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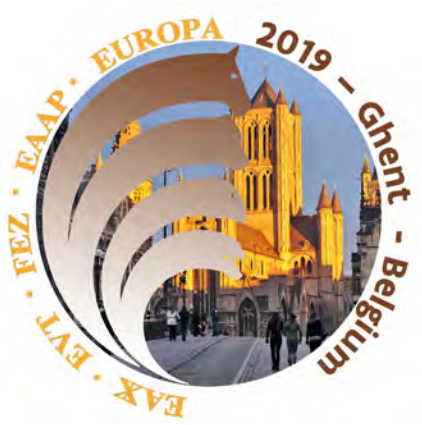

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Assessing pubertal age through testicular and epididymal histology in Bísaro pig

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Bísaro pig (BP) had grown in numbers in the last decade, representing one of the most important native Portuguese breed. This study aims to estimate the age of puberty in male BP through testicular and epididymal morphometry. Fifty-six pairs of testis and epididymis were collected from male BP ranging in age from 1 to 8 month-old. Samples were collected post-mortem (n=26) or from surgical castration (n=30), from May 2017 to April 2018, sourced from six different farms. After collection, testis and epididymis were trimmed, weighed and measured. Tissue samples were processed for paraffin embedding and routine haematoxylin–eosin staining. Studied parameters included spermatogenesis scoring (SS), the diameter of seminiferous tubule (DST), the density of Sertoli (DS) and Leydig (DL) cells, the diameter of Leydig cells (DLC) and nucleus (DLN), and the ratio between tubular/interstitial areas (RTI). Correlations between testicular and epididymal length, width, depth, weight and volume, were highly positive (r: 0.866-0.997; P<0.001; n=56). Positive correlation was also found between DLC and DLN (r: 0.732; P<0.001; n=52). Differently, DST increased proportionally to the animal's age (R2: 0.69; P<0.001; n=52) varying from 52.91 µm to 241.95 µm. RTI acted similarly increasing in older animals (R2: 0.76; P<0.001; n=52). It varied from 25.36 to 77.56%. On average, tubules have 17.52(4.27) sertoli cells with a density varying from 2.32 to 86.45 cells/µm. While DS decreases (R2: 0.72; P<0.001; n=52), DL increases (R2: 0.32; P<0.001; n=52) as the animals gets older. A GLM model was used to predict average testis dimensions and animal's age at pre-defined stages; when SPZ is found in the epididymis, testis are 6.88 cm length and 4.49 cm width, at 118.21 days. At this age, the most likely median SS is 6 and the mean predicted SS is 5.29. When SPZ is found in the vas deferens testis are 7.46 and 4.82 cm, at 145.74 days. The most likely median SS is also 6 and the mean predicted SS is 6.07.