frontiers in Animal Science

REVIEW
published: 17 February 2022
doi: 10.3389/fanim.2022.823188

What Do We Know and Need to Know About Weaning in Sheep? An Overview of Weaning Practises, Stress and Welfare

Aline Freitas-de-Melo[1]*, *Agustín Orihuela*[2], *Maria José Hötzel*[3] and *Rodolfo Ungerfeld*[1]

[1] Departamento de Biociencias Veterinarias, Facultad de Veterinaria, Universidad de la República, Montevideo, Uruguay,
[2] Facultad de Ciencias Agropecuarias de la Universidad Autónoma del Estado de Morelos, Cuernavaca, Mexico,
[3] Laboratório de Etologia Aplicada e Bem-Estar Animal, Departamento de Zootecnia e Desenvolvimento Rural, Universidade Federal de Santa Catarina, Florianópolis, Brazil

Artificial weaning is the separation of the mother and her lamb, involving the forced ending of suckling and milk supply by the ewes to their lambs. Artificial weaning time varies depending on the purpose of each sheep farm and the characteristics of the lambs, including age and weight of the lambs, and their ability to eat solid food. Artificial weaning is performed from 24 h postpartum until around the age of natural weaning (up to 12 mo of age). Artificial weaning results in the breaking of the ewe-lamb bond, the end of suckling and the complete replacement of milk by solid food, frequently added to changes in both the physical and social environment. It triggers behavioural, physiological and immunological changes that generate negative consequences on the health of both ewes and lambs, as well as on the growth of the lambs, rising animal welfare concerns. As artificial weaning is considered one of the most stressful situations in the life of farm mammals, it is important to consider the factors involved in those responses to develop adequate strategies and improve sheep welfare at weaning. The most frequently used method is the abrupt separation of the ewes and their lambs. However, the adaptation of the animals after weaning can be improved by alternative weaning methods (e.g., progressive, two-steps weaning and mixed regimes), hormonal treatments (e.g., administration of progesterone), and pre- and postweaning management (e.g., the use of creeping feeding, and the presence of conspecifics or humans after weaning). This review presents and summarises information on the stress response and animal welfare issues at artificial weaning in sheep and some management practises that can be used to reduce these problems. The major points included are the ewe-lamb bond at birth and during the lactation period; the stress response at weaning; the factors involved in the stress response at weaning; the different types and weaning methods and their repercussions on animal welfare; and some strategies that can be used to improve sheep welfare at weaning. Lastly, we propose directions for research needed to develop practical alternatives to minimise sheep's stress response at weaning.

Keywords: separation, weaning method, distress, behaviour, ewes, lambs, filial attachment, physiology

OPEN ACCESS

Edited by:
Edward Narayan, The University of Queensland, Australia

Reviewed by:
Gregory John Lynn Sawyer, The University of Sydney, Australia
Seyed Abbas Rafat, University of Tabriz, Iran

***Correspondence:**
Aline Freitas-de-Melo
alinefreitasdemelo@hotmail.com

Specialty section:
This article was submitted to Animal Physiology and Management, a section of the journal Frontiers in Animal Science

Received: *26 November 2021*
Accepted: *13 January 2022*
Published: *17 February 2022*

Citation:
Freitas-de-Melo A, Orihuela A, Hötzel MJ and Ungerfeld R (2022) What Do We Know and Need to Know About Weaning in Sheep? An Overview of Weaning Practises, Stress and Welfare. Front. Anim. Sci. 3:823188. doi: 10.3389/fanim.2022.823188

INTRODUCTION

Sheep are precocial species that generally deliver 1–2 newborns, that can see, hear, stand up, and follow the mother shortly after birth. Ewes establish a selective bond with their lamb within a few hours after birth (Poindron et al., 2007; Mora-Medina et al., 2016), nursing only those that were recognised as their own offspring. The ewes' interest in their lambs is greater after birth, and declines gradually along the lactation period, as the lambs gain social and nutritional independence (Weary et al., 2008; Maldonado et al., 2015). Artificial weaning is performed before natural weaning age, which has been reported to occur at up to 12 mo of age in wild sheep (Geist, 1971; Grubb, 1974), and is characterised by the complete replacement of suckling by solid food or artificial rearing with sheep milk or milk substitute (Napolitano et al., 2008; Weary et al., 2008). Weaning age varies according to the production goal and the characteristics of the lambs, like age, weight and ability to eat solid food (Napolitano et al., 2008). In dairy sheep, lambs are often precociously weaned, 1–2 days after birth (Napolitano et al., 2008). Meat and wool sheep farms use early, traditional or late weaning, which is done from 30 days of age until around natural weaning age.

Artificial weaning may be an abrupt or a gradual process. Abrupt weaning involves nutritional, social, and emotional changes that entail an important stress response on ewes and their lambs (Freitas-de-Melo and Ungerfeld, 2016a). Abrupt weaning is associated with decreased weight gain, increased alertness, and locomotion behaviours, and decreased resting and feeding behaviours of the ewes and lambs (Damián et al., 2013; Freitas-de-Melo et al., 2017, 2019). Abrupt weaning also influences ewes and lambs' immunological status (Napolitano et al., 2008), and predisposes animals to diseases like mastitis and parasitosis. Multiple factors influence the stress response at weaning, including age, sex, type of birth, genetic, the mother's nutrition during gestation, and the lambing season. Understanding how these factors affect the weaning response is essential when adapting managements around weaning. Some weaning methods that are characterised by a more gradual process, such as progressive weaning, two-steps weaning, and mixed regimes are developed in an attempt to improve sheep welfare. These methods are based on gradually separating the lamb from the mother or limiting the access to the mothers' udder before abrupt separation. Furthermore, some treatments or managements can be applied before and/or after weaning to help ewes and lambs to cope better with weaning stress. Some examples are inducing ewes into oestrus on the day of separation (Pérez-León et al., 2006), the presence of conspecifics or humans with a close emotional bond (Barton, 1983a,b; Carbajal and Orihuela, 2001), applying positive stimulus as stroking (Pascual-Alonso et al., 2015), or pharmacological treatments to decrease the stress perception, as progesterone treatments (Freitas-de-Melo et al., 2013; Freitas-de-Melo and Ungerfeld, 2016b).

This review summarises information about the stress response and animal welfare issues related to artificial weaning in sheep, and some possible managements to decrease these problems. The major points included are the ewe-lamb bond at birth and during the lactation period; the stress response at weaning; the factors involved in the stress response at weaning; the different weaning types and methods and their consequences on animal welfare; and some strategies to improve sheep welfare at weaning. Lastly, we propose directions for research to develop practical alternatives to minimise sheep's stress response at weaning.

EWE-LAMB BOND AT BIRTH AND DURING LACTATION

In mammals, the establishment of a strong and lasting social bond between the mothers and their offspring ensures the provision of care, immunity and nutrition, which is essential for the survival of the offspring and to maximise the mothers' reproductive success (Trivers, 1974; Godfray, 1995; Nowak, 2006). The mother-young bond has been defined as a preferential mutual emotional attachment of relatively long duration that resists temporal separations (Newberry and Swanson, 2008). It is characterised by affiliative behaviours such as licking, provision of food, warmth and protection, rest in company, synchronisation of many activities, and maintenance of closeness and body care (Newberry and Swanson, 2008). In sheep, a precocial species, the dam develops an exclusive relationship with the newborn, i.e. caring only her own lamb(s) or those that after birth she recognises and accepts as her own lambs, rejecting any others that attempt to suckle later (Poindron, 2005). This bond is established within a few hours after birth (Poindron and Neindre, 1980), during a sensitive period when the ewe displays an attraction for newborn lamb(s), favouring maternal attachment for her lamb(s) or those she recognises as her own during this period (Poindron and Neindre, 1980). When the space allocation allows, the ewe tends to separate from the flock close to the parturition and select a place where she remains with her lamb(s) for some time (Shackleton and Haywood, 1985). This relative isolation helps ensure exclusive recognition of her own offspring, although it appears not to occur in some breeds (da Porciuncula et al., 2021).

The initial recognition process is under physiological control and ensures that even inexperienced ewes recognise and care their lamb(s). The decrease in progesterone concentrations and rise in oestradiol concentrations produced by the placenta at the end of gestation triggers maternal behaviour, which acts in synergism with the release of central secretion of oxytocin stimulated by the delivery of the foetus(es) (Kendrick and Keverne, 1991; Shipka and Ford, 1991; Dwyer, 2014). Oxytocin plays a crucial role in triggering maternal behaviour and is further potentiated by other neuropeptide systems, such as opiates (Lévy and Keller, 2008). Ewes deprived of their young after birth cease displaying maternal behaviours, showing that the newborn's presence plays an essential role in the bonding process (Poindron and Neindre, 1980). Maternal selectivity is established within 2 h after delivery, and later the ewe can recognise her lamb and reject alien lambs (Smith et al., 1966; Poindron, 2005). The sense of olfaction is well developed in sheep and is essential for social recognition in the species (Lévy et al., 1995). The volatile compounds of the amniotic fluid attract the ewe and initiate the bonding and recognition process. The presence of fluids after

birth stimulates the ewe to lick the lamb, motivating teat-seeking behaviours, olfactory learning, and colostrum ingestion by the lamb (Nowak, 2006). The length of the first suckling is related to the latency for the mother to recognise her lamb (Nowak and Poindron, 2006).

Lambs' survival depends on their ability to discriminate their mothers within the first 12 h after birth (Nowak and Lindsay, 1992). The lamb searches for the teat and suckles soon after birth, and the first ingestion of colostrum occurs within the first hour (Nowak et al., 1997). Suckling is essential to establish the bond between the lamb and the ewe, and the ingestion of colostrum mediates the establishment of an exclusive preference for the mother (Goursaud and Nowak, 1999; Val-Laillet et al., 2004). In the first hours following birth, the newborn lamb associates suckling with the presence of its mother, increasing its motivation to seek her company (Nowak et al., 1997). Preventing lambs from suckling for the first hours after birth disrupts or delays the development of a preference for the mother (Alexander et al., 1986; Nowak et al., 1997, 2021). Recent work by Nowak et al. (2021) showed that suckling within 2 h after birth triggers the early bonding of the lamb with the mother. In the same study, Nowak et al. (2021) demonstrated that exposure to an oxytocin antagonist impairs the relationship with the dam, demonstrating a role for the oxytocinergic neural system in shaping the early development of attachment in sheep.

Body weight, body fat, and vigour at birth influence the ability of the lamb to display all the behaviours required to establish the bond with the dam. A delay in this process has negative consequences on lambs' metabolic and hormonal status (Alexander et al., 1986; Nowak et al., 1997, 2021). Several conditions affect the behaviours implicated in neonatal attachment in sheep, including environmental temperature (Fonsêca et al., 2014); factors related to the ewe, like status during conception (dry or lactating) (Ungerfeld et al., 2021a), nutrition during gestation (Dwyer et al., 2003; Freitas-de-Melo et al., 2015a), breed (Lawrence and Dwyer, 1999; Pickup and Dwyer, 2011), litter size (Dwyer and Lawrence, 1998; Hernandez et al., 2009), as well as some intrinsic factors of the lamb, like sex (Hernandez et al., 2009; Freitas-de-Melo et al., 2015a; Gaudin et al., 2015), and body temperature (Menant et al., 2020). Understanding the physiological and behavioural consequences of these conditions, how they synergise with each other, and how they can be changed with management is essential to increase lambs' survival probability.

The primary mechanisms for the establishment of the ewe-lamb(s) bond involve olfaction (Morgan et al., 1985), and after initial recognition, behavioural mechanisms that depend on audition and vision are deployed (Alexander and Shillito, 1985; Ungerfeld et al., 2021b). As lactation progresses, the lamb and ewe are not always close to each other, so they require mechanisms to locate and call each other. Vocal and visual recognition between the ewe and her lamb is established early and is essential for maintaining the bond and ensuring appropriate care as the lactation advances. Ewes emit bleats within 3 h before and after parturition (Sèbe et al., 2007), and by 48 h after birth, the lambs can identify the low-pitched bleats of their dam (Sèbe et al., 2010). Ewes, in turn, can discriminate their own lamb from an alien lamb by vocalisation as early as 24 h postpartum (Sèbe et al., 2007). During the lactation period, ewes and lambs still recognise each other through their vocalisations, from around 20 to 90 d of age (Poindron and Carrick, 1976; Hague et al., 1982; Ungerfeld et al., 2021b). The behaviour of the dam (Terrazas et al., 2002; Dwyer et al., 2003) and the lamb (Alexander and Shillito, 1985) is essential for the recognition, highlighting the role of vision in the process. It has been reported that the lamb's head is an essential visual cue in recognition of the lamb by its mother (Alexander and Shillito, 1985). Although younger lambs can distinguish between face and non-face pictures (Salva et al., 2014), they do not recognise the picture of their mothers' faces at 90 d of age (Ungerfeld et al., 2021b).

