

CASA. Viability (V: sperm with intact plasma and acrosome membranes) using flow cytometry (Hoechst-33342/Propidium iodide/FICT-PNA). The PTX concentration did not influence values of TM, PM and V with respect to the control over time. Mean percentages of TM, PM and V in PTX supplemented samples along the incubation time ranged from 72 to 61%, from 56 to 44% and from 67% to 57%, respectively. Increasing PTX concentration in thawing medium negatively influenced quality of movement parameters ( $p < 0.05$ ) from 60 min of incubation. In conclusion, PTX supplementation in thawing medium did not improve sperm quality at any time post-thaw evaluated. Supported by Seneca Foundation, Murcia, Spain (GERM04543/07).

## OC13

### Testicular histological features and sperm quality in healthy dogs of different body weight

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The aim of this study was to define the histological structure of testicular parenchyma in healthy mongrel dogs and the relationships between the seminal quality and the microscopic testicular findings. Two groups were performed: group 1 ( $n = 10$ ), dogs weighting  $<10$  kg and group 2 ( $n = 8$ ), males weighting between 10 and 25 kg. Semen was collected and seminal quality was evaluated (please, add briefly seminal parameters evaluated). Then, males were orchidectomized and testes were processed, assessing different histological parameters: number of seminiferous tubules, seminiferous tubule diameter and number of Sertoli and Leydig cells by seminiferous tubule. Results showed that the semen volume (2nd fraction) and the total number of spermatozoa were higher ( $p < 0.01$ ) in group 2 (0.7 vs. 1.9 ml and  $376.1 \times 10^6$  vs.  $981.2 \times 10^6$  spermatozoa, dogs  $<10$  kg and dogs 10–25 kg, respectively); however, it did not find any significant differences in the other of seminal parameters between groups. In addition, the number of seminiferous tubules, with or without spermatozoa, and the number of Sertoli and Leydig cells were significantly higher in group 1. By contrast, the seminiferous tubules diameter was higher in group 2 (276.5 vs. 306.7  $\mu\text{m}$ , group 1 vs. group 2,  $p < 0.05$ ). This study confirmed that the dog weight has a direct relationship with the testicular parenchyma structure and, therefore, with the sperm production.

## OC14

### A puppy with vaginitis associated with abnormal anatomical position of the vulva

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A reproductive exam is very important in certain pathological conditions in puppies. Anatomical abnormalities or congenital defects

of the vulva, vestibule and caudal vagina can be predisposed to different pathologies such as vaginitis, discharges, dermatitis, foreign bodies, cystic endometrial hyperplasia or even pyometra in an adult bitch. A prepuberal bitch (5 months age, golden retriever) arrived at our clinic with excessive vulvar licking. A reproductive exam showed small sized and cranial disposition of the vulva (making difficult an internal exploration) and irritation around the vulvar skin. Vaginal cytology revealed neutrophilia and anoestrus cycle stage. Prepuberal vaginitis was diagnosed. Treatment recommended was vulva wiping with antiseptic solution and wait for spontaneous resolution after the first heat. However, first heat appeared but the bitch showed the same purulent vaginal discharge and vulvar conformation. Vaginal cytology showed neutrophilia and diestrus phase and abdominal ultrasound showed little liquid content in cervix and caudal uterine lumen. Vaginoscopy revealed a vaginal septum in cranial vagina that was eliminated at that moment. After that uterine liquid disappeared progressively, but vaginal discharge persists for months. The vulvoplasty was recommended after second heat, with excision of a patch of skin around the vulva to assure good appearance of labia. After surgery, the bitch did not show any vaginal symptom.

## OC15

### Staining technique affects the morphological assessment of epididymal feline spermatozoa

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Morphological spermatozoa examination remains a valuable tool to know the quality of feline sperm to be cryopreserved and/or used for artificial insemination. Numerous staining techniques have been employed in this specie, although several variations have been reported. Thirty-six cats were castrated and epididymal sperm samples were obtained. Sperm morphology was assessed using two staining methods, an air-dried stain (Giemsa-Wright) and a wet stain (eosin-nigrosin), in order to determine the proportion of normal and abnormal spermatozoa. The effect of centrifugation (sperm samples were diluted in DPBS medium and centrifuged at  $300\times$  for 8 min) and cooling process (samples were cooled to  $4^\circ\text{C}$ , at a rate of  $0.3^\circ\text{C}/\text{min}$ , using a controlled refrigerator) on the sperm morphology were also analysed. Mean percentages of morphologically normal spermatozoa were significantly lower in eosin-nigrosin-stained samples than in Giemsa-Wright-stained preparations (46.5% vs. 53.4%;  $p < 0.01$ ). Differentiated analysis of head, midpiece and tail sperm abnormalities suggest that these methods differ in their capacity to identify sperm anomalies in cat. Although no significant differences were observed for secondary abnormalities, distal cytoplasmic droplets were significantly lower in Giemsa-Wright stained samples ( $p < 0.01$ ). Centrifugation or cooling process did not affect the epididymal sperm morphology in evaluated samples. This study highlights the necessity to minimize morphological assessment variations in feline spermatozoa to improve the reproducibility in different laboratories.