## HOW DIFFERENT HUSBANDRY CONDITIONS INFLUENCE GUT MICROBIOME, CORTISOL LEVEL AND BEHAVIOUR IN LAMBS?

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The gut microbiota-brain axis is a bidirectional pathway through which the brain regulates the activity of the gut and vice versa. The intestinal microbiota was proven to influence and be influenced by stressrelated responses caused by management conditions. The aim of this work was to evaluate whether different husbandry conditions in lambs influenced gut microbiome (composition and diversity), cortisol level and behaviour. Fifteen Sarda breed lambs, aged 6 months, were randomly assigned to three groups of 5 animals each: Isolation (tactile and visual isolation; 3 h/day for 8 weeks); Enriched (in group in a outdoor grassy pen with a new enrichment each week; 3 h/day for 8 weeks); Control (habitual farm environment). Every week, 2 video-recordings of 1-hour each were collected respectively during and after each treatment, and analysed using a scan sampling technique (60 scans per video). Every two weeks hair was collected from the right shoulder using a shave and re-shave method. Cortisol concentration was measured by means of an ELISA test conducted on hair samples and expressed as pg/ml. At week 8 faecal material was collected directly from the rectal ampulla and immediately frozen at -20°C until DNA extraction was performed using a QIAmp DNA Stool kit (Qiagen, Hilden, Germany). DNA quality and quantity were assessed using a NanoDrop ND-1000 spectrophotometer (NanoDrop Technologies, Wilmington, DE, USA). The microbiota of enriched animals was clearly different from the other two groups and showed lower within group variability. During treatment, only isolated sheep showed escape attempts (23% of scans during the first week) and freezing (58% of scans during the first week). In the home pen, after treatment, lambs did not show any stress-related behaviour. After one month of treatment, hair cortisol increased only in the isolated group. In conclusion, differences in microbiome could be related to the different stimulating enrichment and addition of grass on the diet. Although, from a behavioural point of view, sheep apparently habituated to the isolation, increase in cortisol levels could be related to experiencing a stressful situation.

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