower pregnancy rate when compared to younger heifers

Animal Reproduction

AI and IATF

Fertility of Bos taurus cows submitted to a 7-day fixed-time AI protocol receiving 0.5 or 1.0 mg of estradiol cypionate as ovulation inducer and with or without GnRH at the time of AI

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The aim of this study was to compare 2 doses of estradiol cypionate (EC; 0.5 vs. 1.0 mg) as ovulation inducer and to evaluate the addition of a GnRH treatment at the time of AI on fertility of Aberdeen Angus cows. A total of 1541 (multiparous and primiparous) cows, with 74.0±57.4 d postpartum and BCS of 5.1±0.03 (1-9 scale) were used. The cows were randomly assigned into 1 of 4 groups in a 2x2 factorial design. On d0, all cows received 2.0 mg of estradiol benzoate (EB, EstroActive, Virbac, Mexico) and insertion of a 1.2 g intravaginal progesterone (P4) implant (DibActive 1200, Virbac). Seven days later (d7), all cows received 0,150 mg D-cloprostenol (PGF, InducelActive, Virbac) and 400 IU of eCG (GonActive eCG, Virbac). For the EC0.5NoG group, the cows received 0.5 mg EC on d7, concomitant with P4 implant withdrawal and did not receive GnRH at AI, whereas in EC0.5G group, the cows received 0.5 mg EC on d7 and 10 µg of buserelin acetate (GnRH, LiberActive, Virbac) on d9, at the time of AI. Cows from group EC1NoG received 1.0 mg EC on d7 and no GnRH at AI, and cows in group EC1G, received 1.0 mg EC on d7 and the GnRH treatment at the time of AI. All cows had their tail base painted on d7 to determine expression of estrus near the time of AI. Fixed-time AI was performed on d9 (48-52 h after the P4 implant removal). Pregnancy diagnosis was performed by transrectal ultrasound evaluation between 30 and 35 d after AI. Statistical analyses were performed with the PROC GLIMMIX of SAS 9.4 (P≤0.05). The overall estrus expression was 63.0% (971/1541) and was not influenced by EC dose (P=0.69), however, primiparous receiving 1.0 mg EC expressed more estrus than primiparous of groups EC0.5 (67.0 [120/179] vs. 58.8% [94/160]; P=0.01), in addition, 1.0 mg EC increased estrus of cows with BCS ≤ 4.0 compared to 0.5 mg EC (76.0% [168/221] vs. 62.3 [139/223]; P=0.001). Dose of EC did no impact expression of estrus in multiparous (P=0.59) or cows with BCS > 4.0 (P=0.27). There was no interaction between treatments on pregnancy per AI (P/AI) on d30 (51.4 [163/317] vs. 59.1 [250/423] vs. 53.7 [197/367] vs. 61.1% [265/434] for EC0.5NoG, EC0.5G, EC1NoG, and EC1G, respectively; P=0.62). Dose of EC did not affect P/AI (55.8 [413/740] vs. 57.7% [462/801] for EC0.5 and EC1, respectively; P=0.94), however, cows receiving GnRH at AI had ~8 percentage points greater P/AI (52.3 [360/684] vs. 60.1% [515/857]; P=0.02).Estrus affected P/AI (P=0.004) and GnRH at AI increased fertility of cows either without estrus (56.9 [201/353] vs. 47.8% [103/217]; P=0.03) or with estrus (62.3 [314/504] vs. 55.0% [257/467]; P=0.02). In conclusion, both doses (0.5 and 1.0 mg) of EC as ovulation inducers promoted similar fertility in Angus beef cows. In addition, the inclusion of a GnRH treatment at the time of AI increased fertility, regardless of EC dose or estrus expression.Acknowledgments: Virbac Mexico SA. de CV.