Once established, the mother-young bond lasts for months. Ewe-lamb bond implies a close physical relationship during the first days after lambing (Ozella et al., 2022). Several factors may influence the strength of the attachment in lambs [e.g., sex, with female lambs showing a stronger attachment (Gaudin et al., 2015), birthweight (Menant et al., 2020)], and in ewes [e.g., parity (Dwyer and Lawrence, 2000; Freitas-de-Melo et al., 2021a), breed (Lawrence and Dwyer, 1999; Pickup and Dwyer, 2011), and temperament (Catherine and Alistair, 2000)]. In early postpartum, ewes have a strong preference for their lamb; but as the strength of the preference decreases, the preference of the lamb for the dam increases (Hernandez et al., 2009; Maldonado et al., 2015). This change in social behaviour characterises the process of natural weaning and is associated with the decrease in suckling frequency, the increase in solid food consumption, and the waning of the bond (Weary et al., 2008). During the first 1 or 2 wk postpartum, the ewes allow their lambs to suckle as often as they want (Munro, 1956; Hinch, 1989). Throughout lactation, most suckling bouts are initiated by the lambs, but terminated by the ewes (Hinch, 1989), except for short suckles, usually terminated by the lambs, which are probably more related to the maintenance of the mother-young bond than to feeding (Shackleton and Haywood, 1985). As the lactation advances, the ewe prevents the lamb from suckling more frequently (Fletcher, 1971; Fonsêca et al., 2016), and the physical distance between the ewe and her lamb increases (Hinch et al., 1987; Hinch, 1989; Galeana et al., 2007).

In the natural weaning process, the reduction in milk production by the dam plays a central role in determining the progression of the process (Arnold et al., 1979). During the first week of life, lambs suckle once or more times per hour (Munro, 1956). As lactation advances, the duration of suckling and consequently milk production by the dam decrease, and in parallel, the intake and time eating solid food by the young increase (Arnold et al., 1979; Freitas-de-Melo et al., 2018a). This reduction in suckling frequency, associated with the increase in solid feeding, favours rumen development (Lyford, 1988). As the lamb grows, it gains not only nutritional but also social independence. The gradual change from milk to solid food favours an increase in social interactions and the time spent in company of other lambs in the group (Weary et al., 2008). The physical distance between the ewe and her lamb increases from birth until natural weaning occurs (Hinch et al., 1987), which occurs between 6 and 12 mo of age in wild sheep (Geist, 1971;

Grubb, 1974). Even after weaning, lambs prefer the company of their mother for more than 1 month after separation (Hinch et al., 1987; Ungerfeld et al., 2018), but after that, they do not show an important motivation to seek for their mother (Ungerfeld et al., 2018).

ARTIFICIAL WEANING IN SHEEP

Artificial weaning is the separation of the mother and her lamb, involving the forced ending of suckling and milk supply by the ewes to their lambs. Artificial weaning time varies depending on the purpose of each sheep farm and the characteristics of the lambs, such as availability of food, age and weight of the lamb, and its ability to eat solid food (Napolitano et al., 2008). Artificial weaning is performed from 24 h postpartum until around the age of their natural weaning. According to the age of the lamb, artificial weaning can be classified as precocious, early, traditional, and late (**Table 1**). In sheep milk production systems, artificial weaning is done early to increase the amount of milk collected by the farmer and thus, obtain more dairy products; however, it requires the artificial rearing of lambs with milk substitutes (Napolitano et al., 2008). In other systems, weaning is performed to improve the body reserves of the ewes after the end of lactation, increasing the reproductive results at the following breeding season, including fertility, prolificacy, and birth weight of their subsequent offspring (Vatankhaha and Salehi, 2010; Vatankhah et al., 2012).

Precocious Weaning

Dairy sheep farms often use precocious weaning, allowing lambs to suckle the colostrum and then separating the lambs from the dams abruptly 24 to 48 h postpartum (Napolitano et al., 2008). However, precocious weaning may also be performed from one day until just before the age of early weaning (**Table 1**). In general, lambs are transferred to pastures or corrals away from their mothers, breaking the contact between them. This early separation is required to increase the amount of milk available for different milk products, mainly cheese. However, this system is often associated with poor performance of the lambs (Napolitano et al., 1995), in addition to the need to use artificial suckling and milk substitutes, which increase digestive problems such as diarrhoea (Wang et al., 2019). Considering the susceptibility to disease in lambs at this age, feeds given to lambs must be carefully formulated, equipment must be disinfected, and the rhythm of milk ingestion should be regulated to ensure their health. Overall, artificial rearing systems can be expensive due to milk replacer and labour costs during the milk-feeding period (Bimczok et al., 2005). In addition to health problems, lambs could suffer chronic stress when subjected to artificial rearing. Lambs prevented from suckling their mothers and receiving milk from a bucket show more distress bleats and spend less time near their companions than dam-suckled lambs (Napolitano et al., 2003). Furthermore, artificial rearing in male lambs generates long-term consequences on the response to stressors and sexual behaviour (Damián et al., 2017, 2018a,b). Separating the young from the mothers at such an early age limits meaningful lambs' learning about the physical and social environment (Thórhallsdóttir et al., 1990; Orihuela et al., 2019).

TABLE 1 | Type of weaning more commonly applied according to the age of lambs and sheep production systems.

Type of weaning	Weaning age	Sheep production system
Precocious	From 24 h until 29 days	Dairy sheep reared intensively or semi-extensively
Early	From 30 until 60 days	Meat and wool sheep reared intensively or semi-intensively
Traditional	From 61 until 90 days	Meat and wool sheep reared intensively, semi-intensively or extensively
Late	After 90 days	Meat and wool sheep reared extensively

Early, Traditional and Late Weaning

Meat and wool sheep farms commonly use early, traditional or late weaning, which is done after the lactation peak, coinciding with enough solid food consumption of lambs (Freitas-de-Melo and Ungerfeld, 2016a). Early weaning is often performed when lambs are 30 to 60 d old, traditional weaning between 61 to 90 d of age, and late weaning when they are older than 90 d, but before natural weaning (**Table 1**). These types of weaning can be done abruptly, progressively, or in steps (see section WEANING METHODS). In extensive sheep production systems based on natural pastures, weaning is frequently done late, as otherwise, the nutritional content of pastures might limit the growth of recently weaned lambs. In contrast, in semi-extensive systems weaning is done at an earlier age, especially when food availability after weaning does not limit lambs' growth. Even though lambs begin to consume solid food between 15 and 30 d of age (Lane et al., 2000), Campbell et al. (2017) demonstrated that delaying the weaning age of lambs to at least 60 d can be beneficial from an animal health standpoint, requiring less harvested grain in the feedlot to reach an appropriate market endpoint in pasture-based systems. A negative aspect of early weaning is the fact that it limits growth rate, as the animals require longer times to adapt their digestive system to solid food consumption due to the physiological delay in ruminal development or lack of familiarity with solid food (Lynch et al., 1992). Thus, weight gain can drop in the weeks following weaning at 45 d due to low pasture consumption and concentrate.

STRESS RESPONSE AT WEANING

Weaning involves several stressors for ewes and lambs, including: (1) emotional response provoked by the end of the mother-lamb bond; (2) the end of suckling/nursing, a pleasant activity that stimulates the delivery of hormones and neurotransmitters that induce relaxed and calm status; (3) in the case of precocious weaned lambs, milk consumption from artificial teats; (4) artificial weaning also involves an acute change in the lambs' nutrition, as milk is completely replaced by solid

food (Weary et al., 2008) or by milk replacer, which has a lower nutritional value (Sevi et al., 2009); (5) the loss of the mother, the first social bond and primary source of social learning, including social organisation and behavioural patterns of the species (Mepham and Kuhn, 1994); (6) changes in the physical and social environment, since lambs are commonly housed in a new environment, often with unknown conspecifics (Weary et al., 2008; Sevi et al., 2009; Freitas-de-Melo and Ungerfeld, 2016a); in some sheep systems, weaning is followed by transportation, another significant stressor (Tadich et al., 2009); (7) the end of nursing might induce inflammation of the ewes' udder, as reported in beef cows (Ungerfeld et al., 2015). Altogether, these stressors trigger strong responses in both members of the dyad, which generally negatively affect their physiological and health status, constituting a significant welfare issue.

Behavioural Responses in Ewes and Lambs

Immediately after ewes and their lambs notice the separation, both increase the frequency of behaviours displayed to search each other and decrease the frequency of feeding and resting behaviours (Freitas-de-Melo et al., 2013, 2017). Ewes and lambs increase the number of vocalisations, and the frequencies of standing up and walking, as well as pacing (when animals walk at a distance of 1 to 2 m parallel to the fence, modifying the direction of back and forth repeatedly) (Alexander, 1977; Freitas-de-Melo et al., 2013, 2019) (**Figure 1**). The behavioural changes more closely related to the stress response at weaning are the increase in the number of vocalisations and pacing (Freitas-de-Melo and Ungerfeld, 2016a). Usually, these behavioural changes are rapidly triggered, peaking in few hours and lasting not more than 2 to 3 d (Freitas-de-Melo et al., 2013, 2017, 2019). These behaviours are energetically demanding (Weary and Fraser, 1995), but they are also ineffective, as the lamb cannot re-join its mother; thus, the increase of the costlier behaviours is not maintained for a long time. In addition, after weaning, the ewe and the lamb decrease their time lying down, grazing and ruminating (**Figures 2**, **3**). Lambs can also redirect their behaviour, even aiming to suck from other lambs (Stephens and Baldwin, 1971). The loss of the mother as a result of weaning is the main factor involved in lambs' behavioural changes. Damián et al. (2013) observed that lambs vocalised and paced after being separated from their mother; however, lambs that are reared with sheep milk provided in artificial teats from 24 to 36 h after birth did not display these behaviours after the provision of milk ended at 75 d of age. This demonstrates that the mother's loss, but not the loss of access to milk and suckling, induces these behaviours. Male lambs increase the aggressiveness after weaning, a behaviour that is maintained along several days despite weaning is performed at 45 or 75 days of age (Çakmakçi et al., 2021). The behavioural circadian rhythms are also disrupted for several days, including many peaks of activity outside the expected moment (Wyse et al., 2018).

Physiological and Immunological Responses in Ewes

Surprisingly, and although it might have important productive and welfare implications, there is scarce, and in some cases contradictory information, on the physiological responses of ewes at weaning. Orgeur et al. (1998, 1999) did not observe changes in cortisol secretion after weaning, but Cockram et al. (1993) reported an initial increase in cortisol concentrations with quick habituation. However, Pérez-León et al. (2006) observed a significant increase in ewes' cortisol concentration after weaning, so future studies should investigate the determinants of this variation. The inconsistency between the results of these studies could be due to the difference in the time taken for ewes to perceive the absence of their lambs. In effect, the response is not triggered simultaneously in all ewes, as many of them respond to separation until the udder is full and produces pain. Thus, the moment in which ewes' stress response begins could be related not only to the separation but also by the udder modifications induced by weaning. Therefore, the udder development, and milk yield produced, among other factors, could explain why some ewes respond with increases in cortisol concentration and others do not. It is also possible that the status on the natural break of the bond differs in different studies, thus, measuring responses in moments different to those in which the responses could be expected.

It has also been reported that the haematocrit remains elevated 5 d after weaning, indicating that weaning is a long-term stressful situation (Freitas-de-Melo and Ungerfeld, 2020). Weaning also induces a decrease in total protein concentration (Freitas-de-Melo et al., 2013, 2017; Freitas-de-Melo and Ungerfeld, 2020) and changes in albumin concentration (Freitas-de-Melo et al., 2013, 2017; Freitas-de-Melo and Ungerfeld, 2020). However, Cockram et al. (1993) did not observe changes in α-endorphin or prolactin concentrations after artificial weaning in Greyface ewes. Weaning also generates an increase in the leukocyte number (Orgeur et al., 1998), and more specifically, in neutrophil concentration, associated with a decrease in the proportion of CD2 lymphocytes and T19 lymphocytes (Cockram et al., 1993). Although there is a general reduction in globulin concentration (Freitas-de-Melo et al., 2013, 2017), the antibody response to a challenge with ovalbumin or the skin response to dinitroflurobenzene is not affected (Cockram et al., 1993). Therefore, the study of the immunological ability of recently weaned ewes still requires deeper understanding.

