## A32
## BLASTOCYST PRODUCTION BY INTERSPECIFIC ICSI IN FELIDS

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The ICSI is an assisted reproductive technique that can be used in wildlife to preserve the biodiversity. The aim of this work was to determine the fertilizing capacity of cheetah spermatozoa (*Acinonyx jubatus*) using the ICSI technique with domestic cat oocytes (DC, *Felis catus*). The oocytes were obtained from cats subjected to ovariectomy and matured for 22h. Mature oocytes were injected with a cheetah or DC spermatozoon (frozen/thawed) obtained from electroeyaculation and epididimis, respectively. Another group was injected without any spermatozoon (sham control). Injected oocytes were cultured immediately or were chemically activated with ionomycin (Io) before culture. The results were analyzed by Fisher test, $p<0,05$. Cleavage rates were: 66,3% (65/98 Cheetah); 73,6% (67/91 Cheetah-Io); 43,5% (37/85 DC); 69.7% (76/109 DC-Io); 19,2% (10/52 Sham) and 65,3% (47/72 Sham-Io). Interspecific embryos reached blastocyst stage equally as DC embryos, 32.6% *vs.* 20%, respectively. On the other hand, ionomycin assistance did not improve blastocyst rates in any of the groups, 21% *vs.* 17.4% for Cheetah-Io and DC-Io. The Sham control showed blastocyst formation only when Io was used (Sham-Io group, 6,9%). In summary, the ICSI technique without Io assistance was an efficient technique for embryo production with cheetah spermatozoa, which is an endangered species. This technique has great potential for poor quality sperm samples of endangered wild felids.

# REPRODUCTION

## A33
## CHANGES IN IGF-1, IGF-2 AND IGF-1R EXPRESSION IN CANINE PLACENTAE OF DIFFERENT PREGNANCY AGE

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The IGF system is involved in human placental development. The aim of the work was to evaluate IGF-1, IGF-2 and IGF1R expression in canine early, mature and term placentae (EP, MP, TP, respectively). Placental samples from 7 dogs were obtained by Hysterectomy and processed by indirect immunohistochemistry for the detection of IGF1, IGF2 and IGF1R.
In EP fetal trophoblast and in labyrinthine cytotrophoblast both IGF were evidenced (IGF2>IGF1). Their expression decreased as pregnancy drawed on. IGF1R was evidenced in MP. IGF1R expression in syncytiotrophoblast was greater than that of both IGF. Labyrinthine and endometrial maternal endothelia were positive to IGFs and IGF1R, with stronger labelling in MP than in EP. Although gland endometrial cells were positive for all the studied markers, IGF2 detection was markedly polarised (apical pole in EP, apical and basal poles in MP), coinciding with IGF1R expression.
IGF are expressed in the main cellular populations of the canine endotheliochorial placenta. They may be involved in cell signalling and mediate endometrial invasion by the trophoblast. IGF-2 expression is stronger than IGF-1´s. IGFs expression is stronger in EP, while IGF1R increases later on. Endometrial gland cells communicate with maternal endothelia and with fetal cells.

## A34
## GLUCOSE FATE DURING BOVINE COC *IN VITRO* MATURATION

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The aim of this work was to estimate the relative fate of the consumed glucose by bovine cumulus-oocyte complexes (COCs) during in vitro maturation (IVM). Hexosamine biosynthesis pathway (HBP), Glycolysis (G) and pentose phosphate pathway (PPP) were analyzed. IVM was carried out in 199 medium, 5 % FBS, FSH + LH, 5 % $CO_2$, 39ºC for 22 h (control) supplemented with increasing concentrations of DON, NaF or 6-AN, inhibitors of the HBP, G and PPP, respectively. The degree of cumulus expansion, the lactate production and the ability to reduce Brilliant Cresyl Blue (BCB) were used as parameters for the evaluation of the HBP, G and PPP, respectively. Glucose uptake during COC IVM and oocyte meiotic maturation was determined in all cases. The addition of DON had a dose dependent inhibitory effect on cumulus expansion and glucose uptake ($p<0.05$), while no effect was observed on oocyte meiotic maturation. The supplementation with NaF inhibited in a dose dependent manner lactate production, glucose uptake and meiotic maturation ($p<0.05$). With the addition