Animal Reproduction

AI and IATF

The influence of the number of antral follicles on in vitro production of Senepol embryos

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The antral follicle count (AFC) is the number of follicles identified by ultrasound in the ovaries. Studies have shown a relationship between AFC and embryo *in vitro* production (PIVE) for some bovine breeds. The aim of this study was to evaluate the relationship of AFC and viable oocytes, cleaved embryos, total embryo *in vitro* production, and conversion rate of viable oocytes in total produced embryos in Senepol females. The study was carried out in the region of the Triângulo Mineiro, Minas Gerais State, Brazil, with 146 females. The animals were classified according to the antral follicle count by ultrasound examination in low AFC (AFC ≤ 20), intermediate AFC (≥ 25 AFC ≤ 45), and high AFC (AFC ≥ 50). Statistical analysis was performed using the SAS Studio software. The relationships between AFC and the number of viable oocytes, cleaved embryos, and total embryo production were evaluated by regression. The relationships between AFC category and the other characteristics were evaluated using PROC GLM and Spearman correlation. We observed that the number of viable oocytes, cleaved embryos, and total embryos increased as the number of antral follicles increased (P< 0.0001). The analyses based on AFC category showed that females classified as high CFA produced higher quantities of viable oocytes (47.35±8.18), cleaved embryos (33.59±6.75), and total embryos (14.47±9.06). In contrast, females classified as low CFA showed the lowest results (9.33±3.51; 7.56±3.48; and 2.17±1.75, respectively; P<0.0001). However, no relationship was found (P=0.3400) between the categories of CFA and the conversion rate of viable oocytes in total produced embryos (high = 30.56%; intermediate = 19.72%; and low = 23.23%). Our results suggest that Senepol females with high CFA show higher production of viable oocytes, cleaved embryos, and total produced embryos *in vitro*, but with the same conversion rate as Senepol females with low and intermediate CFA.Keywords: cattle, precocity, conception rate, *in vitro* production of embryos.

Animal Reproduction

AI and IATF

Influence of chute exit speed score under the pregnancy rate in nelore dams submitted to timed artificial insemination

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The objective of this work was to evaluate the relationship between temperament (chute exit speed) and the pregnancy rate (P/AI) in different categories of Zebu breeds submitted to TAI. The study was conducted at Seriema Farm, located in the municipality of Miranda - MS, from October 2018 to February 2019, in an extensive breeding system, participant in the CEIP selection program where temperament is taken into account in the selection of Zebu breeds. 3,473 Zebu dams were used, separated by category: precocious heifers (14-16 months old, n= 304), heifers (24 months old, n= 818), primiparous (n= 641), secondiparous (n= 331) and multiparous (n= 1,379). The TAI protocol chosen was 11 days with three managements. On day 0, cows received of 2 mg (IM) of estradiol benzoate (Gonadiol®; Zoetis, São Paulo, SP, Brazil) and intravaginal progesterone device (CIDR®, 1.9 g of P4; Zoetis). On day 9, removal of the CIDR, concomitant with the application of 12.5 mg (IM) of PGF2α (Lutalyse®; Zoetis), 0.6 mg (IM) of estradiol cypionate (ECP®; Zoetis) and 300 IU (IM) of eCG (Novormon®, Zoetis), and TAI on day 11. After artificial insemination, the chute exit speed was observed by a trained person and directed only to this function, classifying as: 1= walking; 2= trotting; 3= running. Thirty days after artificial insemination, the diagnosis of pregnancy by ultrasound was performed. The data were analyzed by PROC GLIMMIX (SAS), and the characteristics included in the model were: technician (n= 8), bulls (n= 28), BCS (1-5), estrus (ESCT 1-3), category and chute exit speed, and when not significant, were excluded. The P/AI was influenced by BCS (p= 0.008), technician (p= 0.026), ESCT (p<0.001) and different according to the category (p<0.0016), with multiparous (56.63%), secondiparous (56.50%) and precocious (52.96%) not differing from each other and presenting better results than primiparous cows (46.8%) and heifers (49.75%). The bulls used (p= 0.184), had no effect on P/AI. Also, chute exit speed on the last day of the TAI protocol, there was no significant difference (p= 0.139). It is concluded that the temperament of the dams, evaluated through the chute exit speed, did not change the pregnancy rate of Zebu breeds selected in a CEIP breeding program.