Physiological and Immunological Responses and Their Consequences on Lambs' Health and Growth

Weaning triggers a typical physiological stress response in lambs, including an acute increase in serum cortisol concentration (Mears and Brown, 1997). The combination of stressors at weaning challenges the general homeostasis of the lambs, inducing high energy costs until their physiological status stabilises to the new situation. Weaning also triggers changes in the respiratory and pulse rates and rectal temperature (Mohapatra et al., 2020). According to Fazio et al. (2015) triiodothyronine concentration decreases, but thyroxine concentration increases 2 wk after weaning. However, some authors did not observe the same response (Mears and Brown, 1997), suggesting that these changes

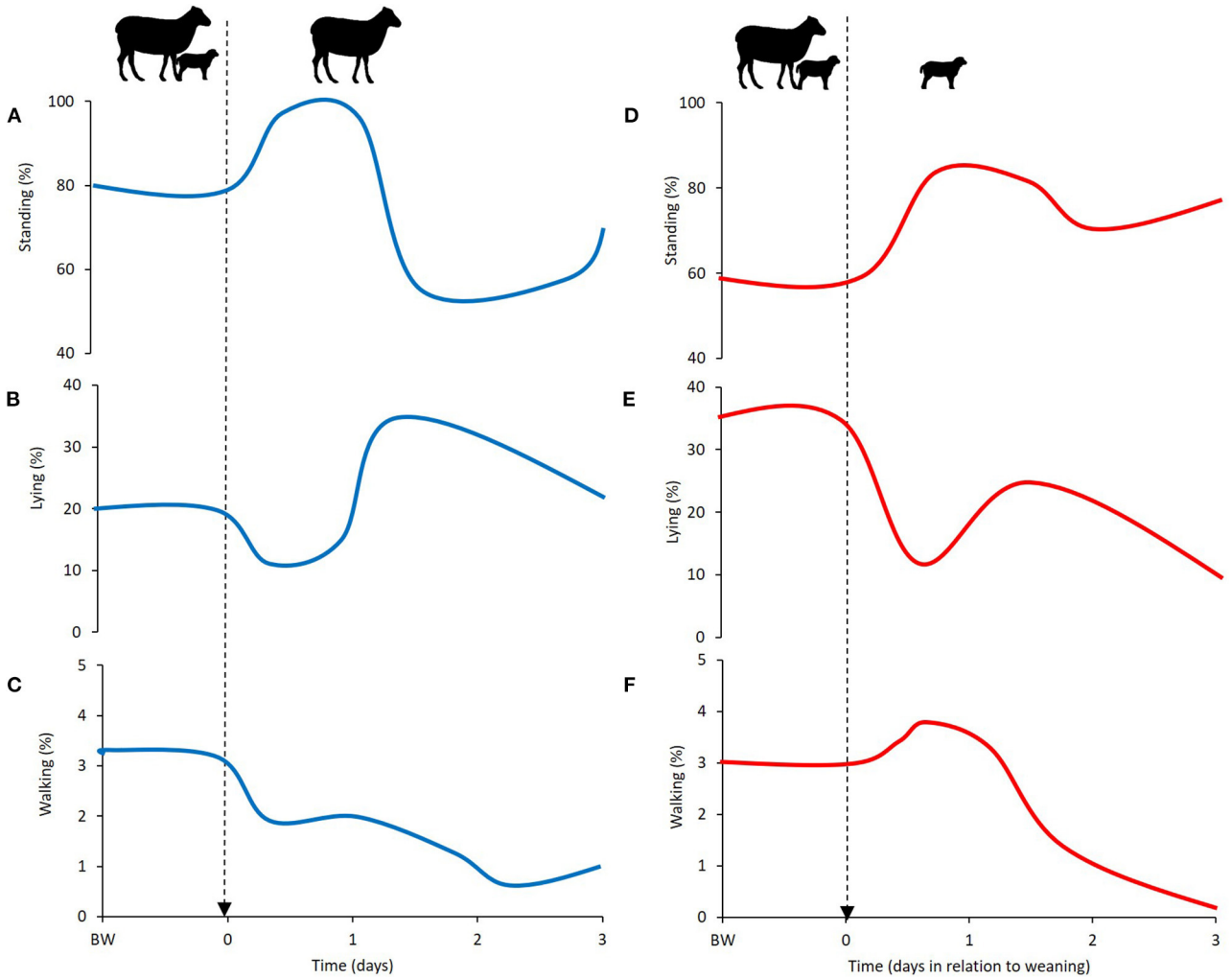

FIGURE 1 | Behavioural changes in ewes (left panel) and lambs (right panel) triggered by weaning (dotted arrow). The upper graphs show the time spent standing **(A, D)**, the medium panel the time spent lying **(B,E)**, and the more below one, the time walking **(C,F)**. The models are based in the data of Freitas-de-Melo and Ungerfeld (2020).

could be related to the specific environmental conditions in which lambs are maintained after weaning. Çakmakçi et al. (2021) found a decrease in triglyceride concentration during 7 d after weaning. Weaning also triggers a decrease in total protein and albumin concentration (Freitas-de-Melo et al., 2017), and the haematocrit remains elevated at least 5 d after weaning (Freitas-de-Melo and Ungerfeld, 2020).

The immune capacity is also affected after weaning (Napolitano et al., 1995), which together with the decrease in IGF-1 (McCoard et al., 2020) and the increase in cortisol concentrations, probably increase lambs' susceptibility to different diseases. The adverse effects of weaning on the lamb's immune system remain for at least 3 wk and are associated with inflammatory responses, including an increase in pro-inflammatory cytokines (Zhang et al., 2018). The number of antibodies synthesised after an antigenic challenge is lower in recently weaned lambs (Napolitano et al., 1995). In accordance, weaned lambs are more susceptible to experimental infections with *Haemonchus contortus* and *Trichostrongylus colubriformis* than unweaned lambs (Watson and Gill, 1991). This is consistent with the increase in the excretion of coccidia oocysts in recently weaned lambs (Orgeur et al., 1998, 1999). Although in other studies there was no apparent increase in parasite load, the associated blood loss is greater in early-weaned lambs (Schichowski et al., 2010). An increase in the incidence of diarrhoea is observed in precocious and early-weaned lambs (Wang et al., 2019). Moreover, lambs naturally infected with a greater number of gastrointestinal nematodes decrease their time lying (Högberg et al., 2021), amplifying the effect of weaning on resting time.

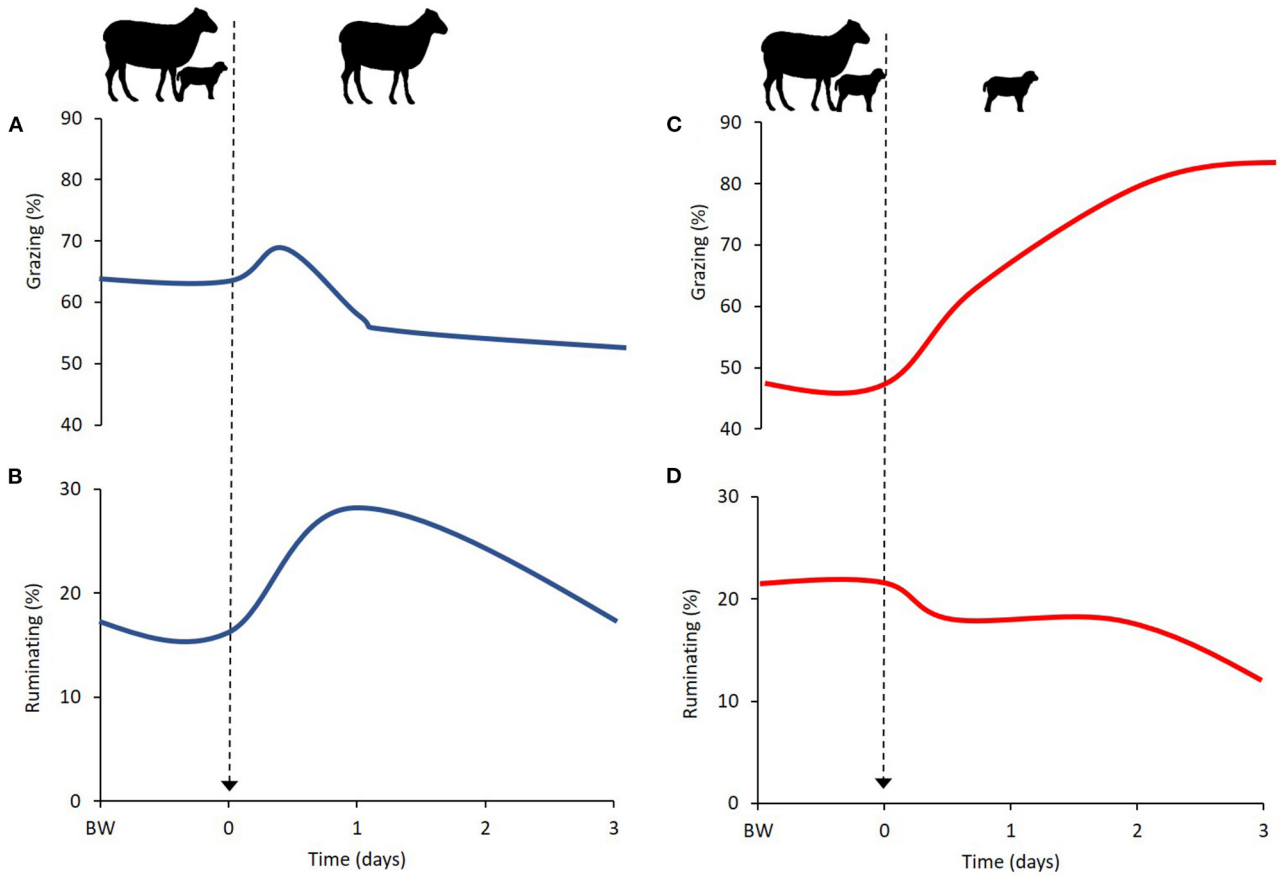

FIGURE 2 | Behavioural changes in ewes (left panel) and lambs (right panel) triggered by weaning (dotted arrow). The upper graphs show the time spent grazing **(A,C)** and the one below, the time ruminating **(B,D)**. The models are based in the data of Freitas-de-Melo and Ungerfeld (2020).

As mentioned earlier, weaning also negatively affects the growth of the lambs, inducing a decrease in body weight shortly after separation (Mohapatra et al., 2020). This effect might be even greater in lambs weaned before rumen development is complete. This delay in growth may be a consequence of a bad adaptation of the digestive system to the acute feeding changes, the reduced immune status, the increased susceptibility to gastrointestinal parasites, and the energy dispended in the display of costly behavioural responses.

In summary, behavioural and physiological responses at weaning negatively affect sheep production and welfare, so it is essential to deeply understand the factors involved in weaning responses and develop alternatives to control them.

FACTORS INVOLVED IN THE WEANING STRESS RESPONSE

Multiple factors influence the responses at weaning and how sheep cope with this stress. Although many more factors need to be studied, at least age, sex, type of birth, genetic, mother's nutrition during gestation and lambing season are directly related to this response (**Table 2**).

Single-born Merino lambs weaned at 6 wk are more anxious and vocalise more times than lambs weaned at 13 wk of age (Schichowski et al., 2010). Similarly, compared to lambs weaned at 100 d of age, those weaned at 50 d secrete more cortisol, which induces an increase in glucose concentration (Sowinska et al., 2001). The response follows a similar pattern in other physiological traits; e.g., concentrations of lactate dehydrogenase and creatine kinase are higher in lambs weaned at 45 than at 75 d of age (Çakmakçi et al., 2021), and total protein concentration is lower in younger than older weaned lambs (Çakmakçi et al., 2021). The earlier the lambs are weaned, the lower their subsequent weight gains are, even when grazing pasture and/or receiving food supplementation (Cañeque et al., 2001; Godfrey and Weis, 2016). This effect might be due to a lack of adaptation to solid food as a unique food resource. The incidence of diarrhoea is also greater in lambs weaned at 21 than at 35 d of age, affecting the daily weight gain in the younger lambs (Wang et al., 2019). Age at weaning modifies the lambs' rumen development (Belanche Gracia et al., 2019) and ileal microbiota (Li et al., 2018), even affecting the amount of expression of

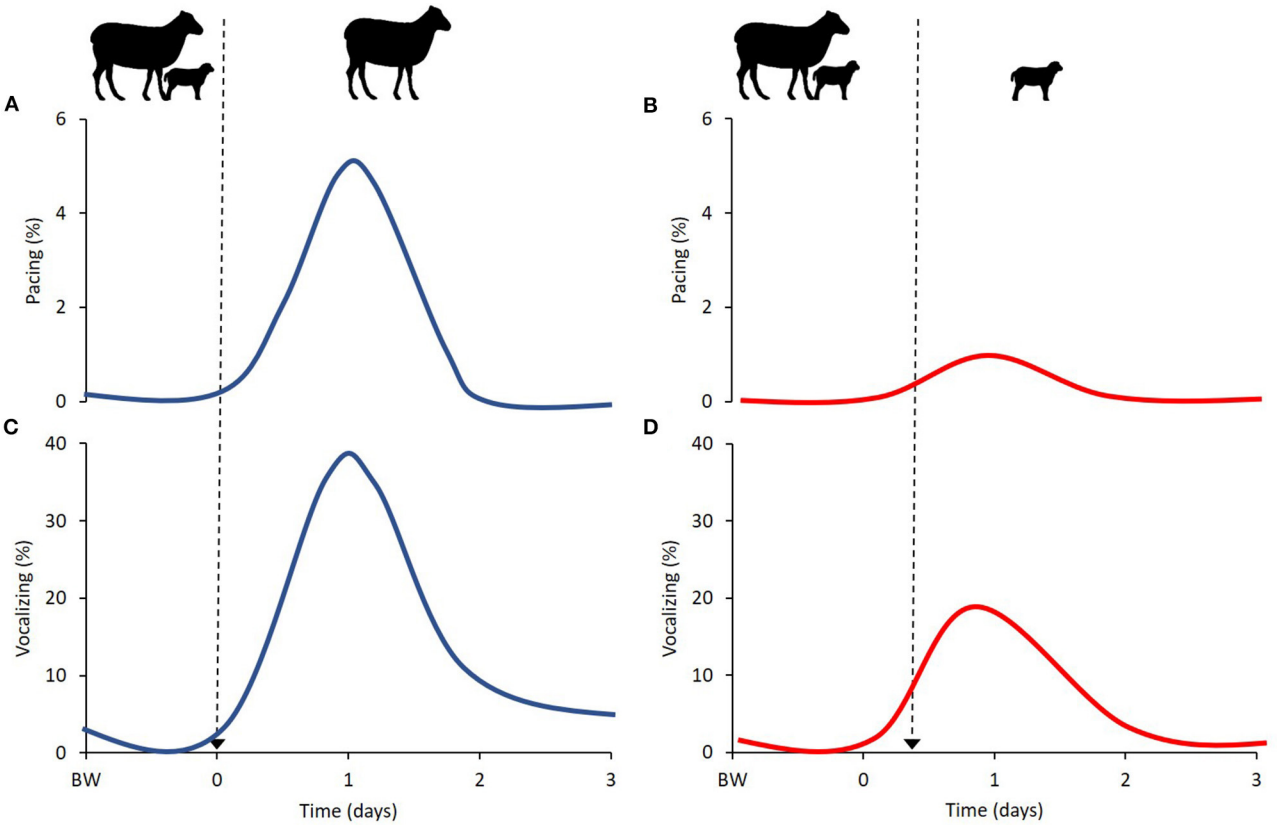

FIGURE 3 | Behavioural changes in ewes (left panel) and lambs (right panel) triggered by weaning (dotted arrow). The upper graphs show the time spent pacing **(A,C)** and the one below, the emission of vocalisations **(B,D)**. The models are based in the data of Freitas-de-Melo and Ungerfeld (2020).

genes regulating the intestinal barrier function (Li et al., 2018). Clean wool grows more in early-weaned than in late-weaned ewes (Smeaton et al., 1983). Although the age at artificial weaning appears to be more challenging for the lambs than for the ewes, to the best of our knowledge, there is no published article studying the stress response of ewes according to lambs' weaning age. Mastitis is also a concern after weaning in wool and meat sheep, and it is more commonly observed in early weaning systems as lambs are separated from their mothers when milk yield is still high (Animal Welfare Approved certification program, 2018). After early weaning, some farmers reduce the amount of food and water offered to lactating ewes to decrease milk production and avoid mastitis, which also constitutes a welfare concern.

