Reactivity in ewes submitted to invasive and non-invasive techniques of samples collections

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The first-time experience of a procedure or with a person should be as positive as possible. If a procedure is very aversive or painful at first contact, it can be difficult to persuade the animal to return to the local where this occurred. For this reason it is important to develop a strategy in experimental and routine activities, taking into account that sheep has often reactive temperament. This study aims to evaluate the possible decreased of the reactivity in sheep across the prior presentation of invasive and non-invasive techniques for collecting saliva and blood samples for cortisol analysis and transepidermal water loss through the use of Vapometer®. Saliva was collected using Sallivetes® and blood was taken from the jugular vein in heparinized tubes, placed on ice, and then centrifuged at 4 °C and 3,000 for 15 min. The tubes containing serum were stored at −20 °C until CORT and IGF-I determination using an enzyme immunoassay kit (Diagnostic Systems Laboratory, Webster, TX). The collection of transepidermal water loss (TEWL) was performed using a Vapometer ® (Dolphin, Finland) device. In the three techniques the ewes were always contained by the same person for the time necessary to collect the variable. To take saliva samples the animals were contained during 2 minutes, for blood samples during 1 minute and for TEWL during 30 seconds. The test consists of six data collections performed with increasing intervals between days, checking the possible adaptation of the animal management factors. The first interval was 2 days and after 3 days, then after 4 days, and the next was 7 days and again 7 days, totaling 24 days of experiment. Before sampling saliva, blood or TEWL the reactivity of the ewes was analyzed by a composite score scale when animals were inside the chute. The composite score (CS) is a combination of the scores of: entrance (1 to 4), respiratory rate (1 to 4), vocalization (0 or 1), movement (1 to 4) and flight speed (1 to 4). After the chute a score of containment (1 to 4) was attributed by the same person, which had contained the animal. The experimental unit was each animal and the average of the observations in each sheep was used for the statistical analysis. The observed data of transepidermal water loss, salivary cortisol and serum cortisol were subjected to analysis of variance, with the days and the groups as fixed effect. In case of significant results ($P<0.05$) it was adopted Tukey-Kramer Test as the procedure for multiple comparisons. Cortisol decreased for saliva and blood groups during the experiment days ($P<0.05$), showing habituation of the animals to the management. In the same way, TEWL had the highest mean value on day 1, and decreased afterwards ($P<0.05$). The score of containment was lower for the TEWL group compared to the other two groups ($P<0.05$), however the higher value was observed on day 5, seven days after a sampling ($P<0.05$). The score of containment of saliva and blood groups decrease in day 2, but return to increase in the next sampling day ($P<0.05$). The CS was higher for saliva group ($P<0.05$). On day 5 CS increased for blood group, but decreased for TEWL ($P<0.05$). The invasive techniques (saliva and blood) increased the reactivity of sheep before and after retention. Sheep are reactive animals therefore need a more gentle handling during the experiment with invasive samples.

Key-words: Cortisol, free-stall; IGF-I, rectal temperature stress.