

## Animal Reproduction

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## Gradual acclimation to repeated handling prior hormonal protocol as an alternative to reduce stress in beef cattle

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The aim of this study was to evaluate the effect of gradual acclimation to repeated handling of cow and calf before TAI hormonal protocol on stress and reproductive indices. Nelore cows (n = 116) with an average of 59.81 ± 17.49 days postpartum were divided into two groups. During the thirty days period prior to initiating the TAI protocol, females in the Treatment group (TG, n = 58) were subjected to gradual acclimation to repeated handling 30 days prior TAI protocol, which consisted of weekly visits to the corral and passage through the cattle chute avoiding physical aggression, shouting or manipulation. At squeezing chute individual containment of each animal was performed by closing the entrance and exit gates. The cows and their calves were then placed in a support paddock where they received positive reinforcement (feed with chopped grass and fresh water) before returning to pasture. The control group (CG, n = 58) only went to the corral for selection and then to start the TAI protocol. Cow weight and body condition score at the beginning of the TAI protocol were not influenced by gradual acclimation (466.48  $\pm$  63.19 vs 467.70  $\pm$ 61.79 kg, respectively for CG and TG; P = 0.916). Time-fixed insemination was considered as Day 0 (D0). In half of the animals from each group, preovulatory follicle volume (at D0) and corpus luteum (CL) volume (at D7 and D16) were measured by the same technician using B-mode transrectal ultrasonography. At D0, D7, D16, and during animal selection, blood was collected via coccygeal vein puncture for cortisol measurement by solid-phase radioimmunoassay technique using commercial kits. Data was subjected to Kolmogorov-Smirnov test for normality assessment. Normal data was compared by ANOVA or t-test. Non-normal data was evaluated by Mann-Whitney test. The impact of gradual acclimation on pre-ovulatory follicular (0.83 ±  $0.32 \text{ vs } 1.06 \pm 0.52 \text{ cm}^3$ , P = 0.107; respectively for CG and TG) and on CL volume on D7 ( $3.49 \pm 1.55 \text{ vs } 3.78 \pm 1.06 \pm 1.06 \pm 1.06 \pm 1.06 \pm 1.06$  $1.73 \text{ cm}^3$ , P = 0.570; CG and TG, respectively) and D16 ( $3.42 \pm 1.19 \text{ vs } 3.31 \pm 1.48 \text{ cm}^3$ , D16, P = 0.779; CG and TG, respectively) was not significant. Cortisol concentrations at the time of animal selection did not differ (35.47 ± 14.48 vs 37.54 ± 16.72 ng/ml, respectively for CG and TG; P = 0.595), however, TG showed lower cortisol concentration at D0 (15.92  $\pm$  9.50 vs 30.89  $\pm$  15.45 ng/ml, respectively for TG and CG; P  $\leq$  0.001). At D7 and D16, cortisol levels were similar between groups (at D7, 13.82  $\pm$  8.98 vs 13.98  $\pm$  7.88 ng/ml, P = 0.936; and at D16,  $11.84 \pm 8.21$  vs  $14.30 \pm 12.66$  ng/ml, P = 0.704, respectively for CG and TG). The gradual acclimation to repeated handling reduced animal stress at the beginning of the TAI protocol, however, no effect was observed on the volume of the follicle and corpus luteum.