Antiapoptotic and antioxidant effects of melatonin on cat vitrified oocytes

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Cryopreservation of oocytes is crucial to bank precious genetic resources of domestic and wild animals. However, cryopreservation is known to cause several damages, including oxidative stress and apoptosis, which hinder oocyte developmental competence. The addition of antioxidant or antiapoptotic compounds could improve the outcomes of preserved oocytes, so the aim of this study was to test the effect of melatonin on reactive oxygen species (ROS) and apoptotic markers (DNA fragmentation, caspase activity) in immature cat vitrified oocytes at warming and after in vitro maturation.
Cumulus oocytes complexes (COCs) collected from spaying-derived ovaries were vitrified-warmed with Cryotop (with/without 10^{-9} M melatonin). Maturation lasted 24h at 38.5°C, 5% CO_2, in M199 supplemented as in [1], with or without melatonin. Fluorescent probes were used for ROS and caspases (CellROX Green and CellEvent Caspase-3/7, Thermofisher) and DNA integrity (Cell Meter TUNEL, AAT Bioquest). Data were analyzed by Kruskal-Wallis (caspases and ROS fluorescence intensity quantified by ImageJ) or Fisher's (TUNEL) tests; significance $p<0.05$.
Melatonin-vitrified COCs had four times less ROS after IVM compared to control vitrified COCs at warming ($p=0.04$). Melatonin did not influence DNA fragmentation, but it halved caspases after IVM in comparison to untreated COCs ($p=0.04$).
In summary, melatonin exerted antioxidant and antiapoptotic effects on cat vitrified COCs and could be used to enhance the quality and the developmental potential of cryobanked gametes, crucial means of fertility and biodiversity preservation.
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[1] Colombo et al., 2020, Reprod Domest Anim, 55 Suppl 2:74-80