Animal Reproduction

AI and IATF

Treatment with EB or injectable P4 in Nellore (Bos indicus) heifers on day 14 of doppler resynchronization protocol for TAI

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The objective of this study was to evaluate the efficiency of super-early resynchronization protocol in Nellore heifers using EB or injectable P4 (iP4) on day 14 (D14) after first TAI (D0; 2×2 factorial arrangement). Heifers (n=1116) were randomly distributed in 4 treatment groups on D14: 1) received only a intravaginal P4 device (P4D; Fertilcare®, MSD, Brazil; n=279); 2) received a P4D and 1mg of EB (Fertilcare sincronização®, MSD, Brazil; n= 277); 3) received 140mg of short-action iP4 (Progecio®, Agener, Brazil; n= 279); and 4) received a P4D, 1mg of EB and 140mg of short-action iP4 (n= 281). P4D was removed on the day of pregnancy diagnosis (D22) using Color Doppler ultrasonography (Mindray® M5Vet, China) (Ginther, 2007). Heifers with a CL vascularization greater than 25% were considered pregnant and were submitted to another US to confirm pregnancy and evaluate false positive rate. Heifers diagnosed as non-pregnant (with low or no CL vascularization) received 0,530 mg of cloprostenol (Ciosin®, MSD, Brazil), 200 IU of eCG (Folligon®, MSD, Brazil) and 0,5 mg of EC (Fertilcare Ovulação®, MSD, Brazil), and the dominant follicle (DF) was measured. Heifers were inseminated 48 hours later and were checked for pregnancy by US. Statistical analyses were performed by GLIMMIX procedure of SAS® (2×2 factorial arrangement). Since there was no interaction between EB and iP4 (P= 0.37), data was aggrouped and it is shown by main effects (EB × No-EB; P4 × No-P4). There was no effect of EB or iP4 on pregnancy rate [EB= 51% (283/558) *vs.* No-EB= 53% (296/558); P=0.43 and P4= 52% (290/560) *vs.* No-P4= 52% (289/556); P=0.95] and false positive rate of first TAI [EB= 10% (28/283) *vs.* No-EB= 14% (40/296); P=0.14 and P4= 13% (38/290) *vs.* No-P4= 10% (30/289); P=0.23]. It was verified that EB and iP4 decreased DF diameter at P4D removal on D22 [EB= 9.2mm *vs.* No-EB= 10.5mm; P<0.0001 and P4= 9.5mm *vs.* No-P4= 10.1mm; P=0.003]. However, only EB treatment decreased pregnancy rate of second TAI [EB= 39% (107/275) *vs.* No-EB= 47% (124/262); P=0.05 and P4= 43% (117/270) *vs.* No-P4= 43% (114/267); P=0.95] and tended to decrease cumulative (1^{st} + 2^{nd} TAI) pregnancy rate [EB= 68% (362/558) *vs.* No-EB= 73% (380/558); P=0.07 and P4= 71% (369/560) *vs.* No-P4= 71% (373/556); P=0.94]. It is concluded that iP4 and EB did not affect pregnancy rate of first TAI, and the use of iP4 did not increase the efficiency of Doppler resynchronization protocol. Furthermore, EB treatment decreased DF diameter and pregnancy rate of second TAI. Keywords: estradiol benzoate, resynchronization, Color Doppler ultrasonography.

Animal Reproduction

ABSTRACTS: 34TH ANNUAL MEETING OF THE BRAZILIAN EMBRYO TECHNOLOGY SOCIETY (SBTE)

AI and IATF

Fertility of Nelore heifers submitted to 7 or 9d fixed-time artificial insemination protocols with prostaglandin F2α administered at different times of the protocol

