

Abstracts - 37th Annual Meeting of the Association of Embryo Technology in Europe (AETE) Embryology, developmental biology, and physiology of reproduction First evidence of nanoplastic uptake by the maturing oocyte

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Plastic pollution has become a growing environmental problem as the result of its widespread use and improper treatment of plastic waste. Over time, plastics that leaked into nature will degrade into smaller particles including micro- and nano plastics (MNPs). MNPs can enter the human body through ingestion, inhalation, and dermal contact (Parta et al., Sci Total Environ, 702:134455, 2020). Previous studies have shown that MNPs can induce an immune response and can cause a neurologic, oxidative, and toxicity response in somatic cells, including those of the reproductive tract in both mammals and aquatic organisms (Xie et al., Ecotoxicol Environ Saf, 190:110133, 2020; Liu et al., J Hazard Mater, 424:127629, 2022; Nie et al., Nanotoxicology, 15;885, 2021). However, the impact of MNPs on oocyte and embryonic development remains largely unknown. Furthermore, it is unknown whether oocytes take up MNPs from the environment.

In this study, the uptake of MNPs by developing bovine cumulus-oocyte-complexes (COCs) and their effects on development have been studied. Given the large similarities in reproduction and early development during early embryonic development between human and cow, the bovine model is an excellent model to study human oocyte and early embryo development (Sirard, In: Animal Models and Human Reproduction, 127-144, 2017; Eds: Constantinescu & Schatten; Wiley & Sons Inc.; ISBN:9781118881606). COCs, collected from slaughterhouse ovaries, were exposed to our standard maturation condition without, control condition (NaHCO3-buffered M199 supplemented, with 100 IU/ml Penicillin-streptomycin, 0.05 IU/mL FSH, 0.1 µM cysteamine, and 10 ng/mL EGF) or with green-fluorescent polystyrene (PS) nanoplastics (Polysciences, Inc., Hirschberg an der Bergstrasse, Germany) of 50nm or 200nm (10 µg/mL) during the 23 h *in vitro* maturation (39 °C, 5% CO2 in air). After maturation, COCs were stained with Hoechst (DNA) and phalloidin (actin), to score the nuclear stage of matured oocytes (oneway ANOVA), and uptake of MNPs, respectively.

Confocal microscopy showed that MNPs of 200nm were only taken up by some cumulus cells, while MNPs of 50nm were taken up by cumulus cells and oocytes. In total, 351 oocytes in 3 replicates were analysed, there was no difference between the nuclear maturation status after exposure to MNPs of 200nm (59.9%) and 50nm (46.5%) during maturation, in comparison to control oocytes (60.8%). In conclusion, bovine oocytes are able to take up 50nm PS particles while cumulus cells are able to take up both 50 and 200 nm PS particles. However, the current experiment did not show an effect of the uptake of plastics on the nuclear stage of the oocyte after maturation. Future studies need to unravel whether the uptake of MNPs may impact oocyte competence.

Keywords: micro- and nanoplastics, oocyte, bovine embryo model