The response to abrupt weaning also differs in both ewes and lambs according to the sex of the lamb. After abrupt weaning at 90 d of age, single castrated male lambs grazed, ruminate and gain more weight than female lambs. Some responses are reported only in female lambs, such as an increase in albumin concentration (Freitas-de-Melo and Ungerfeld, 2020). On the other hand, weaning performed around 120 d of age affects more the growth of male than female bighorn sheep until puberty (Festa-Bianchet et al., 1994). In the conditions of that study, food availability changed throughout the year, including periods of food restriction. Therefore, the periods of food restriction and the greater growth rate and energy demand of males could explain this sex difference. The postweaning mortality rate is greater in male than female lambs when weaning is performed either at 90 or 150 d of age (Turkson and Sualisu, 2005; Mandal et al., 2007). On the weaning day, ewes that reared male lambs pace more than those that reared female lambs (Freitas-de-Melo and Ungerfeld, 2020), probably because mothers of male lambs invest more energy during gestation and lactation than mothers of females (Clutton-Brock et al., 1981; Berube et al., 1994). Male and female lambs have different productive goals after weaning, as in general male lambs are finished for slaughter, and female lambs replace breeding animals (Kenyon and Cranston, 2017). Therefore, considering that ewes cope with weaning stress differently according to the sex of their lambs, farmers might apply different strategies to improve animal welfare after weaning considering the sex of the offspring.

Ewes and their twin or single lambs respond differently to early weaning, probably because ewes and their single lambs have a stronger bond than ewes and their twins (O'Connor et al., 1992; Dwyer and Lawrence, 1998). In fact, after early weaning (at 1.5 mo old), single lambs pace more than twin lambs and reduce their grazing and lying down frequencies, whilst twin lambs do not change it (Freitas-de-Melo et al., 2021b). Similarly, ewes rearing

TABLE 2 | Factors involved in the physiological, behavioural and immunological responses at artificial weaning in ewes and lambs.

Type of sheep	Physiological response	Behavioural response	Other responses	References
Younger lambs	↑cortisol ↑glucose lactate ↑dehydrogenase ↑creatine kinase ↓total protein	↑vocalising ↑anxiety	↓weight gain ↑diarrhoea rate	Cañeque et al. (2001) Sowinska et al. (2001) Schichowski et al. (2010) Çakmakçi et al. (2021) Godfrey and Weis (2016) Wang et al. (2019)
Mothers rearing female Lambs	↑total protein ↑globulin	↓pacing ↑walking	–	Freitas-de-Melo and Ungerfeld (2020)
Female lambs	↑albumin	↓grazing ↓ruminating	↓weight gain ↑survival rate	Turkson and Sualisu (2005) Mandal et al. (2007) Freitas-de-Melo and Ungerfeld (2020)
Mothers with twin lambs	↑albumin	↓vocalising ↓pacing	–	Freitas-de-Melo et al. (2021b)
Single lambs vs twins	↑*albumin	↑pacing ↓grazing ↓lying down	↑bodyweight	Freitas-de-Melo et al. (2021b)
Mothers that received a low pasture allowance during gestation	–	↓grazing	↑bodyweight	Freitas-de-Melo et al. (2017)
Lambs from mothers that received a low pasture allowance during gestation	↓variation in albumin	↓vocalising ↓pacing	–	Freitas-de-Melo et al. (2017)
Texel × Corriedale vs. Corriedale × Correidale lambs	–	↑ruminating ↓pacing	↑bodyweight	Freitas-de-Melo et al. (2019)
Merino vs. Rhoenschaf lambs	–	↓vocalising	↑bodyweight	Schichowski et al. (2008)
Spring-born vs. autumn-born lambs	–	↓walking ↓grazing ↑standing up	↑bodyweight	Menant et al. (2022)

Tended to decrease; –no difference was reported, or this response was not studied. Up arrow or down arrow mean increase or decrease in the recorded variables.

singles pace and vocalise more than the mothers of twins (Freitas-de-Melo et al., 2021b). In general, more studies are needed in prolific breeds to determine more deeply the effects of litter size on the artificial weaning responses in sheep.

Biotype also modulates the response of lambs to artificial weaning, as it can modify growth rate and thus, the development of the natural weaning process. Therefore, at the same age, lambs from different breeds or biotypes advance differently in the stages of their natural weaning, responding differently to separation from the mother. For example, Merino lambs have a greater body development and vocalise less than Rhoenschaf lambs after being weaned at 8 or 16 wk of age (Schichowski et al., 2008). When weaning is performed at 143 d of age, Texel × Corriedale crossbreed lambs grow faster and ruminate more frequently than purebred Corriedale lambs, and the former do not pace, while purebred Corriedale lambs do (Freitas-de-Melo et al., 2019). These crossbreed lambs developed different feeding strategies during the pre-weaning period, depending less on their mother, influencing their behavioural response at weaning (Freitas-de-Melo et al., 2019). Although different temperaments or copying styles could also explain the effect of biotypes on the lambs' stress response at weaning, this factor does not influence the response of lambs to artificial weaning at different ages (Çakmakçi et al., 2021).

Ewes' nutrition during gestation also affects the lambs' weaning response. Food restriction during gestation negatively affects the lambs' birth weight, the mother-lamb bond at lambing (Dwyer et al., 2003; Freitas-de-Melo et al., 2015a), and reduces the number of rejecting behaviours towards alien lambs by selective mothers (Freitas-de-Melo et al., 2018b). Furthermore, the lambs from undernourished ewes advance their natural weaning process since they present a greater grazing frequency before weaning, probably to compensate for the access to fewer amounts of milk (Freitas-de-Melo et al., 2017). Lambs from feed-restricted ewes during gestation vocalise and pace less, and the variation of albumin concentration before and after weaning is lower than that of lambs from well-fed ewes (Freitas-de-Melo et al., 2017).

Lambing season also influences the behavioural response of lambs at weaning. In out-of-season lambing systems, ewes get pregnant in spring, and consequently, lambing and lactation take place during autumn and winter, respectively. In these conditions, lambs have a lower birth weight, poorer vitality, and greater mortality rate (Chniter et al., 2011; Dhaoui et al., 2020) than spring-born lambs. Autumn-born lambs display different

strategies to cope with weaning compared to spring-born lambs, as the former walk more frequently but were observed less often standing up than the latter (Menant et al., 2022). While spring-born lambs decrease their grazing frequency at weaning, autumn-born lambs increase it (Menant et al., 2022). In addition, mothers of autumn-born lambs produce less milk, with less fat, lactose, and protein content than mothers of spring-born lambs (Menant et al., 2022). Consequently, spring-born lambs are heavier before and after weaning than autumn-born lambs (Menant et al., 2022).

Overall, it is necessary to consider several animal and environmental factors that modify the strength of the ewe-lamb bond, and thus, the response to separation. Although we presented some of these factors in this section, there are probably several more that can affect this response but usually are not considered. Therefore, there is room to study the conditioning factors that might modify the ewe and lamb responses to separation.

WEANING METHODS

The most frequently used method for artificial weaning is abrupt separation of mothers and lambs (Freitas-de-Melo and Ungerfeld, 2016a). However, there are some studies aiming to handle the weaning promoting animal welfare. These alternative weaning methods are progressive weaning, two-steps weaning, and mixed regimes (**Table 3**).

Abrupt Weaning

Regardless of lamb's age at weaning, the most frequently used weaning method is abrupt separation (Freitas-de-Melo and Ungerfeld, 2016a), perhaps because sheep farmers can easily apply it. This separation method consists of the sudden and complete separation of the ewe and her lamb(s), avoiding visual, auditory, or olfactory contact after weaning (Freitas-de-Melo et al., 2013). As mentioned above, abrupt weaning generates behavioural, physiological, and immunological responses with consequences on lambs' health and growth. At abrupt weaning, ewes and lambs are very anxious and vocalise frequently, but most of the signs of disturbance are no longer noticeable 2–3 d later (Orgeur et al., 1998; Pérez-León et al., 2006). Abrupt weaning also can increase lambs' susceptibility to diseases and decrease weight gain (Freitas-de-Melo and Ungerfeld, 2016a). Some procedures might be used to reduce the stress generated by abrupt weaning, including the induction of oestrus in ewes on the day of separation (Pérez-León et al., 2006), the presence of conspecifics or humans (Barton, 1983a,b; Carbajal and Orihuela, 2001), stroking (Pascual-Alonso et al., 2015) or progesterone treatments (Freitas-de-Melo et al., 2013; Freitas-de-Melo and Ungerfeld, 2016b). Pre and postweaning handling procedures are described in detail in section STRATEGIES TO IMPROVE SHEEP WELFARE AT WEANING.

Progressive Weaning

During progressive weaning, the time of ewe-lamb separation gradually increases until definitive separation (Freitas-de-Melo and Ungerfeld, 2016a). Farmers can perform progressive weaning, separating ewes and lambs for 4 to 6 h/d during the first wk, increasing 4 to 6 h each wk, until the definitive separation. This method is based on some early studies (Owen et al., 1969a,b) that concluded that the transition from the initial to substantial consumption of solid food is strongly influenced by milk intake. Restriction of milk intake advances the consumption of greater amounts of solid food. Besides that, lambs and ewes habituate to the gradual separation process. Another practical advantage of this method is that restricted suckling advances the resumption of postpartum ovarian activity and reduces the anoestrous period (Morales-Terán et al., 2004, 2011). Furthermore, restricted suckling in the ewes stimulated by close contact with rams decreases the stress responses at weaning (Orihuela et al., 2016) and favours the reproductive response of postpartum ewes (Ungerfeld and Sánchez-Dávila, 2012). However, the easiness of this practise depends on the productive systems, given that in extensive grazing systems would be impractical to move all the flock every day to separate ewes and lambs, in addition to adverse consequences on their body weight and on pasture maintenance.

Although this type of weaning demands more farmers labour force, it decreases the stress response at definitive separation. Ewes tend to present a higher cortisol concentration, and the percentages of mothers feeding are lower after progressive weaning from 25 to 90 d than in the abrupt weaning method (Orgeur et al., 1998). Furthermore, after definitive separation, the percentages of mothers and lambs resting are higher, and ewes and lambs vocalise less after progressive than abrupt weaning (Orgeur et al., 1998). Cortisol concentration is lower in lambs, and body weight is greater after progressive weaning from 39 to 45 d than abrupt weaning at 45 d (Henrique et al., 2017). However, the proportion of lambs with the highest number of oocysts per gramme of faeces is higher in progressive than abrupt weaning (Orgeur et al., 1998). The response to progressive weaning likely differs according to the age at which it begins, as progressive weaning from 5 to 10 d of age induces behavioural, endocrine, and immune disturbances in lambs (Sevi et al., 2003).

Galeana et al. (2007) found that lambs that maintain a greater distance from their mothers during lactation are less stressed at weaning. Progressive weaning also generates long-term consequences on the perception of stressors in lambs. Lambs weaned progressively from 5 to 10 d of age (Sevi et al., 2003) or 30 to 60 d of age (Freitas-de-Melo et al., 2021c) displayed different reactions to social stress later in life compared to lambs weaned abruptly.

Two-Steps Weaning

Two-steps weaning consists of limiting the access to the mothers' udder before definitive ewe-lamb separation, either continuously or temporarily. This can be accomplished by removing lambs from their mothers' pen but maintaining their contact through wire-fence, covering the udder, or using nose flaps to avoid suckling for a limited period before ewe-lamb separation.

