

## **PROLACTIN IN SALIVA OF PIGS AS A POSSIBLE BIOMARKER OF STRESS: ANALYTICAL VALIDATION OF AN IMMUNOASSAY AND CHANGES AT SLAUGHTERHOUSE**

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Prolactin (PRL) is considered a biomarker of stress response, both acute and chronic, in several species. Although the scientific literature reports divergent results, some studies suggest that prolactin secretion during stress acts to maintain homeostasis within the immune system. Several studies have investigated the possibility of measuring PRL in saliva in different species such as rats, domestic ruminants, donkeys, dogs, cattle, and sheep for diagnosis or research purposes. Results obtained in animal models have indicated that the secretion of this hormone is dependent on the type and intensity of the stress. However, to our knowledge, PRL has not been measured in the saliva of pigs, species in which is especially appropriate due to its non-invasive collection. The objective of this study was to validate a commercial immunoassay for measuring PRL in porcine saliva and, to evaluate how prolactin can change in saliva after a model of stress based on transport, approximately 30 min, to the slaughterhouse. Saliva samples were collected using saliva collection tubes (Salivette®) and synthetic sponges of pigs from a commercial farm in Totana, Murcia, Spain. An analytical validation was made of pig PRL enzyme immunoassay (Cusabio®). In addition, salivary PRL was measured in ten pigs (5 male and 5 female) at different times: before the transport (BT), at the time of arrival (T0) immediately after unloading at the slaughterhouse, and 4h after arrival to the slaughterhouse (T4). PRL assay showed an average of intra- and inter-assay coefficient of variation < 12%. Linearity under dilution presented a linear regression equation with  $r^2$  close to 1 and recovery percentage ranged from 90%. The limit of detection was 0.23 pg/mL. Salivary PRL was lower ( $P<0.05$ ) at T0 (median 0.79 (0.58<sup>25th</sup>-1.12<sup>75th</sup>) pg/mL) and at T4 ( $P<0.001$ ) (median 0.37 (0.32<sup>25th</sup>-0.40<sup>75th</sup>) pg/mL) as compared with BT (median 1.26 (1.05<sup>25th</sup>-1.70<sup>75th</sup>) pg/mL). In conclusion, PRL can be measured in saliva of pigs with the immunoassay used in this study that presented adequate precision and accuracy. In addition, although more studies are needed because of the low number of animals used, PRL showed a decrease after transportation and remains low during 4 hours at slaughter, indicating that it could be a biomarker of stress.