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Peripheral insulin resistance in Holstein (*Bos taurus*) repeat breeder cows

B.M. Bayeux[1], R.D. Mingoti[1], A.E. Vercesi Filho[3], G.D. Melo[1], B.L.C. Catussi[1], L.G. Silva[1], Y.F. Watanabe[2], P.S. Baruselli[1]

[1]USP - Universidade de São Paulo, São Paulo, SP, Brasil; [2]VITROGEN, Cravinhos, SP, Brasil; [3]IZ - Insitituto de Zootecnia, Nova Odessa, SP, Brasil.

The aim of this study was to evaluate the establishment of peripheral insulin resistance (PIR) in Holstein (*Bos taurus*) repeat breeder (RB) cows and the effect on the *in vitro* embryo production (IVEP). Twenty-eight cows were distributed in three experimental groups: Initial Lactation [IL; DIM <50 days; n=9; 24.9±1.7kg/d of milk; BCS=2.9±0.07]; End Lactation and Repeat Breeders (EL-RB) [DIM>300 days; n=10; 17.2±0.9kg/d of milk; BCS=3.4±0.12] and End Lactation and Pregnant [EL-P; DIM>300 days; n=9; 19.6±1.3kg/d of milk; BCS=3.4±0.06]. For the glucose tolerance test (GTT), blood samples were collected after 4 hours of fasting at -20, -10 and 0 min, and an infusion of 0.3 g / kg of sterile glucose solution [50% of glucose (iv)] was performed at the 0 minutes moment during 5 min. At 5, 10, 20, 30, 40, 60, 80, 100 and 120 subsequent minutes, blood samples were collected to calculate the metabolism rate (k), and half-life (T½) of the plasmatic glucose. The insulin response (INS) after GTT was evaluated by the circulating insulin concentration and by the insulin increase ([INS] peak - [INS] basal; Δmax). An ovarium pick-up (OPU) were performed in a random day of estrus cycle to determinate oocytes quality and a US (Mindray® DP2200 Vet) evaluation was performed for antral follicle count (CFA). Statistical analyzes were performed by orthogonal contrasts [Contrast 1 (C1) = Lactation Phase (IL *vs* FL-RS + FL-P) and Contrast 2 (C2) = Reproductive Status (FL-RB *vs* FL-P)] using logistics regression by GLIMMIX PROC of SAS. Statistical differences were found considering lactation phase, for the variables area under the curve (AUC) between 5 and 120 minutes after infusion [IL=4367.8±927.1 *vs* RB=9524.5±1746.3 + EL-P=8047.6±1113.8; P=0.02]. Antral follicle count (CFA) was higher in cows at the beginning of lactation when compared to cows at the end of lactation [IL=21.3±6.0 *vs* EL-RS=13.6±4.3 + EL-P=10.8±2.2; P=0.03 (C1)] but the number of recovered oocytes, viable oocytes, oocyte recovery rate, and cleavage rate did not differ between groups (P> 0.05). A difference was found for blastocyst rate [IL = 10.0% *vs* EL-RB = 18.9% + EL-P = 22.8%; P = 0.01] and for embryo development rate [IL = 14.0% *vs* EL-RB = 41% + EL-P = 38.1%; P = 0.03] considering the moment of lactation. In conclusion, repeat breeder cows and pregnant cows at the end of lactation have higher insulin in response to TTG, evidencing the establishment of the PIR. However, poor oocyte quality was not demonstrated when compared to cows at the beginning of lactation.

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