Two-steps weaning can be easily applied by separating ewes and lambs through a wire fence. In this case, ewes and lambs can communicate with each other, and thus visual, auditory, and olfactory contact is maintained before definitive ewe-lamb separation. Ewes can see, smell, and hear their lambs, separated only by a wire fence, but cannot make full physical contact

TABLE 3 | Weaning method and physiological, behavioural and immunological responses in lambs and ewes and their consequences in health and growth of lambs after ewe-lamb separation.

Weaning method	Ewe/lamb	Physiological response	Behavioural response	Immunological response	Other responses	References
Abrupt	Ewe	↑cortisol ↓total protein ↓albumin or ↑albumin ↑hematocrit	↑vocalisation ↑pacing and walking ↓rumination ↓grazing ↓lying down ↑ standing up	↓globulin ↑leukocyte number ↑neutrophil concentration ↓proportion of CD2 and T19 lymphocytes	–	Cockram et al. (1993) Orgeur et al. (1998) Pérez-León et al. (2006) Freitas-de-Melo et al. (2013, 2017) Freitas-de-Melo and Ungerfeld (2020)
Abrupt	Lamb	↑cortisol ↓total protein ↓albumin ↑hematocrit ↓respiratory and pulse rates ↓rectal temperature ↓T3 and ↑T4 concentration ↓Triglyceride concentration	↑vocalisation ↑pacing and walking ↓rumination ↓grazing ↓lying down ↑standing up	↑pro-inflammatory cytokines ↓number of antibodies after an antigenic challenge	↓body weight gain ↑excretion of coccidia oocysts ↑diarrhoea	Napolitano et al. (1995) Mears and Brown (1997) Fazio et al. (2015) Mohapatra et al. (2020) Çakmakçi et al. (2021) Freitas-de-Melo et al. (2017) Zhang et al. (2018) Freitas-de-Melo and Ungerfeld (2020)
Progressive	Ewe	↑*cortisol	↓high pitched bleats ↓low piched bleats ↑resting ↓feeding	↑leucocyte count	–	Orgeur et al. (1998)
Progressive	Lamb	↓cortisol	↓high pitched bleats ↓low piched bleats ↑resting ↑feeding	–	↑body weight ↑excretion of coccidia oocysts	Henrique et al. (2017) Orgeur et al. (1998)
Wire fence separating vs. abrupt	Ewe	–	↑high piched bleats ↑low piched bleats ↓feeding	–		Orgeur et al. (1999)
Wire fence separating vs. abrupt	Lamb	↓cortisol	↑high piched bleats feeding	–	Avoid the ↑excretion of coccidia oocysts	Orgeur et al. (1999)
Two-step (by cover the udder)	Lamb	–	↓vocalisation ↓agitation score	–	–	Schichowski et al. (2008) Norouzian (2015)

Tended to increase; –no difference was reported, or this response was not studied.
T3, triiodothyronine concentration.
T4, thyroxine concentration.

(or suckle) (Orihuela et al., 2004). Lambs that had a close spatial association with their mothers before weaning are more likely to approach a wire fence that separates them from their mother during weaning at 64 d of age (Galeana et al., 2007). A similar approach in cattle indicated that during the first days after separation, calves remained for long periods close to the fence, aiming to get together with their mother, probably generating frustration as the goal cannot be reached (Enríquez et al., 2010). Ewes and lambs weaned by fence-line separation vocalise more and eat less than those weaned abruptly (Orgeur et al., 1999). However, females weaned from fence-line separation weigh more after weaning and present a smaller coccidial oocyst output than those weaned abruptly (Orgeur et al., 1999).

After covering the mothers' udder for 1 wk before definitive separation, lambs display fewer behaviours indicative of agitation (Schichowski et al., 2008; Norouzian, 2015) and vocalise less than those weaned abruptly (Schichowski et al., 2008).

Another strategy in two-step weaning is to prevent suckling some time before weaning by placing nose flaps in lambs but maintaining them in free contact with their dams. Nose flaps are more used in beef cattle, so there are more studies regarding the use of nose flaps in two-steps weaning in calves and cows (Haley et al., 2005; Enríquez et al., 2011; Ungerfeld et al., 2015; Alvez et al., 2016). Currently, there are some commercial nose flaps available to use in lambs (**Figure 4**), but to the best of our knowledge, there are no published articles that studied the effect of these products on the stress response or lambs' weight gains using this procedure. We observed that at least some commercial nose flaps should be strongly adjusted to prevent falling off, producing important injuries in the nasal septum of different breeds of lambs. However, that can depend on the product design,

FIGURE 4 | Lamb with a nose-flap **(A,B)**. Pictures provided by Daniela Casuriaga.

as some farmers in Uruguay are using a commercial nose flap with apparently good results.

It remains to be determined the amount of early stress response induced by the initial limitation of the access to the udder, either by nose flaps or by covering it, or by separation with a wire-fence. This lack of knowledge limits recommending these strategies due to the possibility that the stress response is redistributed over time but not necessarily decreased. In fact, if this is the case, it would be difficult to know what is really better from an animal perspective. Another limitation of the scarce knowledge is that in these studies, it was studied the response of the lambs, but not that of the ewes. The permanent contact with her offspring maintains the stimulus for milk production, but the udder would remain full as the lamb could not suckle, producing pain and probably mastitis. Therefore, more studies, amplifying the approaches and the perspectives should be performed before practical recommendations can be given.

Mixed Regimes

Mixed regimes are applied in some dairy sheep farms during the first month postpartum, where lambs continue accessing their mother for short periods of time, allowing a restricted period for suckling, alternated with periods of separation before milking (Sevi et al., 2009). During these periods of contact, lambs suckle *ad libitum* and interact freely with their mother. In this system, the ewes and the lambs are habituated to the separation, also promoting the early consumption of solid food by the lamb. This regime varies in different farms and regions, but it may begin as early as 1 d after birth, separating ewes and lambs at evening, milking the ewes once daily in the morning, and allowing full ewe-lamb contact during the day (McKusick et al., 2001). As the greater milk production occurs in the first month, the amount of milk is enough for the normal development of the lambs and to produce commercial milk (Sevi et al., 2009).

Interestingly, there was no difference in total commercial milk produced by ewes abruptly weaned from their lambs 24 h postpartum and ewes maintained in the mixed regime (McKusick et al., 2001), probably because the contact with the lamb stimulates a greater milk yield. In a mixed regime, the decrease in milk production with the advance of lactation might be compensated with the administration of lambs' commercial starter concentrate, which can begin when lambs are 2 wk old or even earlier. The concentrate can be initially offered to the lambs through a creep feeding system while they are still with their mother, so they can learn to consume solid food from a trough. After lambs are habituated to consume solid foods, solid supplements can be offered during periods of separation. In summary, this system does not affect milk production nor the growth of the lambs (McKusick et al., 2001) and allows the lambs' gradual adaptation to consume solid food while they are separated from their mother.

STRATEGIES TO IMPROVE SHEEP WELFARE AT WEANING

As mentioned earlier, different managements can be applied before and/or after weaning to improve how ewes and lambs cope with weaning. Some can be applied in lambs around weaning to promote an earlier maturation of the digestive system. For instance, the provision of feed supplements before and after weaning can help lambs cope with the nutritional changes at weaning. The use of creep feeding is recommended for stimulating solid food consumption even at an early age, facilitating the transition to the digestion of solid food (Martínez et al., 2015). Moreover, the administration of starter feed in weaned lambs at 10 d of age stimulates the development of ruminal epithelia, accelerating the cell cycle, inhibiting apoptosis in the rumen epithelium, and promoting the uptake of volatile fatty acids (Sun et al., 2018). Furthermore, faecal worm egg counts are lower in supplemented-weaned lambs than unsupplemented lambs (Shaw et al., 1995), probably because supplemented-weaned lambs are less exposed to parasites, improving the health status of lambs. Oral administration of fresh rumen fluid in lambs weaned at 28 d of age improves apparent digestibility of dry matter and neutral detergent fibre and growth rate (Zhong et al., 2014).

There are also some postweaning treatments that facilitate the transition of the lambs to their new status, especially considering the social environment. Lambs are pushed to substitute completely milk for solid food, so being placed together with other conspecifics that are adapted to this might facilitate their learning process. For example, lambs that are placed to graze together with adult ewes after weaning have a greater bite rate and ruminate and graze more than single weaned lambs (Youssef et al., 1995). Although the separation from their mother is stressful, keeping the lambs in a familiar environment with other lambs may allow them to adjust to the separation from their mother more easily than moving them to an unfamiliar pen or putting them with new conspecifics. Besides, postweaning social enrichment also improves lambs' welfare (Pascual-Alonso et al., 2015). The stress response of the lambs diminishes when they remain for 1 h/d after weaning in close contact with a stockperson who handles them gently (stroke and spoke to them) or if they stay in contact with adult ewes. The lambs who receive these treatments grow more during the fattening period, and the contact with humans after being weaned reduces the reaction to human manipulation (Pascual-Alonso et al., 2015).

It is important to consider also the relationship with other lambs that are placed together. For example, twin lambs display more intensive behavioural response (greater frequency of vocalise and pace) and have poorer immunological status at weaning when housed together than separated from their sibling (Casuriaga et al., 2021). Therefore, separating twins after weaning and coupling them with non-siblings is probably better to reduce their stress and improve their health status after early weaning in prolific breeds.

Long-term treatment with progesterone reduces ewes' responses at weaning (Freitas-de-Melo et al., 2013), as progesterone metabolites have an anxiolytic effect (Freitas-de-Melo and Ungerfeld, 2016a). On the day of weaning, ewes that receive an intravaginal progesterone releasing device (CIDR) for the 32 days before artificial weaning, pace and vocalise less than untreated controls. Moreover, 4 days after artificial weaning, globulin concentration is lower in control ewes than in treated ewes, reaching levels of hipo-globulinemia, which could be related to a state of immunodeficiency. Therefore, this treatment can also potentially reduce susceptibility to diseases of ewes after weaning. Although this treatment reduces the stress response at weaning in ewes, it is not practical, as it is too long and expensive, reducing the possibility of practical use. For this reason, an injection of oil-based progesterone to ewes immediately before weaning to simplify the handling of animals and reduce the costs might be a practical alternative. Although this treatment decreases the behavioural response (Freitas-de-Melo et al., 2015b), the reduction of the negative responses is not as great as long-term treatment (Freitas-de-Melo et al., 2013). Another practical management that provided interesting results is the induction of oestrus in ewes simultaneously with weaning, to superimpose another goal to the ewe, reducing the attention to their lambs. In effect, this management reduces the number of vocalisations and the cortisol concentration on the day of weaning (Pérez-León et al., 2006). This treatment can be easily included in accelerated lambing systems to have pregnant ewes during the early postpartum period.

OVERVIEW AND DIRECTIONS

Different ages and methods to wean lambs might be adapted and recommended in different production systems. The effects of weaning on health, production, and welfare are highly variable due to several factors and conditions, as age, sex, type of birth, genetics, mother's nutrition during gestation, lambing season, and physical and social environments after weaning. Although weaning is one of the most stressful events in farm animals for both, mothers and offspring, there are important gaps in the knowledge required to decrease the negative concerns of a widely used handling. In general, it seems that regardless of the system used, practises such as inducing oestrous in ewes, and promoting early consumption of solid food in lambs, and the inclusion of strategies of social facilitation and environmental enrichment in the postweaning allocation of lambs, reduce the stress caused by weaning. In addition, the adaptation of both members of the dyad after their definitive separation can be improved by alternative weaning methods, such as progressive, two-steps weaning, and mixed regimes. It should also be considered that the weaning method has short- and long-term effects, affecting how the animals cope with weaning but also modifying several behaviours of the offspring later in life. This review highlights the need of future studies focusing on the factors that might modify the ewe and lamb responses to separation. Moreover, as social demands require a sustained decrease of practises that might affect animal welfare, the development of new strategies can impact not only in productivity, but also in the future sustainability of the productive systems. Accordingly, it is essential to develop studies that focus on strategies to decrease the adverse effects of weaning on sheep's welfare, and productivity.

AUTHOR CONTRIBUTIONS

All authors listed have made a substantial, direct, and intellectual contribution to the work and approved it for publication.

REFERENCES

Alexander, G. (1977). Role of auditory cues in mutual recognition between ewes and lambs in Merino sheep. *Appl. Anim. Ethol.* 3, 65–81. doi: 10.1016/0304-3762(77)90072-4

Alexander, G., Poindron, P., Le Neindre, P., Stevens, D., Lévy, F., and Bradley, L. (1986). Importance of the first hour post-partum for exclusive maternal bonding in sheep. *Appl. Anim. Behav. Sci.* 16, 295–300. doi: 10.1016/0168-1591(86)90122-X

Alexander, G., and Shillito, E. E. (1985). "Importance of visual clues from various body regions in maternal recognition of the young in Merino sheep (*Ovis aries*) " in *Reproductive and Developmental Behaviour in Sheep*, ed A.F. Fraser (New York, NY: Elsevier), 229–235.

Alvez, P., Quintans, G., Hötzel, M. J., and Ungerfeld, R. (2016). Two-step weaning in beef calves: permanence of nose flaps for 7 or 21 days does not influence the behaviour response. *Anim. Prod. Sci.* 56, 866–870. doi: 10.1071/AN14643

Animal Welfare Approved certification program (2018). *Age of Weaning Lambs*. Available online at: https://agreenerworld.org/wp-content/uploads/2018/04/TAFS-17-Age-of-weaning-lambs-v1-updated.pdf [accessed October 21, 2021].

