



A140 Embryology, developmental biology and physiology of reproduction

### **miRNAs identified in corpus luteum of IVF recipient cows are absent in SCNT recipient animals on day 19 of pregnancy**

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Corpus luteum (CL) is responsible by P4 production and it is necessary for establishing and maintenance of pregnancy in cattle. Normal luteal function is regulated by miRNAs, that are small non-coding RNA molecules involved in post-transcriptional regulation of target genes. However, the role of miRNAs and its potentially regulated pathways are poorly known in the CL of cows carrying conceptus derived from different biotechnologies (IVF or SCNT). Therefore, our hypothesis is that miRNAs are exclusively expressed in the CL of cows carrying IVF or SCNT conceptus on day 19 pregnancy. For this, COCs recovered from ovaries collected at local abattoir were used to produce SCNT blastocyst and, IVF embryos were made from oocytes collected by OPU. Nellore cows had the estrus synchronized and received one embryo (IVF or SCNT), on day 7 after expected estrus. CL were collected on day 19 of pregnancy, in animals carrying IVF (n=3) or SCNT (n=3) conceptus. The CL function was evaluated by P4 concentration in blood serum collected from the jugular vein on days 9, 14 and 19 of pregnancy. Mature miRNAs were reverse transcribed using MiScript HiSpec Buffer. The relative levels of 384 miRNAs were evaluated. The data were normalized by the geometric mean of miR-99b, RNU43 snoRNA and Hm/Ms/Rt U1 snRNA. miRNAs were considered exclusively expressed when the expression was detected in all CL samples of one group and not detected (not expressed) in all samples of the other group. On days 9, 14 and 19, serum P4 concentrations were similar between IVF and SCNT groups (Bridi, et al., *Animal Reproduction*, 15:480, 2018). A total of 288 mature miRNAs were identified in CL samples from both groups, with one exclusive miRNA in the SCNT-CL and three exclusive miRNAs in the IVF-CL. The identification of bovine genes modulated by each miRNA was performed using TARGETSCAN software. After gene identification, the code Ensembl Transcript ID was used to determine enriched pathways regulated by these miRNAs using DAVID Bioinformatics Resources 6.8, NIAID/NIH. The miRNA bta-miR-129-3p, uniquely detected in the SCNT CL group, modulated signaling pathways that include MAPK (14 genes), oxytocin (9 genes), GnRH (7 genes) and estrogen (6 genes). Moreover, bta-miR-141, bta-miR-302a and bta-miR-875, which were uniquely detected in the IVF CL, regulate PI3K-Akt (40 genes), MAPK (33 genes), Hippo (25 genes) and oxytocin (18 genes) pathways. The results show that, on day 19, the expression of different miRNAs in the CL of recipient cows can be modulate by conceptus of different origins (IVF or SCNT). Furthermore, the exclusive miRNAs of both groups regulate oxytocin signaling pathway in the CL, that have an important biological role in the maintenance of luteal function and, consequently in the establishing of pregnancy. Funding: FAPESP grants 2014/22887-0; 2016/50433-9; 2017/19681-9, 2017/50438-3 and 2018-13155-6. Acknowledgments: WTA.