Proceedings of the
8th International Symposium
on Canine and Feline Reproduction
ISCFR

June 22-25, 2016
Paris, France

In a joint meeting with the XIX EVSSAR Congress

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Expression of steroidogenic enzymes and steroid receptors in fetal gonads of domestic cat – sex similarities and differences.

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Already fetal gonads produce steroid hormones and by this influence the further development of external and internal genitalia as well as of the brain. For many species it was shown that fetal steroid biogenesis usually starts earlier in male than in female gonads, but sexual differences in the receptivity towards steroid hormones in fetal gonads are not so well studied. The aim of the study was to analyze the steroidogenic enzyme expression in fetal gonads of domestic cats, together with determination of steroid receptors. By this, we aimed to determine the time course and sexual difference of endo-/auto-/paracrine actions of steroid hormones during ontogenesis in the domestic cat. For our analysis, we were able to collect pregnant uteri (one per analyzed day) from domestic cat obtained after castration from mid-pregnancy. From each fetus, we isolated both gonads for histological and molecular analysis (d34: n(male) = 4, n(female) = 2; d36: n(m) = 3, n(f) = 3; d39: n(m) = 1, n(f) = 1; d48: n(m) = 2, n(f) = 4). Gene expressions of 17 genes of interest were analyzed by quantitative PCR relative to the expression of reference genes (GAPDH, TBP and RPS7). The histomorphological classification of fetal gonad gender was confirmed by gene expression of sex-determining region (SRY) and Anti-Müllerian hormone (AMH), their corresponding proteins are known as testes-developing factors. For some factors an immunohistochemical analysis was performed. The mRNA expression of the steroidogenic enzymes Steroidogenic acute regulatory protein (StAR), Cholesterol side-chain cleavage enzyme (CYP11A1), 3-beta-hydroxysteroid dehydrogenase/delta(5)-delta(4)isomerase type I (HSD3B1), steroid 17-alpha-monooxygenase (CYP17A1) and hydroxysteroid dehydrogenase type 3 (HSD17B3), were remarkably higher in male gonads (between 10 to 500fold) compared to female ones on all analyzed days. All these genes, except HSD17B3, were detectable in female gonads too, there with a tendency for an increase towards day 48. The HSD17B type 1 and 2 genes (HSD17B1, HSD17B2) were expressed at comparable levels in both sexes, likewise with higher levels on day 48 in female gonads compared to the other tested days. In contrast to the other genes, expression of aromatase (CYP19A1) was detectable in female gonads but almost negligible in male, indicating that only female fetal gonads are capable of aromatizing androgens to estrogens at the studied developmental phases. By immunohistochemical HSD3B1 analysis a strong staining of the interstitium of male gonads was observed, in female gonads undefined small structures exhibited a strong staining. The gene expression of the following nuclear and membrane steroid receptor were analyzed: androgen receptor (AR), estrogen receptor 1 (ESR1) and 2 (ESR2), progesterone receptor (PGR), progesterone receptor membrane component 1 (PGRMC1) and 2 (PGRMC2) and G protein-coupled estrogen receptor (GPER). Slightly higher expressions of AR, PGR and ESR2 in female compared to male gonads on all tested days were detected, only for GPER we observed the opposite; the other receptors were almost equally expressed in both genders. In immunohistochemistry for ESR1, PGR and AR the antibodies react on gonads of both genders. In summary male and female domestic cats gonads exhibit both the enzyme composition to produce steroid hormones, but in males the capacity is noticeable higher. Furthermore hints for an increase in fetal gonad ovarian steroid hormone production towards the third trimester of pregnancy are given. Fetal gonads of both genders express the tested hormone receptors and seem therefore receptive for androgens, gestagens and oestrogens. Gender differences in steroid receptor expression are not strongly pronounced.