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Research

Behavior of lambs at different ages during brief periods of increased sensorial isolation from their mothers



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ABSTRACT

To determine the behavior of different-aged lambs during short periods of increased sensorial isolation from their mothers, 6S0 ewe-lamb pairs were randomly assigned to one of the following groups: "C," intact lambs that remained with their mothers; "A," lambs with restricted auditory capacity; "A+V," lambs with restricted auditory and visual capacities; "A+V+W," lambs with restricted auditory and visual capacities plus separation from their mothers by an open wire mesh; and "A+V+S," lambs with restricted auditory and visual capacities plus separation from their mothers by a solid wall. These treatments were repeated in the same animals when lambs were $3 (G_3)$, $10 (G_{10})$, and $20 (G_{20})$ days old. The behavior of lambs was recorded during each 15-minute separation period, and data were examined using covariance analysis to determine differences for repeated measures over time. In general, after the physical separation from the ewes (A+V+W and A+V+S groups), lambs emitted more vocalizations (3, 10, and 20 days old) and displayed more frequent urination (3 days old), exploration of objects (20 days old) and conspecifics (3 and 20 days old), escape attempts (A+V+W at 10 and 20 days of age; A+V+S at 20 days old), or walking (A+V+W at 10 and 20 days of age; A+V+S at 3, 10 and 20 days old) and lying down (10 and 20 days old for A+V+W and 3 days old for A+V+S) in comparison to the other treatments. G_{10} and G_{20} lambs of the A+V, A+V+W, and A+V+S groups also displayed greater values for walking activity (P < 0.05) in comparison with G₃ lambs. In general, it is concluded that the exhibition of the examined behavioral parameters was increased with the degree of sensorial isolation, and particularly when the physical contact between the lambs and their dams was restricted. As lambs became older, they showed an increased behavioral reactivity in response to the increased sensorial isolation.

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Introduction

Lambs stay with their mothers during the first weeks after birth (Morgan et al., 1974) when ewes display various signals that trigger specific behavioral patterns in their newborns. In turn, young lambs exhibit a variety of signals that induce care-giving responses from their mothers (Terrazas et al., 2002). Efficient communication

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between ewe and lamb is crucial for the survival and development of the lambs. This communication includes acoustic, olfactory, visual, and tactile stimuli. The ewe-lamb bond is reinforced after birth through their interaction during the first few minutes postpartum, and this early period appears to be critical for the survival of the lambs. After studying some of those communication stimuli, Morgan et al. (1974) concluded that ewes recognize their lambs primarily by sight, hearing, and smell. Other studies have demonstrated that newborns also use acoustic signals to catch their mothers' attention or alert their conspecifics from dangerous situations (Rödel et al., 2013). In addition, ewes tend to keep their lambs in close proximity for several days after birth and offer the care they require, a behavior that intensifies their bond. At first, this bond seems stronger for the ewe but decreases as the lamb grows, whereas in the lambs, the opposite is true (Maldonado et al., 2015), though individual variation exists (Galeana et al., 2007).

Sheep are, by nature, gregarious animals, so isolation is perceived as a threat to their homeostasis (Moberg, 2000), learning (Bouissou and Gaudioso, 1982), and emotional reactivity (Doyle et al., 2009), in such an extent that any restriction during the early stages of their life may have consequences on several behavioral, hormonal, and immunological indicators (Orgeur et al., 1998; Orihuela et al., 2004). As a result, when separated, both ewes and lambs manifest distress reactions that are also followed by adrenal responses. Appeasement of the distress is obtained only on being reunited with the attachment figure, that is, the ewe (Nowak et al., 2011). Alexander (1977) found that when lambs are separated from their mothers even temporarily, they show clear signs of stress, including a higher frequency of bleating and increased locomotion, 2 behavioral parameters that demonstrate their efforts to maintain contact with the ewes by locating, encountering, and identifying them. Moreover, Hinch et al. (1990) observed a general increase in the spatial distance between family members at older ages, a finding that was interpreted as reflecting a gradual decline in the intensity of attachment over a period of 2.5 years.

