Germplasm banks are a tool for the preservation of native, rare and/or genetically relevant breeds. We present a partial study of the SERIDA semen bank for the cattle breed "Asturiana de la Montaña", assessing chromatin status (a critical parameter for sperm fertility) in 8 bulls that were sampled in 4 seasons. Two straws were thawed (37°C, 30 s) for each bull and season and analysed by Sperm chromatin Structure Assay (SCSA, flow cytometry) just after thawing and after 5 hr at 37°C (for detecting susceptibility to post-thawing damage), obtaining DNA fragmentation (%DFI) and chromatin immaturity (%HDS). Data were analysed by linear mixed-effects models (results as mean ± SD of %). All values indicate very low chromatin alterations (ranging  $1.0 \pm 0.9$  to  $3.6 \pm 1.0$  for %DFI,  $2.5 \pm 0.4$  to  $6.3 \pm 0.7$  for %HDS). Bull effect was significant in both cases (p < .001). We detected a significant interaction bull  $\times$  season (p < .001), with season being significant in 4 bulls for %DFI (p < .05) and in 2 bulls for %HDS (p < .001). The seasonal dynamics differed between bulls, but changes were small. Incubation induced a small, non-significant increase of %DFI and a decrease of %HDS (p < .1). In conclusion, this survey shows excellent chromatin status after thawing, with no susceptibility to undergo further damage. Chromatin status seems to be an individual characteristic of each bull. Grant: INIA RZP2013-00006-00-00.

## P 11 | Surgical treatment of the intersex condition in the dog

MJ Sánchez<sup>1,2</sup>; JF Pérez<sup>1</sup>; LV Monteagudo<sup>3</sup>; M Pera<sup>1</sup>; I Garcia<sup>1</sup>; A Rodríguez<sup>1</sup>; M Sánchez<sup>1</sup>

 $^1{\rm Veterinaria},$  UCM, Madrid, Spain;  $^2{\rm INIA},$  Madrid, Spain;  $^3{\rm Veterinaria},$  UZ, Zaragoza, Spain

Intersexuality is a rare congenital condition that leads to the development of an ambiguous urogenital tract. Removal of the reproductive tract is recommended to avoid genital diseases; however there is scant information about the procedure. The objective of this report is to describe the surgical procedure used to treat the canine intersex condition. Surgical corrections were performed on four intersex dogs: three 78 XX SRY-negative (one bilateral ovotestis, two sertoli-cell-only syndrome) and one 78 XX SRY-positive (sertoli-cell-only syndrome). Despite the different genetic and histological features, the anatomical characteristics were similar. The patients presented a vulva that lacked of dorsal fold placed ventro-cranially to its anatomically normal position, a hypertrophied os clitoris protruding from the vulva and a urinary opening near to its normal position in the bitch. Under general anaesthesia dogs were positioned in dorsal or sternal decubitus depending on the more o less caudal position of the vulva. Clinical examination revealed a midline orifice at the base of the os clitoris in the medial perineal raphe, which was excised completely to expose the clitoris, the vulvar vault and to verify that the urethral orifice followed a groove previously identified on the clitoris. The study describes the os clitoris excision, closure of the urethral plate to the mucosa of the clitoral fossa and of the vulvar mucosa. The procedure reduced the clinical signs that resulted from the exposure of the clitoris.

## P 12 | Effect of environmental factors on ram sperm quality under tropical conditions

M Carvajal-Serna<sup>1</sup>; JA Cardozo-Cerquera<sup>2</sup>; HA Gajales-Lombana<sup>1</sup>; JA Cebrián-Pérez<sup>3</sup>; T Muiño-Blanco<sup>3</sup>; R Pérez-Pé<sup>3</sup>; A Casao<sup>3</sup>

<sup>1</sup>Department of Animal Production, National University of Colombia, Bogotá, Colombia; <sup>2</sup>Research Center TIBAITATÁ, Colombian Corporation of Agricultural Research-CORPOICA, Colombia; <sup>3</sup>Department of Biochemistry and Molecular and Cellular Biology, Instituto de Investigación en Ciencias Ambientales de Aragón (IUCA), University of Zaragoza, Zaragoza, Spain

Environment can have a negative effect on animal reproduction. This study aimed to determine the relationship between the environmental conditions and sperm quality of three sheep breeds in tropical regions. Semen from twelve adult rams (4 Colombian Creole, 4 Romney Marsh, 4 Hampshire) was collected weekly by artificial vagina for 1 year, and volume, motility (total, progressive and kinematic parameters), viability and morphological normality were evaluated using a CASA system (Hamilton Thorne Research, IVOS, Bedford, MA). Air temperature and relative humidity were monitored each day and the temperaturehumidity index (THI) was calculated. Results were grouped into four environmental seasons; two rainy (Mar-May, Sep-Nov) and two dry (Dec-Feb, Jun-Aug) and analysed by two-way ANOVA followed by Bonferroni post-test. Pearson's correlation between sperm quality parameters and THI was calculated for each breed. Breed and season, affect the sperm quality parameters (p < .05). Likewise, THI correlated positively with morphology in all three breeds and with total motility (p < .05) in Creole and Romney Marsh. In conclusion, THI and breed can influence significantly sperm quality in rams under tropical conditions. Grants: Colombia 110157635854, Col576-2012, Spain AGL2014-57863-R.

## P 13 | Seasonal variation of melatonin synthetizing enzymes and melatonin receptors in ram testes

JA Cebrián-Pérez; S Lázaro-Gaspar; P Martínez-Marcos; R Pérez-Pé; T Muiño-Blanco; A Casao

Department of Biochemistry and Molecular and Cellular Biology, Instituto de Investigación en Ciencias Ambientales de Aragón (IUCA), University of Zaragoza, Zaragoza, Spain

Pineal melatonin is known to regulate seasonal reproduction in sheep, but melatonin can also be synthetized in extrapineal tissues, such as the testis. We have recently reported the presence of the melatonin synthetizing enzymes serotonin-*N*-acetyltransferase (AANAT) and *N*-acetylserotonin *O*-methyltransferase (ASMT), along with melatonin receptors MT1 and MT2 in ram testes. The objective of this study was to determine whether the gene expression of these enzymes and receptors present seasonal variation. Testis from 27 adult rams were collected from the slaughterhouse (13 during the reproductive season and 14 during the non-reproductive season), and RNA was extracted from testicular tissue by the TRI-reagent protocol. After c-DNA