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The aim of this study was to compare reproductive outcomes of Nelore heifers (n=831) submitted to a 7 or 9 d estradiol (E2)/progesterone (P4)-based FTAI protocols, with PGF administered at different times, considering synchronization (n=533) and resynchronization (n=298) protocols. Prior to the onset of the breeding season (BS), heifers with corpus luteum (CL; n=89) on the first ultrasound evaluation were assigned to the treatments and those without CL received a protocol for induction of cyclicity (Day -35: 7 d-used intravaginal P4 implant [0.5 g]; Day -23: P4 withdrawal and 0.5 mg E2 cypionate [EC]). Twelve (for 9 d protocols) or 14 (for 7 d protocol) d later, heifers, regardless of CL presence, received a P4 device (0.5 g) and 1.5 mg E2 benzoate (EB). On the day of implant removal (7 or 9 d later), 0.5 mg PGF, 0.5 mg EC and 200 IU eCG were administered. For estrus evaluation, all heifers had the base of their tailhead painted with tail-chalk at the time of P4 removal and were checked 48 h later, at FTAI. Therefore, on Day -11 heifers were assigned to 1 of 4 treatments (n): The groups 9dP4-PGd9 (216) and 9dP4-PGd7 (198) received PGF on Day -2 or -4, respectively, whereas the groups 9dP4-PGd0&9 (215) or 7dP4-PGd0&7 (202) were treated with PGF on Day -11 and -2 or -9 and -2, respectively. Hormones were from Globalgen vet science and semen from STGenetics. Statistical analyses were performed by PROC GLIMMIX of SAS 9.4 ($P \leq 0.05$). The percentage of cyclic heifers at the onset of the BS was 16.7% (533) and from those without CL submitted to the protocol for cyclicity induction, 85.0% (440) had CL on Day -11. Heifers with body condition score (BCS) ≥ 3.0 had greater percentage of CL than those with BCS < 3.0 (84.5 [666] vs. 74.5% [161]). Group 9dP4-PGd0&9 had greater CL regression (94.4% [107]), between Day -11 and Day -2, than groups 9dP4-PGd7 and 9dP4-PGd9 but did not differ from 7dP4-PGd0&7. The diameter of the largest follicle (LF) at the time of P4 removal did not differ among groups, but the presence of CL at this time had a negative impact on follicle size (9.4±0.4 vs. 10.5±0.2 mm). The LF diameter at AI was smaller in group 7dP4-PGd0&7 (12.1±0.3 mm) compared to 9dP4-PGd7 and 9dP4-PGd0&9 (13.2±0.3 mm) but did not differ from 9dP4-PGd9 (12.9±0.4 mm). Expression of estrus (91% [831]) did not affect pregnancy per AI (P/AI; 52.5% [831]) and both variables were similar among groups. Ovulation after AI (96.1% [204]) and double ovulation (2.0% [200]) were similar among groups. Cyclic heifers at the onset of the BS had greater P/AI than heifers with induced cyclicity with or without CL on Day -11 (64.0 [89], 51.3 [374], and 47.0% [66], respectively). In conclusion, despite differences in ovarian dynamics, Nelore heifers submitted to either 7 or 9 d E2/P4-based FTAI protocols, with PGF administered in different times, had similar expression of estrus and P/AI. Acknowledgments: FAPESP Grant # 2018/03798-7, CNPq, CAPES, GlobalGen vet science, ST Genetics.

Animal Reproduction

AI and IATF

Influence of the analogue and dose of GnRH on the LH release and ovulatory response in Bos indicus heifers and cows with high circulating progesterone

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This study aimed to evaluate the influence of GnRH analogues (gonadorelin [GON] *vs*. buserelin [BUS]), as well as GnRH dose (single *vs*. double) on the LH release and ovulatory response in Nelore females with high circulating progesterone (P4) concentrations. Cycling heifers (n = 59) and non-lactating cows (n = 56) were previously enrolled in a presynchronization protocol (D-17: P4 implant and 2 mg estradiol benzoate; D-9: implant removal and 0.53 mg cloprostenol sodium [PGF]; D-7: 25 µg lecirelin [GnRH]). Females that ovulated to GnRH at D-7 were randomly assigned to receive one of the following treatments on D0: 100 µg GON (**G**); 200 µg GON (2G); 10 µg BUS (B); or 20 µg BUS (2B). At GnRH treatment, a P4 implant was inserted on heifers (0.5 g) and cows (1 g). Ultrasound examinations were performed on Days -9, -7, -5, 0, 2, 5, and 7 to check the ovulatory response to presynchronization, the diameter of the dominant follicle (DF) and ovulation to the GnRH treatments. Moreover, blood samples were taken on Day 0 at 0, 2, and 4 h after GnRH, to evaluate circulating P4 and LH concentrations. Five d after the treatments, the P4 implant was removed, females received two PGF treatments, 24 h apart, and 2 d later, 25 µglecirelin was given to reassign the females into the next replicate. Statistical analyses were performed by SAS 9.4 and results are presented as mean ± SEM ($P \leq 0.05$). In heifers, regardless of the dose, BUS induced greater LH release than GON. Additionally, the double dose increased the LH release only in heifers treated with BUS. Likewise, the ovulatory response on D0 was greater in heifers treated with BUS than GON (88.9 [24/27] *vs*.16.7%[5/30]), but there was no effect of dose or interaction. In cows, the GnRH-induced LH peak was greater for BUS than GON. Moreover, the double dose induced a higher LH peak than the single dose, regardless of the GnRH analogue. However, no interaction effect on LH peak was observed in cows. Otherwise, the ovulatory response on D0 was affected by an interaction effect, in which the double dose increased ovulation only in cows treated with BUS (single = 35.7% [5/14]; double = 90.9% [10/11]), whereas for GON treatments, there was no effect of dose (single = 35.7% [5/14]; double = 35.7% [5/14]). Moreover, cows presented a higher LH peak than heifers only with the 2G treatment, and the ovulatory response was greater in heifers than cows only with the B treatment. In conclusion, regardless of the category, BUS treatment produced greater LH release and ovulatory response, under high circulating P4. In addition, the double dose increased the LH release for both analogues in cows, but only for BUS treatment in heifers, and improved the ovulation only in cows treated with BUS. Finally, regardless of treatment, the amplitude of GnRH-induced LH peak positively affected the ovulatory response. However, apparently, heifers were more sensitive to this effect.Acknowledgments: FAPESP (Grant # 2018/03798-7), CAPES, and CNPq.