Arnold, G. W., Wallace, S. R., and Maller, R. A. (1979). Some factors envolved in natural weaning processes in sheep. *Appl. Anim. Ethol.* 5, 43–50. doi: 10.1016/0304-3762(79)90006-3

Barton, M. A. (1983a). Behaviour of group-reared calves on acid milk replacer. *Appl. Anim. Ethol.* 11, 77. doi: 10.1016/0304-3762(83)90088-3

Barton, M. A. (1983b). The effects of management and behavioural factors on intake of acidified milk and concentrates by group-reared calves. *Anim. Prod.* 36, 512.

Belanche Gracia, A., Yáñez-Ruiz, D. R., Detheridge, A., Griffith, G., Kingston-Smith, A., and Newbold, C. (2019). Maternal vs. artificial rearing shapes the rumen microbiome having minor long-term physiological implications. *Environ. Microbiol.* 21, 4360–4377. doi: 10.1111/1462-2920.14801

Berube, H. C., Festa-Bianchet, M., and Jorgenson, J. T. (1994). Reproductive costs of sons and daughters in Rocky Mountain bighorn sheep. *Behav. Ecol.* 7, 60–68. doi: 10.1093/beheco/7.1.60

Bimczok, D., Röhl, F. W., and Ganter, M. (2005). Evaluation of lamb performance and costs in motherless rearing of German Grey Heath sheep under field conditions using automatic feeding systems. *Small Rumin. Res.* 60, 255. doi: 10.1016/j.smallrumres.2004.12.008

Çakmakçi, C., Karaca, S., and María, G. A. (2021). Does coping style affect behavioral responses and growth performance of lambs weaned at different ages?. *J. Vet. Behav.* 42, 64–74. doi: 10.1016/j.jveb.2020.10.009

Campbell, B. J., Pullin, A. N., Pairis-Garcia, M. D., McCutcheon, J. S., Lowe, G. D., Campler, M. R., et al. (2017). The effects of alternative weaning strategies on lamb health and performance. *Small Ruminant. Res.* 156, 57. doi: 10.1016/j.smallrumres.2017.09.006

Cañeque, V., Velasco, S., Díaz, M., Pérez, C., Huidobro, F., Lauzurica, S., et al. (2001). Effect of weaning age and slaughter weight on carcass and meat quality of Talaverana breed lambs raised at pasture. *Anim. Sci.* 73, 85–95. doi: 10.1017/S1357729800058082

Carbajal, S., and Orihuela, A. (2001). Minimal number of conspecifics needed to minimise the stress response of isolated mature ewes. *J. Appl. Anim. Welfare Sci.* 4, 249. doi: 10.1207/S15327604JAWS0404_02

Casuriaga, D., Ungerfeld, R., Guerrero, M., Ciappesoni, G., del Pino, L., and Freitas-de-Melo. (2021), Twin lambs: emotional attachment and behavioral response at weaning. *Rev. Fac. Nac. Agr. Med. (Colombia)* 74, 166.

Catherine, M. D., and Alistair, B. L. (2000). Maternal behaviour in domestic sheep (Ovis aries): constancy and change with maternal experience. *Behaviour*. 137, 1391–1413.

Chniter, M., Hammadi, M., Khorchani, T., Krit, R., Lahsoumi, B., Sassi, M. B., et al. (2011). Phenotypic and seasonal factors influence birth weight, growth rate and lamb mortality in D'man sheep maintained under intensive management in Tunisian oases. *Small Rumin. Res.* 99, 166–170. doi: 10.1016/j.smallrumres.2011.03.046

Clutton-Brock, T. H., Albon, S. D., and Guinness, F. E. (1981). Parental investment in male and female offspring in polygynous mammals. *Nature* 289, 487–489. doi: 10.1038/289487a0

Cockram, M. S., Imlah, P., Goddard, P. J., Harkiss, G. D., and Waran, N. K. (1993). The behavioural, endocrine and leucocyte response of ewes to repeated removal of lambs before the age of natural weaning. *Appl. Anim. Behav. Sci.* 38, 127–142. doi: 10.1016/0168-1591(93)90061-S

da Porciuncula, G. C., da Costa, J. A. A., Catto, J. B., Reis, F. A., and Fischer, V. (2021). Behavior of pantaneiro ewes and their offspring. *J. Vet. Behav.* 44, 40–49. doi: 10.1016/j.jveb.2021.05.006

Damián, J. P., Beracochea, F., Machado, S., Hötzel, M. J., Banchero, G., and Ungerfeld, R. (2018b). Growing without a mother results in poorer sexual behaviour in adult rams. *Animal* 12, 98–105. doi: 10.1017/S1751731117001574

Damián, J. P., Hötzel, M. J., Banchero, G., and Ungerfeld, R. (2013). Behavioural response of grazing lambs to changes associated with feeding and separation from their mothers at weaning. *Res. Vet. Sci.* 95, 913–918. doi: 10.1016/j.rvsc.2013.08.001

Damián, J. P., Hötzel, M. J., Banchero, G., and Ungerfeld, R. (2017). Competition for oestrous ewes between rams reared by their mothers or artificially reared: Effects on sexual behaviour and testosterone and cortisol serum concentrations. *Theriogenology* 100, 134–138. doi: 10.1016/j.theriogenology.2017.06.009

Damián, J. P., Hötzel, M. J., Banchero, G., and Ungerfeld, R. (2018a). Growing without a mother during rearing affects the response to stressors in rams. *Appl. Anim. Behav. Sci.* 209, 36–40. doi: 10.1016/j.applanim.2018.08.022

Dhaoui, A., Chniter, M., Lévy, F., Nowak, R., and Hammadi, M. (2020). Does lambing season affect mother-young relationships and lamb vigor in D'man sheep reared in oases? *Animal* 14, 2363–2371. doi: 10.1017/S1751731120001342

Dwyer, C. M. (2014). Maternal behaviour and lamb survival: from neuroendocrinology to practical application. *Animal* 8, 102–112. doi: 10.1017/S1751731113001614

Dwyer, C. M., and Lawrence, A. B. (1998). Variability in the expression of maternal behaviour in primiparous sheep: effects of genotype and litter size. *Appl. Anim. Behav. Sci.* 58, 311–330. doi: 10.1016/S0168-1591(97)00148-2

Dwyer, C. M., and Lawrence, A. B. (2000). Maternal behaviour in domestic sheep (Ovis aries): constancy and change with maternal experience. *Behaviour* 137, 1391–1413. doi: 10.1163/156853900501999

Dwyer, C. M., Lawrence, A. B., Bishop, S. C., and Lewis, M. (2003). Ewe-lamb bonding behaviours at birth are affected by maternal undernutrition in pregnancy. *Brit. J. Nutr.* 89, 123–136. doi: 10.1079/BJN2002743

Enríquez, D., Hötzel, M. J., and Ungerfeld, R. (2011). Minimising the stress of weaning of beef calves: a review. *Acta Vet. Scand.* 53, 28–36. doi: 10.1186/1751-0147-53-28

Enríquez, D. H., Ungerfeld, R., Quintans, G., Guidoni, A. L., and Hötzel, M. J. (2010). The effects of alternative weaning methods on behaviour in beef calves. *Livest. Sci.* 128, 20–27. doi: 10.1016/j.livsci.2009.10.007

Fazio, E., Ferlazzo, A., Cravana, C., and Medica, P. (2015). Effects of weaning on total and free iodothyronines in lambs. *Vet. Q.* 35, 16–20. doi: 10.1080/01652176.2014.989624

Festa-Bianchet, M., Jorgenson, J. T., and Wuhart, W. D. (1994). Early weaning in bighorn sheep, Ovis canadensis affects growth of males but not of females. *Behav. Ecol.* 5, 21–27. doi: 10.1093/beheco/5.1.21

Fletcher, I. C. (1971). Relationships between frequency of suckling, lamb growth and post-partum oestrous behaviour in ewes. *Anim. Behav.* 19, 108–111. doi: 10.1016/S0003-3472(71)80142-2

Fonsêca, V. F. C., Saraiva, E. P., Arruda, M. F., Pereira, W. E., Pimenta Filho, E. C., Santos, S. G. C. G., et al. (2016). Mother-offspring relationship in Morada Nova sheep bred in a tropical semiarid environment: a perspective on maternal investment and parental conflict. *Appl. Anim. Behav. Sci.* 183, 51–58. doi: 10.1016/j.applanim.2016.07.002

Fonsêca, V. F. C., Saraiva, E. P., Pimenta Filho, E. C., Furtado, D. A., Mariz, T. M. A., Silva, A. L., et al. (2014). Influence of the climatic environment and duration of labor on the mother-offspring interaction in Morada Nova sheep. *J. Anim. Sci.* 92, 4123–4129. doi: 10.2527/jas.2014-7322

Freitas-de-Melo, A., Angélica, T., Ungerfeld, R., Orihuela, A., Hötzel, M., and Pérez-Clariget, R. (2018b). Influence of low pasture allowance during pregnancy on the attachment between ewes and their lambs at birth and during lactation. *Appl. Anim. Behav. Sci.* 199, 9–16. doi: 10.1016/j.applanim.2017.10.016

Freitas-de-Melo, A., Banchero, G., Hötzel, M. J., Damián, J. P., and Ungerfeld, R. (2013). Progesterone administration reduces the behavioural and physiological responses of ewes to abrupt weaning of lambs. *Animal* 8, 1367–1373. doi: 10.1017/S1751731113000621

Freitas-de-Melo, A., Pérez-Clariget, R., Terrazas, A., and Ungerfeld, R. (2021a). Ewe-lamb bond of experienced and inexperienced mothers undernourished during gestation. *Sci. Rep.* 11, 1–9. doi: 10.1038/s41598-021-84334-2

Freitas-de-Melo, A., and Ungerfeld, R. (2016a). Destete artificial en ovinos: respuesta de estrés y bienestar animal. *Revisión. Rev. Mex. Cienc. Pecu.* 7, 361–375. doi: 10.22319/rmcp.v7i3.4215

Freitas-de-Melo, A., and Ungerfeld, R. (2016b). Progesterona y respuesta de estrés: mecanismos de acción y sus repercusiones en rumiantes domésticos. *Rev. Mex. Cienc. Pecu.* 7, 185–199. doi: 10.22319/rmcp.v7i2.4173

Freitas-de-Melo, A., and Ungerfeld, R. (2020). The sex of the offspring affects the lamb and ewe responses to abrupt weaning. *Appl. Anim. Behav. Sci.* 229, 105008. doi: 10.1016/j.applanim.2020.105008

Freitas-de-Melo, A., Ungerfeld, R., Bernal Barragán, H., Lombardo Morales, J. A., and Sánchez-Dávila, F. (2021c), Lambs repeatedly separated from their dams display a greater response to social isolation after weaning. *Rev. Fac. Nac. Agr. Med.* 74, 166.

Freitas-de-Melo, A., Ungerfeld, R., Hötzel, M., Abud, M., Alvarez-Oxley, A., Orihuela, A., et al. (2015a). Mother-young bonding in grazing ewes: Effects of sex and food restriction in pregnancy. *Appl. Anim. Behav. Sci.* 168, 31–36. doi: 10.1016/j.applanim.2015.04.009

Freitas-de-Melo, A., Ungerfeld, R., Hötzel, M., Orihuela, A., and Pérez-Clariget, R. (2017). Low pasture allowance until late gestation in ewes: behavioural and physiological changes in ewes and lambs from lambing to weaning. *Animal* 11, 285–294. doi: 10.1017/S1751731116001427

Freitas-de-Melo, A., Ungerfeld, R., and Orihuela, A. (2021b). Behavioral and physiological responses to early weaning in ewes and their single or twin lambs. *Trop. Anim. Health Prod.* 53, 1–12. doi: 10.1007/s11250-021-02589-3

Freitas-de-Melo, A., Ungerfeld, R., Orihuela, A., Hötzel, M., and Pérez-Clariget, R. (2018a). Early mother-young relationship and feeding behaviour of lambs are unaffected by low pasture allowance until the beginning of the last third of gestation in single bearing ewes. *Anim. Prod. Sci.* 58, 930–936. doi: 10.1071/AN16157

Freitas-de-Melo, A., Ungerfeld, R., and Pérez-Clariget, R. (2015b). Progesterone administration reduces the behavioural response of ewes at weaning. *Book Abst. 66th Ann. Meet. Eur. Ass. Anim. Prod. (EAAP)*, Warsaw, Poland, 31 Aug.-—4 Sep., 21, 369.

Freitas-de-Melo, A., Ungerfeld, R., and Pérez-Clariget, R. (2019). Behavioral pattern in Texel × Corriedale terminal crossbreeding: Maternal behavior score at birth, lambs' feeding behaviors, and behavioral responses of lambs to abrupt weaning. *J. Vet. Behav.* 30, 9–15. doi: 10.1016/j.jveb.2018.10.007

Galeana, L., Orihuela, A., Aguirre, V., and Vázquez, R. (2007). Mother-young spatial association and its relation with proximity to a fence separating ewes and lambs during enforce weaning in hair sheep (*Ovis aries*). *Appl. Anim. Behav. Sci.* 108, 81. doi: 10.1016/j.applanim.2006.10.016

Gaudin, S., Chaillou, E., Cornilleau, F., Moussu, C., Boivin, X., and Nowak, R. (2015). Daughters are more strongly attached to their mother than sons: a possible mechanism for early social segregation. *Anim. Behav.* 102, 33–43. doi: 10.1016/j.anbehav.2015.01.003

Geist, V. (1971). *Mountain Sheep: A Study in Behaviour Evolution*. Chicago: University of Chicago Press.

