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Lipids characteristics in bovine preimplantation embryos originated from in vitro fertilization or parthenogenetic activation

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Lipids are essential elements of the cells since they build biological membranes and are involved in many processes, including energy metabolism. They are stored in the cytoplasm in lipid droplets (LD). Lipid content alters during the preimplantation development of embryos, mainly due to the changes in energy metabolism requirements. Except for classic in vitro fertilization (IVF), embryos have the ability to activate their development without the involvement of male gametes (parthenogenesis, PA). The question arises of whether it affects embryonic lipid content.

The aim of this study was lipid characteristics in embryos: 1. at crucial stages of development, 2. originating from IVF and PA systems.

Bovine oocyte-cumulus complexes were matured in vitro and either in vitro fertilization (bIVF group) or parthenogenetic activation (5uM ionomycin/2mM 6DAMP; bPA group) was performed. Embryos were cultured in SOF medium and collected in the following developmental stages: zygote, 2-cell, 4-cell, 8-16-cell, morula, early and expanded blastocyst (approx. 20 per group). They were stained with BODIPY 493/503 and DAPI for LD and chromatin visualization, respectively, then captured and z-stacked with Zeiss LSM 880 confocal microscope. The following parameters were analyzed in ImageJ Fiji software: total lipid content, LD number, LD size, and % area of LD. Statistics included appropriate tests in the R statistical package.

Our results show that in bIVF group, total lipid content reaches the highest level at the zygote stage, and it drops to the lowest values at the 8-16-stage ($P<0.01$), following a significant increase at the expanded blastocyst stage ($P<0.05$). A similar decrease at the 8-16-cell stage is observed for the LD number (113+/-41 vs 57+/-21 μm^2), LD size (5.15+/-2.59 vs 2.16+/-0.77), and % area occupied by LD (3.3+/-1.14 vs 1.06+/-0.83) ($P<0.01$). When bIVF and bPA at 2-cell, 4-cell, and expanded blastocyst stages are compared, a significantly lower value of total lipid content ($P<0.05$) is observed at the 2-cell stage in bIVF embryos. Yet, there areno differences between expanded blastocysts.

LD parameters decrease at the 8-16-cell stage is observed at the same time-point as the embryo genome activation in cattle. It suggests that early embryos may strongly utilize lipids as a source of energy without the possibility of replenishment due to the lack of transcription. Moreover, others suggest a lower contribution of lipids in the late preimplantation embryo development since embryos switch their energy metabolism into glucose. However, it is inconsistent with our data due to the observed rise of lipid parameters at the blastocyst stage. Our results also indicate that lipid content is altered depending on the embryo origin (IVF or PA) only in the early stages of development (2-cell embryos). It suggests that during further development, embryos are able to compensate for the lipid deficiencies, however further studies in every step of development are necessary.

Keywords: embryo, cattle, lipids

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