Animal Reproduction

ABSTRACTS: 34TH ANNUAL MEETING OF THE BRAZILIAN EMBRYO TECHNOLOGY SOCIETY (SBTE)

AI and IATF

Hormonal combinations aiming to optimize fertility outcomes of Nelore cows submitted to 7-d fixed-time AI protocols

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The aim was to study reproductive outcomes of Nelore (*Bos indicus*) cows submitted to a 7d estradiol (E2)/progesterone (P4)-based fixed-time AI (FTAI) protocol, using combinations of hormones or doses. Primiparous (n=962) and multiparous (n=1935) cows were submitted to synchronization (n=2012) and resynchronization (n=885) protocols, following a 2×2×2 factorial arrangement. On D0, cows were randomly assigned to experimental groups and received an intravaginal P4 implant (1g) and 2mg E2 benzoate (EB). On D7, every cow received 0.5mg cloprostenol (PGF) and 300IU eCG, concomitant with P4 withdrawal. FTAI was performed 48h later (D9). Experimental treatments were: administration (P1) or not (P0) of PGF on D0, 1 (EC1.0) or 0.5 (EC0.5) mg E2 cypionate (EC) on D7, and 8.4µg (G1) buserelin acetate (GnRH) or no GnRH (G0) on D9, resulting in 8 treatments (n): P0-EC0.5-G0 (364), P0-EC0.5-G1 (363), P1-EC0.5-G0 (363), P1-EC0.5-G1 (360), P0-EC-1.0-G0 (360), P0-EC1.0-G1 (363), P1-EC1.0-G0 (361), and P1-EC1.0-G1 (363). Hormones were from GlobalGen vet science and semen from STGenetics. Statistical analyses were done by PROC GLIMMIX of SAS 9.4 (P≤0.05). Presence of CL on D0 was greater in first AI than in resynch cows (39 [907] vs 19% [1914]). There was no interaction between the 3 factors of the study (PGF on D0, EC dose on D7, and GnRH at AI), and pregnancy per AI (P/AI) was greater at first AI compared to resynch (59 [2012] vs 55% [885]). Cows with BCS≥3 had greater expression of estrus (80 [1441] vs 62% [1371]) and P/AI (62 [1476] vs 53% [1421]) than thinner cows. There was no difference in expression of estrus or P/AI between cows that received or not PGF on D0. However, presence of CL on D0 resulted in higher expression of estrus (81 [680] vs 67% [2033]) and P/AI (66 [692] vs 55% [2106]). Cows that received 1.0mg EC expressed more estrus than those treated with 0.5mg (74 [1414] vs 68% [1398]) and had greater P/AI (60 [1447] vs. 55% [1450]). P/AI was greater when cows received GnRH at FTAI (60 [1449] vs 56% [1448]), particularly in cows that did not show estrus (53 [393] vs 38% [420]). Moreover, administration of GnRH on D9 increased P/AI in cows with BCS<3.0 (57 [723] vs 49% [698]), in primiparous (50 [465] vs 42% [497]) and in cows that received 0.5mg EC (59 [723] vs 51% [727]). Multiparous had larger follicles (mm) than primiparous cows on D7 (9.6±0.1 vs 8.6±0.1) and 9 (12.4±0.1 vs 11.0±0.1), and higher ovulation frequency after AI (93 [494] vs 82% [202]). In conclusion, although PGF treatment on D0 did not affect P/AI, 1.0mg of EC on D7 or GnRH at the time of AI improved P/AI, but the association between them did not enhance this effect. Moreover, GnRH improved P/AI especially in cows with lower expression of estrus, such as primiparous, thinner cows (BCS<3.0), and cows treated with 0.5mg of EC on D7.Acknowledgments: FAPESP Grant # 2018/03798-7, CNPq, CAPES, Figueira Farm, GlobalGen vet science, STGenetics.