Godfray, H. C. J. (1995). Evolutionary theory of parental-offspring conflict. *Nature* 376, 133–138. doi: 10.1038/376133a0

Godfrey, R. W., and Weis, A. J. (2016). Effect of weaning age on hair sheep lamb and ewe production traits in an accelerated lambing system in the tropics. *J. Anim. Sci.* 94, 1250–1254. doi: 10.2527/jas.2015-9987

Goursaud, A. P., and Nowak, R. (1999). Colostrum Mediates the Development of Mother Preference by Newborn Lambs. *Physiol. Behav.* 67, 49–56. doi: 10.1016/S0031-9384(99)00037-2

Grubb, P. (1974). "Social organization of Soay sheep and the behaviour of ewes and lambs" in *The Ecology of the Soay Sheep of St. Kilda*, ed. P. Jewell, C. Milner, J. Boyd (London: Athlone Press), 131–159.

Hague, P., Shillito Walser, E., and Walters, E. (1982). Vocal communication between ewes and their own and alien lambs. *Behaviour* 81, 140–150. doi: 10.1163/156853982X00120

Haley, D. B., Bailey, D. W., and Stookey, J. M. (2005). The effects of weaning beef calves in two stages on their behavior and growth rate. *J. Anim. Sci.* 83, 2205–2214. doi: 10.2527/2005.8392205x

Henrique, F. L., Titto, E. A. L., Zanella, A. J., Hooper, H. B., Pulido-Rodriguez, L. F., Longo, A. L. S., et al. (2017). Simulated disease process during late pregnancy compromises developmental outcomes of lambs independently of the weaning method applied. *Small Ruminant Res.* 155, 39–44. doi: 10.1016/j.smallrumres.2017.09.004

Hernandez, C. E., Matthews, L. R., Oliver, M. H., Bloomfield, F. H., and Harding, J. E. (2009). Effects of sex, litter size and periconceptional ewe nutrition on the ewe–lamb bond. *Appl. Anim. Behav. Sci.* 120, 76–83. doi: 10.1016/j.applanim.2009.04.012

Hinch, G. N. (1989). The sucking behaviour of triplet, twin and single lambs at pasture. *Appl. Anim. Behav. Sci.* 22, 39–48. doi: 10.1016/0168-1591(89)90078-6

Hinch, G. N., Lecrivain, E., Lynch, J. J., and Elwin, R. L. (1987). Changes in maternal-young associations with increasing age of lambs. *Appl. Anim. Behav. Sci.* 17, 305–318. doi: 10.1016/0168-1591(87)90154-7

Högberg, N., Hessle, A., Lidfors, L., Enweji, N., and Höglund, J. (2021). Nematode parasitism affects lying time and overall activity patterns in lambs following pasture exposure around weaning. *Vet. Parasitol.* 296, 109500. doi: 10.1016/j.vetpar.2021.109500

Kendrick, K. M., and Keverne, E. B. (1991). Importance of progesterone and estrogen priming for the induction of maternal behavior by vaginocervical stimulation in sheep: effects of maternal experience. *Physiol. Behav.* 49, 745–750. doi: 10.1016/0031-9384(91)90313-D

Kenyon, P. R., and Cranston, L. M. (2017). "Nutritional management" in *Advances in Sheep Welfare*, eds. M.D. Ferguson, C. Lee, A. Fisher (United Kingdom: Elsevier), 153–175.

Lane, M. A., Baldwin, R. L., and Jesse, B. W. (2000). Sheep rumen metabolic development in response to age and dietary treatments. *J. Anim. Sci.* 78, 1990. doi: 10.2527/2000.7871990x

Lawrence, A. B., and Dwyer, C. M. (1999). Does the behaviour of the neonate influence the expression of maternal behaviour in sheep? *Behaviour* 136, 367–389. doi: 10.1163/156853999501379

Lévy, F., and Keller, M. (2008). "Neurobiology of maternal behavior in sheep" in *Advances in the Study of Behavior*, eds. Brockman H.J., Roper T.J., Naguib M., Wynne-Edwards K.E., Barnard C., Mitani, M. (Amsterdam: Elsevier), 399–437.

Lévy, F., Locatelli, A., Piketty, V., Tillet, Y., and Poindron, P. (1995). Involvement of the main but not the accessory olfactory system in maternal behavior of primiparous and multiparous ewes. *Physiol. Behav.*, 57, 97–104. doi: 10.1016/0031-9384(94)00200-O

Li, C., Wang, W., Liu, T., Zhang, Q., Wang, G., Li, F., et al. (2018). Effect of early weaning on the intestinal microbiota and expression of genes related to barrier function in lambs. *Front. Microbiol.* 9, 1431. doi: 10.3389/fmicb.2018.01431

Lyford, S. J. (1988). "Growth and development of the ruminant digestive system" in *The Ruminant Animal*, ed. D.C. Curch (New Jersey: Prentice-Hall), 44–63.

Lynch, J. J., Hinch, G. N., and Adams, D. B. (1992). *The Behaviour of Sheep: Biological Principles and Implications for Production*. Melbourne: CSIRO.

Maldonado, A., Orihuela, A., Aguirre, V., Vázquez, R., and Flores-Pérez, I. (2015). Changes in mother-offspring relationships with the increasing age of the lamb in hair sheep (*Ovis aries*). *J. Vet. Behav.* 10, 166–170. doi: 10.1016/j.jveb.2014.11.008

Mandal, A., Prasad, H., Kumar, A., Roy, R., and Sharma, N. (2007). Factors associated with lamb mortalities in Muzaffarnagari sheep. *Small Ruminant Res.* 71, 273–279. doi: 10.1016/j.smallrumres.2006.07.010

Martínez, M. E., de la Barra, R., and de la Fuente, F. (2015). Effect of early creep feeding in the performance of Chilota Breed lambs. *Livestock Sci.* 6, 56–64.

McCoard, S. A., Cristobal-Carballo, O., Knol, F. W., Heiser, A., Khan, M. A., Hennes, N., et al. (2020). Impact of early weaning on small intestine, metabolic, immune and endocrine system development, growth and body composition in artificially reared lambs. *J. Anim. Sci.* 98, skz356. doi: 10.1093/jas/skz356

McKusick, B. C., Thomas, D. L., and Berger, Y. M. (2001). Effect of weaning system on commercial milk production and lamb growth of East Friesian dairy sheep. *J. Dairy Sci.* 84, 1660–1668. doi: 10.3168/jds.S0022-0302(01)74601-2

Mears, G. J., and Brown, F. A. (1997). Cortisol and b-endorphin responses to physical and psychological stressors in lambs. *Can. J. Anim. Sci.* 77, 689–694. doi: 10.4141/A97-051

Menant, O., Ungerfeld, R., Lévy, F., Pérez-Clariget, R., and Freitas-De-Melo, A. (2022). Out-of-season breeding and ewe-lamb bond from birth to weaning in Corriedale sheep. *Appl. Anim. Behav. Sci.* 21:542. doi: 10.1016/j.applanim.2021.105542

Menant, O., Ungerfeld, R., Pérez-Clariget, R., and Freitas-De-Melo, A. (2020). Is body surface temperature measured on the single lambs' back a reliable indicator of the ewe-lamb bond around birth*? J. Thermal Biol.* 93, 102699. doi: 10.1016/j.jtherbio.2020.102699

Mepham, T. B., and Kuhn, N. J. (1994). "Physiology and biochemistry of lactation" in *Marshall's physiology of reproduction, Pregnancy and Lactation Part One: Ovulation and early pregnancy*., ed. G.E. Lamming (New York, NY: Springer), 1102–1186.

Mohapatra, A., De, K., Saxena, V. K., Mallick, P. K., Devi, I., and Singh, R. (2020). Behavioral and physiological adjustments by lambs in response to weaning stress. *J. Vet. Behav.* 41, 47–51. doi: 10.1016/j.jveb.2020.07.009

Mora-Medina, P., Orihuela, A., Arch-Tirado, E., Roldán-Santiago, P., Terrazas, A., and Mota-Rojas, D. (2016). Sensory factors involved in mother-young bonding in sheep: a review. *Vet. Med.-Czech.* 61, 595–611. doi: 10.17221/255/2014-VETMED

Morales-Terán, G., Herrera, C. C. A., Pérez, H. P., Salazar, O. J., and Gallegos, S. J. (2011). Influence of controlled suckling and the male effect on the resumption of postpartum ovarian activity in Pelibuey sheep. *Trop. Subtrop. Agroecosyst.* 13, 493.

Morales-Terán, G., Pro, M. A., Figueroa, S. B., Sánchez del Real, C., and Gallegos, S. J. (2004). Continuous or restricted suckling and its relationship to the length of postpartum anestrous in Pelibuey ewes. *Agrociencia* 38, 165. http://dx.doi.org/urn:ISSN:1870-0462-tsaes.v13i3.1269

Morgan, P. D., Boundy, C. A. P., Arnold, G. W., and Lindsay, D. R. (1985). "The roles played by the senses of the ewe in the location and recognition of lambs," in *Reproductive and Developmental Behaviour in Sheep*, ed. A.F. Fraser. (Amsterdam: Elsevier), 181–192.

Munro, J. (1956). Observations on the suckling behaviour of young lambs. *Brit. J. Anim. Behav.* 4, 34–36. doi: 10.1016/S0950-5601(56)80014-2

Napolitano, F., Annicchiarico, G., Caroprese, M., De Rosa, G., Taibi, L., and Sevi, A. (2003). Lambs prevented from suckling their mothers display behavioural, immune, and endocrine disturbances. *Physiol. Behav.* 78, 81. doi: 10.1016/S0031-9384(02)00892-2

Napolitano, F., Marino, V., De Rosa, G., Capparelli, R., and Bordi, A. (1995). Influence of artificial rearing on behavioral and immune response of lambs. *Appl. Anim. Behav. Sci.* 45,245–253. doi: 10.1016/0168-1591(95)00637-8

Napolitano, F., Rosa, D., and Sevi, A. (2008). Welfare implications of artificial rearing and early weaning in sheep. *Appl. Anim. Behav. Sci.* 110, 58–72. doi: 10.1016/j.applanim.2007.03.020

Newberry, R. C., and Swanson, J. C. (2008). Implications of breaking mother–young social bonds. *Appl. Anim. Behav. Sci.* 110, 3–23. doi: 10.1016/j.applanim.2007.03.021

Norouzian, M. A. (2015). Effect of weaning method on lamb behaviour and weight gain. *Small Ruminant Res.* 133, 17–20. doi: 10.1016/j.smallrumres.2015.10.028

Nowak, R. (2006). "Suckling, milk, and the development of preferences towards maternal cues by neonates: from early learning to filial attachment? " in *Advances in the Study of Behaviour*, eds. H.J. Brockmann, P.J.B. Slater, C.T. Snowdon, T.J. Roper, M. Naguib, K.E. Wynne-Edwards (Amsterdam: Elsevier), 1–358.

Nowak, R., Lévy, F., Chaillou, E., Cornilleau, F., Cogni,é, J., Marnet, P.-G., et al. (2021). Neonatal suckling, oxytocin, and early infant attachment to the mother. *Front. Endocrinol.* 11, 2651. doi: 10.3389/fendo.2020.612651

Nowak, R., Murphy, T. M., Lindsay, D. R., Alster, P., Andersson, R., and Uvnäs-Moberg, K. (1997). Development of a preferential relationship with the mother by the newborn lamb: importance of the sucking activity. *Physiol. Behav.* 62, 681–688. doi: 10.116/S0031-9384(97)00079-6

Nowak, R., and Poindron, P. (2006). From birth to colostrum: early steps leading to lamb survival. *Reprod. Nutr. Dev.* 46, 431–446. doi: 10.1051/rnd:2006023

Nowak, R. F., and Lindsay, D. R. (1992). Discrimination of merino ewes by their newborn lambs: important for survival? *Appl. Anim. Behav. Sci.* 34, 61–74. doi: 10.1016/S0168-1591(05)80057-7

O'Connor, C. E., Lawrence, A. B., and Wood-Gush, D. G. M. (1992). Influence of litter size and parity on maternal behaviour at parturition in Scottish Blackface sheep. *Appl. Anim. Behav. Sci.* 33, 345–355. doi: 10.1016/S0168-1591(05)80071-1

Orgeur, P., Bernard, S., Naciri, M., Nowak, R., Schaal, B., and Lévy, F. (1999). Psycological consequences of two different weaning methods in sheep. *Reprod. Nutr. Dev.* 39, 231–244. doi: 10.1051/rnd:19990208

Orgeur, P., Mavric, N., Yvore, P., Bernard, S., Nowak, R., Schaal, B., et al. (1998). Artificial weaning in sheep: consequences on behavioural, hormonal and immuno-pathological indicators of welfare. *Appl. Anim. Behav. Sci.* 58, 87–103. doi: 10.1016/S0168-1591(97)00140-8

Orihuela, A., Mota-Rojas, D., Ungerfeld, R., Napolitano, F., Strappini, A., Giuseppe, R., et al. (2019). "Mecanismos neurofisiológicos de la impronta en la búfala y otros animales de granja. " in *El Búfalo de Agua en las Américas, Enfoques Prácticos y Experimentales*. eds. I. Guerrero Lagarreta, F. Napolinato, D. Mota-Rojas (México: BM Editores), 381–425.

