Treatment with c-type natriuretic peptide precursor (NPPC) during in vitro prematuration of bovine cumulus oocyte complex: effects on transcript profile and meiotic arrest

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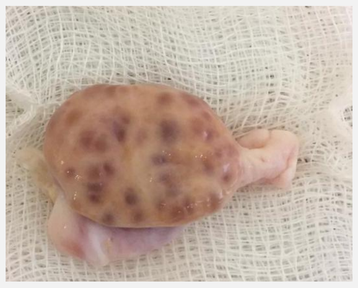
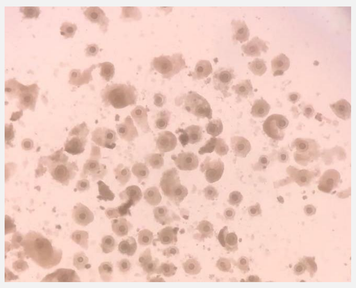
INTRODUCTION

Oocytes that undergo in vitro maturation (IVM) come from follicles with 3 to 8 mm in diameter, these characterize a heterogenous oocyte population, which are at different stages of development. Furthermore, when oocytes are removed from follicular environment, they spontaneously resume meiosis (from germinal vesicle - GV - to metaphase II - MII - stage). C-type natriuretic peptide precursor (NPPC) is mainly synthesized by granulosa cells and acts to maintain oocyte meiotic arrest. In this way, the use of NPPC during a pre-in vitro maturation (preIVM) culture may be helpful in promoting meiosis blockade, allowing additional time for oocytes to acquire competence. So, the aim of this study was to evaluate the transcript profile in cumulus cells derived from bovine COCs submitted to preIVM with NPPC and investigate its effects on meiosis blockade.

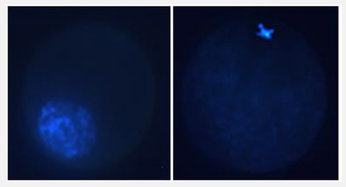
MATERIAL AND METHODS



Abattoir-derived ovaries
Follicles 3-8 mm



preIVM for 8h
Groups: NPPC, controls preIVM (C8) and immature (C0)



Data were analyzed by ANOVA followed by Tukey's test (P<0.05).

RESULTS AND DISCUSSION

There was no significant difference (P>0.05) in GV rates (mean ± SEM) between NPPC (52.8% ± 15.5) and C8 groups (26.1% ±7.2), but both differ (P<0.05) from C0 (96,4% ± 2.4). A total of 7 genes were upregulated (↑; P<0.05) and 7 genes were downregulated (↓; P<0.05) in NPPC group compared to C0, as follows: genes related to embryonic quality and development (↓*CLIC3*, ↑*IGF1R*, ↑*KRT8*, ↑*LUM*, ↓*PRDX3*,↓*S100A14*), oxidative stress (↓*CAT*, ↓*GPX1*, ↓*HSPA1A*, ↑*GFPT2*, ↑*GLRX2*, ↓*DDIT3*) and apoptosis (↑*TNFRSF21*, ↑*TP53*, ↑*BCL2*) (P<0.05). When N8 group was compared to C8 group, genes like *KRT8*, *CAT* and *TP53* were upregulated (P<0.05), in contrast 7 genes related to embryonic quality and development (*LUM and S100A14*) and oxidative stress (*GFPT2, GLRX2, ATF4, DDIT3* and *XBP1*) were downregulated (P<0.05). Analysis of gene ontology (GO) was performed with the R package clusterProfiler and 46 enriched GO biological processes and 31KEGG pathways were identified.

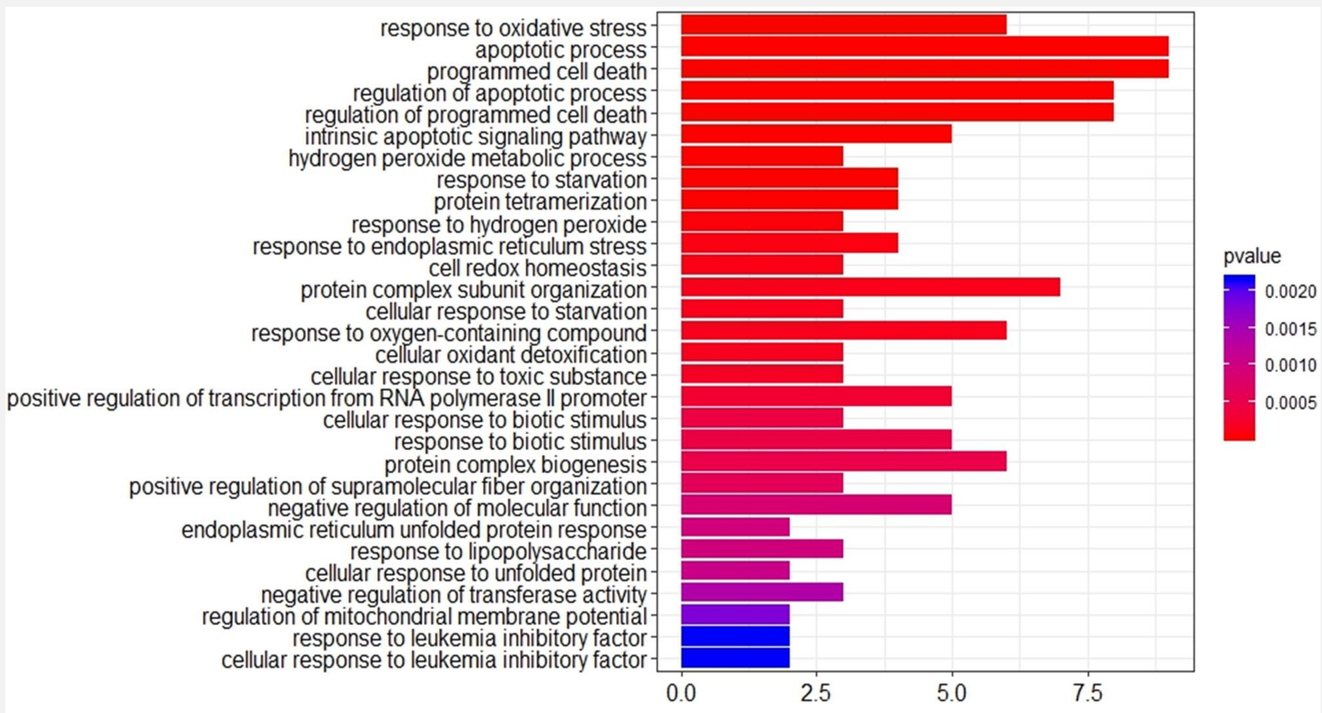


Figure 1. Gene ontology analysis output – biological processes

CONCLUSIONS

Although the meiosis progression in the NPPC group did not differ from the control group, gene expression results demonstrate that the preIVM culture may be beneficial, suggesting a higher antioxidant response and lower activation of endoplasmic reticulum stress, apoptosis, and cellular stress response pathways.

Acknowledgements

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