

Evaluation of salivary cortisol as a biomarker for plasma cortisol during ACTH challenge and at variable salivary consistency

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Modern and sustainable dairy farming implies the consideration and improvement of animal welfare. Non-invasive sampling is crucial for unbiased results. The implementation of feasible biomarkers, however, must be robust to various physiological and environmental conditions. Two experiments were performed to validate the suitability of saliva cortisol measurement: the first during stimulation of the adrenal glands and the second one during various events affecting the composition and consistency of saliva. In experiment 1, twenty-three multiparous Holstein dairy cows were fitted with an indwelling jugular catheter in week 3 postpartum. ACTH (16 µg/100 kg body weight) was administered i.v., and blood was sampled in parallel to saliva (collected by Salimetrics swabs) at 15 minutes intervals for 3 h relative to ACTH injection. Experiment 2 aimed to evaluate the effects of variable saliva consistency and composition on its cortisol concentration. Saliva and blood samples were taken in parallel from one cow before, during and after (5 and 15 minutes) drinking, feeding and ruminating. Each saliva sample was taken immediately after the respective blood sample. Cortisol in saliva was measured by EIA (Salimetrics) and cortisol in blood plasma by RIA. In both experiments, the profiles of cortisol concentrations were similar in blood plasma and saliva. Saliva and blood cortisol concentrations were closely correlated during the ACTH challenge ($r = 0.75$) with significantly increased plasma cortisol concentrations following ACTH administration when compared to basal pre-injection values. Cortisol concentrations in plasma were consistently higher compared to saliva during the ACTH challenge. In saliva samples taken during different feeding and drinking actions, cortisol concentration in saliva was highly correlated to the cortisol concentration in blood ($r = 0.90$). Thus, saliva cortisol concentration is not affected by salivary composition and consistency. This is an important finding emphasizing the power of saliva cortisol mirroring plasma cortisol for non-invasive stress estimation. In conclusion, saliva can be confirmed an ideal non-invasive substrate for robust cortisol determination in dairy cows, independent of physiological status and consistency of saliva.

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