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Profile of type I and II interferon receptor transcripts in peripheral blood mono and polymorphonuclear cells during early gestation in Nelore heifers

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We aimed with this study to analyze the abundance levels of type I and II interferon-tau (IFNT) receptors (IFNAR I and II) in peripheral blood mono (PBMC) and polymorphonuclear (PMN) cells in pregnant Nelore heifers. Twenty-nine heifers (18-20 months) had their estrous cycle synchronized and were subjected to fixed-time artificial insemination (FTAI) on D0. Pregnancy diagnosis was performed by transrectal ultrasonography on D25 and D28 through the detection of the embryonic vesicle and heartbeat. On days 0, 10, 14, 16, 18 and 20, 25 mL of blood were collected in heparinized tubes by puncture of the jugular vein for the isolation of PBMCs and PMNs cells. The isolation was performed by Ficoll® Paque Plus gradient (GE Healthcare, Chicago, USA), in an adapted method (Jientaweeboon S et al. 2011. *ReprodBiol and Endoc.*, 9:79-89). Samples from 8 pregnant and 9 non-pregnant heifers were submitted to RNA extraction using the Direct-Zol RNA Miniprep Kit (Zymo Research, Irvine, USA) according to the manufacturer's instructions. The expression of the target genes (*IFNAR I* and *II*) was normalized in relation to the reference genes (*GAPDH* and *PPIA* for PBMCs; and *GAPDH* and *ACTB* for PMNs). For statistical analysis, the transcript abundance levels were evaluated by analysis of variance (ANOVA) with repeated measures of time, considering the random effect of heifer and the fixed effects of group (pregnant or non-pregnant), day and interaction of group by day using the PROC MIXED SAS software (SAS Institute). For PMNs, no significant ($P>0.1$) differences were detected in the *IFNAR I* expression, while for *IFNAR II*, only a time effect ($P= 0.01$) was observed, indicating an increase on transcript abundance from D0 to D16, with a progressive decrease on D20 in pregnant heifers. For PBMCs, only a time effect ($P= 0.02$) was observed for *IFNAR I* expression, characterized by an increase on the transcript abundance between D10 and D16, followed by progressive reduction on D18 and D20. Although an interaction of group by time was not significant ($P=0.11$), a subsequent analysis indicated that *IFNAR I* abundance on PBMC in the pregnant heifers progressively increased from D0 to D16 and followed a progressive decrease from D16 to D20; whereas, no difference ($P>0.05$) was detected along the evaluated days in the non-pregnant heifers. Also, the *IFNAR I* abundance on D20 was greater ($P=0.04$) in the pregnant than non-pregnant heifers. No significant ($P>0.1$) effects were detected in the *IFNAR II* expression. In conclusion, for PMN, only *IFNAR II* transcript abundance varied during early pregnancy but its expression is independent of the pregnancy status; whereas, for PBMC the pregnancy status may affect the temporal expression of *IFNAR I* at D20, which could be involved in the IFNT signaling mechanisms to guarantee the success of maternal recognition of gestation. Acknowledgments: FAPESP (2015/10606-9; 2017/13472-9; 2018/25393-9).