Animal Reproduction

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AI and IATF

Efficiency of super-early resynchronization protocol without eCG or PGF use in Nellore (*Bos indicus*) cows

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Two experiments were carried out to evaluate the need of eCG and PGF use on D22 of super-early resynchronization protocol in Nelore cows, which in turn would make resynchronization protocol more profitable. In experiment 1, fourteen days after 1st TAI (D14), 1065 suckled Nelore cows (BCS= 2.75±0.4) received P4 device (P4D) (Fertilcare, MSD, Brazil). After 8 days the P4D was removed (D22), and cows were submitted to a pregnancy diagnosis (PD) by luteal vascularization using Color Doppler ultrasonography. Non-pregnant cows (low or absent luteal vascularization; n=546) were treated with 0,530 mg of sodium cloprostenol (Ciosin®, MSD, Brazil), 1mg of estradiol cypionate (EC; Fertilcare Ovulação®, MSD, Brazil) and then were allocated in a 2×2 factorial design to either receive or not 300UI of eCG (Folligon ®, MSD, Brazil) according to DF diameter (smaller or larger than 10.7mm). For estrus detection, cows were painted with chalk on their tailheads. TAI was performed 48h after P4D removal, concomitant with estrus evaluation. PD was performed 30days after TAI. In experiment 2, the use or not of PGF in non-pregnant cows that received a super-early resynchronization protocol treatment on P/AI was evaluated. Fourteen days after 1st TAI (D14), 325 Nelore heifers (Age: 24.5±0.1 months; BCS: 3.5±0.1), received a P4D (Fertilcare®, MSD, Brazil) and 1mg of EB (Fertilcare Sincronização®, MSD, Brazil). By P4D removal (D22), PD was performed by Color Doppler ultrasonography. Heifers diagnosed as non-pregnant (N:216) received 200 IU of eCG (Folligon®, MSD, Brazil), 0,5 mg of EC (Fertilcare Ovulação®, MSD, Brazil), and heifers were randomized to receive or not 0,530mg of PGF (Ciosin®, MSD, Brazil). Estrus detection evaluation and PD were done as previously described. Statistical analyses were performed by PROC GLIMMIX of SAS. In experiment 1, the estrus expression rate was similar between eCG treatment (P=0.45). However, cows with larger DF diameter had more estrus expression than smaller DF [DF≥10.7= 81.2% (310/382) vs. DF<10.7= 65.1% (95/146)]. The P/IA was similar for cows with DF larger than 10.7mm, but increased when eCG was given to cows with DF smaller than 10.7mm [eCG_FD≥10.7= 50.7% (99/195)^{A}; No_eCG_ FD≥10.7= 47.8% (90/188)^{A}; eCG_ FD<11= 39.7% (27/68)^{AB}; No_eCG_ FD<11= 22.9% (17/74)^{B}; P=0.05]. In experiment 2, there were no difference for estrus expression rate [PGF=83.8% (83/99) vs. No_PGF= 90.2% (92/102); P=0.40] and for P/IA (PGF=44.5% (45/101) vs No_PGF=45.1% (46/102); P=0.93). It can be concluded that cows with large DF express more estrus than cows with small DF. Regardless eCG treatment, the P/IA is similar for cows with larger DF. However, the P/IA increased when eCG was given to cows with small DF. Thus, it is not necessary to apply eCG in Nelore cows with DF larger than 10.7mm. In addition, there is no difference for estrus expression rate and P/IA in heifers treated with or without PGF. Thus, it is not necessary to apply PGF in Nelore heifers.