Despite the conclusions of the former studies, our understanding regarding the behavioral consequences of short periods of restriction on the sensorial signals displayed by dams and nursing lambs is still relatively poor. This information could be of practical importance because in several management practices, lambs are separated from their mothers during short periods at early ages (Kent et al., 1998; Mellor and Stafford, 2000). Thus, our objective was to determine the behavior of nursing lambs subjected to different sensorial impairment at different ages. In detail, 2 hypotheses were tested: (1) Do lambs manifest greater behavioral reactivity as the degree or intensity of sensorial isolation increases? and (2) Are younger lambs less affected by the separation from their dams?

Materials and methods

The study was conducted at a research station in the National Autonomous University of Mexico located at $99^{\circ}11'42''$ W and $19^{\circ}41'35''$ N, with a mean temperature of 15.7° C.

Animals

Sixty multiparous Columbia ewes aged 2-5 years old, and their single-born male lambs, were housed permanently under semiintensive handling conditions at a density of 10 m²/animal. Diet consisted of alfalfa hay, commercial concentrate mix, and minerals according to the animals' physiological needs. Water was *ad libitum* offered. In addition, from 10:00 to 16:00 h, ewes were left to pasture in alfalfa meadows. After birth, the ewes remained with their newborns as a group in a single pen. The experiment consisted of 3 phases according to the lambs' age: 3, 10, and 20 days. At 48 hours of age, lambs were earmarked, sexed, and each ewe-lamb pair was identified by painting a number with a nontoxic aerosol on their sides. This number indicated the treatment group to which they were randomly assigned. In case of the 10- and 20-day-old lambs, 24 hours before testing, ewe-lamb pairs were re-marked.

Experimental groups

All lambs were tested between 9:00 and 10:00 h for 15 minutes at the ages of 3 (G_3), 10 (G_{10}), and 20 (G_{20}) days. They were randomly allocated into 5 treatment groups (n = 12 ewe-lamb pairs each). In the control group, "C," ewes and lambs remained in constant contact in the same pen, free to generate and receive all sensorial stimuli. In group "A," lambs were only subjected to auditory restriction, where the handler placed cotton plugs in the lambs' ears, which were then covered with a headband to reduce perception of acoustic stimuli. In group "A+V," lambs were subjected to both auditory + visual restriction, where in addition to the previous treatment, lambs were also visually restricted to impede reception of visual stimuli. In group "A+V+W," in addition to the auditory and visual restrictions, the lambs were physically separated from their ewes by an open wire mesh that divided the pen, whereas in the "A+V+S" group, lambs were treated as in the previous group but the open wire mesh was replaced by a solid wall.

Behavioral observations

For each test, the 12 pairs of ewe-lambs were moved to a 4×4 m pen located 100 m from their home pen. Preparation of the lambs according to its respective restriction regimen lasted less than 3 minutes. All the preparatory works were performed by the same person to avoid variations in handling, and after this period, behavioral parameters were recorded for a period of 15 minutes. At the end of the observation period, the lambs returned to their dams.

The behavioral responses of the lambs were continuously recorded using 2 portable *SONY* Handycam video cameras for later analysis. One camera was placed on top of the pen to provide a panoramic view of the entire available area, whereas the second one provided a lateral view of the pen. Videos were analyzed using Observer TM XT Software, version 10.0 (Noldus IT, The Netherlands). The examined behavioral variables were as follows: the number of vocalizations, urinations and defecations, exploration of objects, exploration of conspecifics, escape attempts, number of walking and lying episodes, defined as in the ethogram illustrated in Table 1.

Statistical analysis

Descriptive statistics were obtained for all the examined variables using the SAS statistical package (PROC MEANS) (SAS, 2004). Normality tests were performed for all variables in all groups and the 3 ages, using the SAS 9.2 program (PROC UNIVARIATE) (SAS, 2004). Because the study analyzed the responses of the same lambs at 3 ages, data were assessed using a mixed model appropriate for repeated measures (SAS, 2004). The model included the treatment, the time, the replication of each treatment, as well as the interaction between treatments and time, and the interaction between treatments and replication for each treatment as fixed effects. The lamb in each treatment and repetition was considered as a random effect. Differences between treatments and time were detected using the Tukey-Kramer test. In all cases, the level of significance was set at $\alpha = 0.01$. The researchers who conducted the

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