Orihuela, A., Suárez, E., and and, Vázquez, R. (2004). Effect of restricting suckling on the social bond between ewes and their 10-week-old lambs. *Livest. Prod. Sci.* 87, 259. doi: 10.1016/j.livprodsci.2003.08.003

Orihuela, A., Valdez, D., and Ungerfeld, R. (2016). The effect of permanent or temporary contact with the lamb and contact with males on the lambing to first ovulation interval in Saint Croix sheep. *Appl. Anim. Behav. Sci.* 181, 100 doi: 10.1016/j.applanim.2016.05.009

Owen, J. B., Davies, D. A. R., and Ridgman, W. J. (1969a). Effects of varying the quantity and distribution of liquid feed in lambs reared artificially. *Anim. Prod.* 2, 1. doi: 10.1017/S0003356100026556

Owen, J. B., Davies, D. A. R., and Ridgman, W. J. (1969b). The control of voluntary food intake in ruminants. *Anim. Prod.* 2, 511. doi: 10.1017/S000335610002852X

Ozella, L., Price, E., Langford, J., Lewis, K. E., Cattuto, C., and Croft, D. P. (2022). Association networks and social temporal dynamics in ewes and lambs. *Appl. Anim. Behav. Sci.* 246, 105515. doi: 10.1016/j.applanim.2021.105515

Pascual-Alonso, M., Miranda-de la Lama, G. C., Aguayo-Ulloa, L., Ezquerro, L., Villarroel, M., Marín, R. H., et al. (2015). Effect of postweaning handling strategies on welfare and productive traits in lambs. *J. Appl. Anim. Welfare Sci.* 18, 42–56. doi: 10.1080/10888705.2014.941107

Pérez-León, I., Orihuela, A., Lidfors, L., and Aguirre, V. (2006). Reducing mother young separation distress by inducing ewes into oestrous into day of weaning. *Anim. Welfare* 15, 383–389.

Pickup, H. E., and Dwyer, C. M. (2011). Breed differences in the expression of maternal care at parturition persist throughout the lactation period in sheep. *Appl. Anim. Behav. Sci.* 132, 33–41. doi: 10.1016/j.applanim.2011.03.010

Poindron, P. (2005). Mechanisms of activation of maternal behaviour in mammals. *Reprod. Nutr. Dev.* 45, 341–351. doi: 10.1051/rnd:2005025

Poindron, P., and Carrick, M. J. (1976). Hearing recognition of the lamb by its mother. *Anim. Behav.* 24, 600–602. doi: 10.1016/S0003-3472(76)80073-5

Poindron, P., Lévy, F., and Keller, M. (2007). Maternal responsiveness and maternal selectivity in domestic sheep and goats: the two facets of maternal attachment. *Dev. Psychobiol.* 49, 54–70. doi: 10.1002/dev.20192

Poindron, P., and Neindre, P. L. (1980). "Endocrine and sensory regulation of maternal behavior in the ewe," in *Advances in the Study of Behavior*, eds. J.S. Rosenblatt, R.A. Hinde, C. Beer and M.C. Busnel (New York, NY: Academic Press), 75–119.

Salva, O. R., Normando, S., Mollo, A., and Regolin, L. (2014). Novelty preference in face perception by week-old lambs. *Interact. Stud.* 15, 113–128. doi: 10.1075/is.15.1.05ros

Schichowski, C., Moors, E., and Gauly, M. (2008). Effects of weaning lambs in two stages or by abrupt separation on their behavior and growth rate. *J. Anim. Sci.* 86, 220. doi: 10.2527/jas.2007-0198

Schichowski, C., Moors, E., and Gauly, M. (2010). Influence of weaning age and an experimental Haemonchus contortus infection on behaviour and growth rates of lambs. *Appl. Anim. Behav. Sci.* 125, 103–108. doi: 10.1016/j.applanim.2010.04.014

Sèbe, F., Duboscq, J., Aubin, T., Ligout, S., and Poindron, P. (2010). Early vocal recognition of mother by lambs: contribution of low- and high-frequency vocalizations. *Anim. Behav.* 79, 1055–1066. doi: 10.1016/j.anbehav.2010.01.021

Sèbe, F., Nowak, R., Poindron, P., and Aubin, T. (2007). Establishment of vocal communication and discrimination between ewes and their lamb in the first two days after parturition. *Dev. Psychobiol.* 49, 375–386. doi: 10.1002/dev.20218

Sevi, A., Caroprese, M., Annicchiarico, G., Albenzio, M., Taibi, L., and Muscio, A. (2003). The effect of a gradual separation from the mother on later behavioral, immune and endocrine alterations in artificially reared lambs. *Appl. Anim. Behav. Sci.* 83, 41–53. doi: 10.1016/S0168-1591(03)00088-1

Sevi, A., Casamassima, D., Pulina, G., and Pazzona, A. (2009). Factors of welfare reduction in dairy sheep and goats. *Ital. J. Anim. Sci.* 8, 81–101. doi: 10.4081/ijas.2009.s1.81

Shackleton, D. M., and Haywood, J. (1985). Early mother–young interactions in California bighorn sheep, *Ovis Canadensis californiana*. *Can. J. Zool.* 63, 868–875. doi: 10.1139/z85-129

Shaw, K. L., Nolan, J. V., Lynch, J. J., Coverdale, O. R., and Gill, H. S. (1995). Effects of weaning, supplementation and gender on acquired immunity to Haemonchus contortus in lambs. *Int. J. Parasitol.* 25, 381–387. doi: 10.1016/0020-7519(94)00098-9

Shipka, M. P., and Ford, S. P. (1991). Relationship of circulating estrogen and progesterone concentrations during late pregnancy and the onset phase of maternal behavior in the ewe. *Appl. Anim. Behav. Sci.* 31, 91–99. doi: 10.1016/0168-1591(91)90156-R

Smeaton, D. C., Sumner, R. M. W., Knight, T. W., and Wadams, T. K. (1983). Effects of time of weaning, pasture allowance, and shearing time on ewe and lamb liveweight, wool growth, and subsequent ovulation rate of the ewe. *New Zeal. J. Exp. Agr.* 11, 41–45. doi: 10.1080/03015521.1983.10427725

Smith, F. V., Van-Toller, C., and Boyes, T. (1966). The 'critical period' in the attachment of lambs and ewes. *Anim. Behav.* 14, 120–125. doi: 10.1016/S0003-3472(66)80019-2

Sowinska, J., Brzostowski, H., Tanski, Z., and Czaja, K. (2001). The weaning stress response in lambs of different age. *Czech J. Anim. Sci.* 46, 465–468.

Stephens, D. B., and Baldwin, B. A. (1971). Observations on the behaviour of groups of artificially reared lambs. *Res. Vet. Sci.* 12, 219–224. doi: 10.1016/S0034-5288(18)34182-1

Sun, D. M., Mao, S. Y., Zhu, W. Y., and Liu, J. H. (2018). Effect of starter diet supplementation on rumen epithelial morphology and expression of genes involved in cell proliferation and metabolism in pre-weaned lambs. *Animal* 12, 2274–2283. doi: 10.1017/S1751731118000290

Tadich, N., Gallo, C., Brito, M. L., and Broom, D. M. (2009). Effects of weaning and 48 h transport by road and ferry on some blood indicators of welfare in lambs. *Livest. Sci.* 121, 132–136. doi: 10.1016/j.livsci.2008.06.001

Terrazas, A., Nowak, R., Serafín, N., Ferreira, G., Lévy, F., and Poindron, P. (2002). Twenty-four-hour-old lambs rely more on maternal behavior than on the learning of individual characteristics to discriminate between their own and an alien mother. *Dev. Psychobiol.* 40, 408–418. doi: 10.1002/dev.10041

Thórhallsdóttir, A. G., Provenza, F. D., and Balph, D. F. (1990). The role of the mother in the intake of harmful foods by lambs. *Appl. Anim. Behav. Sci.* 25, 35. doi: 10.1016/0168-1591(90)90067-N

Trivers, R. L. (1974). Parent-offspring conflict. *Am. Zool.* 14, 249–264. doi: 10.1093/icb/14.1.249

Turkson, P. K., and Sualisu, M. (2005). Risk factors for lamb mortality in Sahelian sheep on a breeding station in Ghana. *Trop. Anim. Health Prod.* 37, 49–64. doi: 10.1023/B:TROP.0000047935.78168.46

Ungerfeld, R., Fernández-Werner, A., Gökdal, Ö., Atay, O., and Freitas-De-Melo, A. (2021b). Lambs identify their mothers' bleats but not a picture of her face. *J. Vet. Behav.* 46, 69–73. doi: 10.1016/j.jveb.2021.07.009

Ungerfeld, R., Freitas-De-Melo, A., Nowak, R., and Lévy, F. (2018). Preference for the mother does not last long after weaning at 3 months of age in sheep. *Appl. Anim. Behav. Sci.* 205, 28–33. doi: 10.1016/j.applanim.2018.05.018

Ungerfeld, R., Hötzel, M. J., and Quintans, G. (2015). Changes in behaviour, milk production and bodyweight in beef cows subjected to two-step or abrupt weaning. *Anim. Product. Sci.* 55, 1281–1288. doi: 10.1071/AN13453

Ungerfeld, R., Pérez-Clariget, R., Menant, O., and Freitas-de-Melo, A. (2021a). Conception during the lactation and rearing period affects the ewe-lamb bond at birth. *Appl. Anim. Behav. Sci.* 236, 105256. doi: 10.1016/j.applanim.2021.105256

Ungerfeld, R., and Sánchez-Dávila, F. (2012). Oestrus synchronisation in postpartum autumn-lambing ewes: effect of postpartum time parity, and early weaning. *Span. J. Agric. Res.* 10, 62. doi: 10.5424/sjar/2012101-233-11

Val-Laillet, D., Simon, M., and Nowak, R. (2004). A full belly and colostrum: Two major determinants of filial love. *Dev. Psychobiol.* 45, 163–173. doi: 10.1002/dev.20024

Vatankhah, M., Talebi, M. A., and Zamani, F. (2012). Relationship between ewe body condition score (BCS) at mating and reproductive and productive traits in Lori-Bakhtiari sheep. *Small Ruminant Res.* 106, 105–109. doi: 10.1016/j.smallrumres.2012.02.004

Vatankhaha, M., and Salehi, S. A. (2010). Genetic and non-genetic factors affecting Lori-Bakhtiari ewe body weight and its relationship with productivity. *Small Ruminant Res.* 94, 98–102. doi: 10.1016/j.smallrumres.2010.07.006

Wang, S., Ma, T., Zhao, G., Zhang, N., Tu, Y., Li, F., et al. (2019). Effect of age and weaning on growth performance, rumen fermentation, and serum parameters in lambs fed starter with limited ewe–lamb interaction. *Animals* 9, 825. doi: 10.3390/ani9100825

Watson, D. L., and Gill, H. S. (1991). The effect of weaning on antibody responses and nematode parasitism in Merino lambs. *Res. Vet. Sci.* 51, 128–132. doi: 10.1016/0034-5288(91)90002-6

Weary, D. M., and Fraser, D. (1995). Signalling need: costly signals and animal welfare assessment. *Appl. Anim. Behav. Sci.* 44, 159–169. doi: 10.1016/0168-1591(95)00611-U

Weary, D. M., Jasper, J., and Hötzel, M. J. (2008). Understanding weaning distress. *Appl. Anim. Behav. Sci.* 110, 24–41. doi: 10.1016/j.applanim.2007.03.025

Wyse, C. A., Zhang, X., McLaughlin, M., Biello, S. M., Hough, D., Bellingham, M., et al. (2018). Circadian rhythms of melatonin and behaviour in juvenile sheep in field conditions: Effects of photoperiod, environment and weaning. *Physiol. Behav.* 194, 362–370. doi: 10.1016/j.physbeh.2018.06.001

Youssef, M. Y. I., Phillips, C. J. C., and Metwally, M. (1995). The effect of pre-weaning grazing experience and presence of adult ewes on grazing behaviour of weaned lambs. *Appl. Anim. Behav. Sci.* 2, 281. doi: 10.1016/0168-1591(95)92372-Z

Zhang, Q., Chong, L., Xiaolin, N., Zhian, Z., Fadi, L., and Fei, L. (2018). An intensive milk replacer feeding program benefits immune response and intestinal microbiota of lambs during weaning. *BMC Vet. Res.* 14, 366. doi: 10.1186/s12917-018-1691-x

Zhong, R. Z., Sun, H. X., Li, G. D., Liu, H. W., and Zhou, D. W. (2014). Effects of inoculation with rumen fluid on nutrient digestibility, growth performance and rumen fermentation of early weaned lambs. *Livest. Sci.* 162, 154–158. doi: 10.1016/j.livsci.2013.12.021

Conflict of Interest: The authors declare that the research was conducted in the absence of any commercial or financial relationships that could be construed as a potential conflict of interest.

Publisher's Note: All claims expressed in this article are solely those of the authors and do not necessarily represent those of their affiliated organizations, or those of the publisher, the editors and the reviewers. Any product that may be evaluated in this article, or claim that may be made by its manufacturer, is not guaranteed or endorsed by the publisher.