Overview of the response of anoestrous ewes to the ram effect

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Abstract. The present review summarises knowledge of the reproductive response of anoestrous ewes to the introduction of rams - in other words, the ram effect. The ovarian and endocrine response, the factors that determine whether ewes will respond or not (associated with both the stimulus and the receptivity of the ewes) and some aspects of practical management are discussed. Information on the use of the ram effect to stimulate post-partum, prepubertal and cyclic ewes is also given. New insights are provided on ovarian response patterns, including recently collected information on luteal responses. The existence of delayed ovulation (5-7 days after the introduction of the rams) followed by luteal phases of normal or short length, luteal cysts and luteinised follicles is reported after scanning the ovaries with ultrasound. Endocrine parameters for depth of anoestrus, such as TH pulsatility and FSH concentrations, and how the concentrations of these hormones should be considered are discussed. Particular attention is paid to the observation of spontaneous, higher TH pulsatility and higher FSH concentrations in anoestrous ewes that respond to rams with luteal phases than in those that fail to respond. The use of progestogen priming and single progestogen administration and the possible advantages for synchronisation of oestrus are also discussed. Other factors that should be considered before the ram effect is applied, such as the strength of the stimulus and some practical considerations, are also included.

Extra keywords: biostimulation, pheromones, sheep, seasonal anoestrus.

Introduction

The reproductive response of anoestrous ewes to the introduction of rams (i.e. the ram effect) has long been established (Underwood *et al.* 1944). Part of the flock may ovulate, come into oestrus and become pregnant as a result of the stimulus provided by rams. The ram effect is a useful and suitable tool for out-of-season oestrus induction, especially because its cost is negligible, so that it has become widely included in reproductive management (Martin *et al.* 1986). Similar reproductive effects, also referred to as biostimulation, have been reported in other ruminants, including goats (Chemineau 1987), cattle (Alberio *et al.* 1987), red deer (Moore and Cowie 1986), Eld's deer (Hosack *et al.* 1999), reindeer (Shipka *et al.* 2002), springbok (Skinner *et al.* 2002), moose (Miquelle 1991), oryx(Blanvillaine^a/. 1997),impala and blesbok (Skinner *et al.* 1992).

The evolution of sheep social structure

Few attempts (Lindsay 1988; Walkden-Brown *et al.* 1999) have been made to relate the existence of the ram effect in domestic sheep to similar phenomena in non-domesticated sheep. The high degree of reproductive synchrony observed in wild and feral female sheep might at least in part be a consequence of male introduction and other social interactions (Signoret *et al.* 1984). Throughout the year, the social structure is similar in wild and feral sheep breeds (Soay sheep: Grubb and Jewell 1973; Roclcy Mountain bighorn *(Ovis canadiensis canadiensis):* Geist 1971; Punjab Urial *(Ovis orientalis punjabiensis):* Schaller and Mirza 1974; mou-flon sheep: McClelland 1991) and farmed breeds (Romney sheep: Knight *et al.* 1998). Outside the rutting period, social groups are composed of several females with their offspring, or of males belonging to small, exclusively male groups (Stricklin and Mench 1987). When male offspring become mature they leave the female group (Shackleton and Schank 1984). Wethers remain together or join a group of females (Jewell 1997), suggesting that the testes - probably through androgens - are involved in the segregation.

As the time of breeding approaches, males join the female groups. Nudging, blocking and rubbing (Jewell 1976) and aggressive behaviour (Lincoln and Davidson 1977) by males begin before females come into oestrus, probably because the male reproductive system is activated before the female reproductive system. Activation of the reproductive system includes increased pulsatility of luteinising hormone (LH) and increasing concentrations of follicle-stimulating hormone (FSH) and testosterone (for a review, see Lincoln and Short 1980). In sheep, natural joining may trigger - through the ram effect - an earlier onset of the breeding season, as has been seen in ewes allowed to remain in permanent contact with rams (Eldon 1993; O'Callaghan *et al.* 1994). In addition, there is evidence that blind ewes in permanent contact with rams (Legan and Karsch 1983) or pinealectomised ewes in a flock of rams and intact ewes (Wayne *et al.* 1989) have a more synchronous onset of the breeding season than do those that remain isolated.

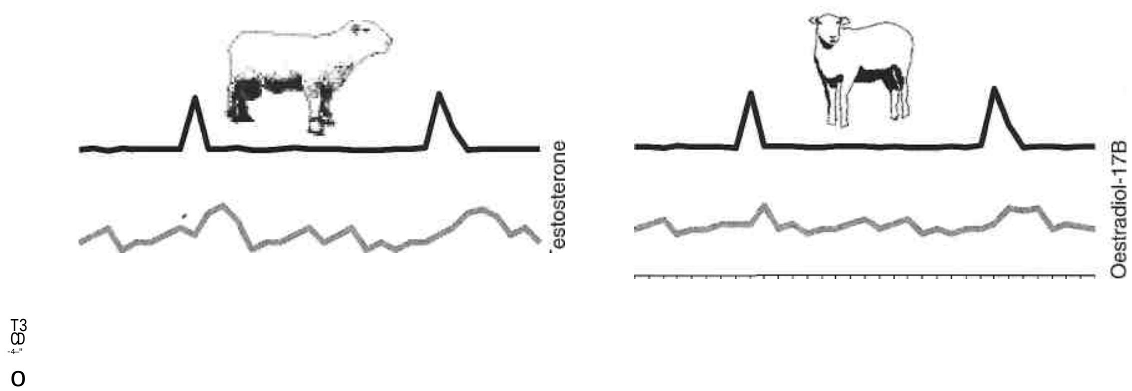
Since the rutting period causes an increase in energy expenditure in rams (Jewell 1997), we may ask what is the significance of beginning the reproductive season before ewes become cyclic? Moreover, does it make sense to have males triggering the female reproductive system? One answer is that the mechanism may be especially important in breeds that display a very short breeding season, such as Soay sheep (with only one to three oestrous cycles per year; Grubb and Jewell 1973). Moreover, late conception in Rocky Mountain bighorn sheep during the breeding season increases both lamb and ewe mortality (Hogg *et al.* 1992), so there is an advantage in any process that advances conception. The stimulus may also promote advancement of puberty in females and this may increase their lifetime reproductive success (Berube *et al.* 1999). The period from the joining of males and females until the peak of oestrus is also useful for males to establish hierarchical rank (Jewell 1976). Oestrus synchronisation allows different males to mate with different females, decreasing the risks of inbreeding.

How do ewes respond to the ram effect?

Endocrine and ovarian responses

The introduction of rams induces an increase in the pulsatile secretion ofLH in the ewes (Martin *et al.* 1983; Fig. 1), which may end with an LH surge followed by ovulation. This ovulation is not associated with oestrous behaviour. In some ewes, the first heat appears in conjunction with the second ovulation, 17-20 days after ram introduction. In others, there is an initial short luteal phase of 4-5 days, then a second ovulation without any signs of oestrus, followed by a luteal phase of normal duration. Thereafter, a third ovulation associated with oestrus occurs.

Laparoscopic (Cushwa *et al.* 1992) and ultrasonographic (Ungerfeld *etal.* 2002) observations have led to the discovery



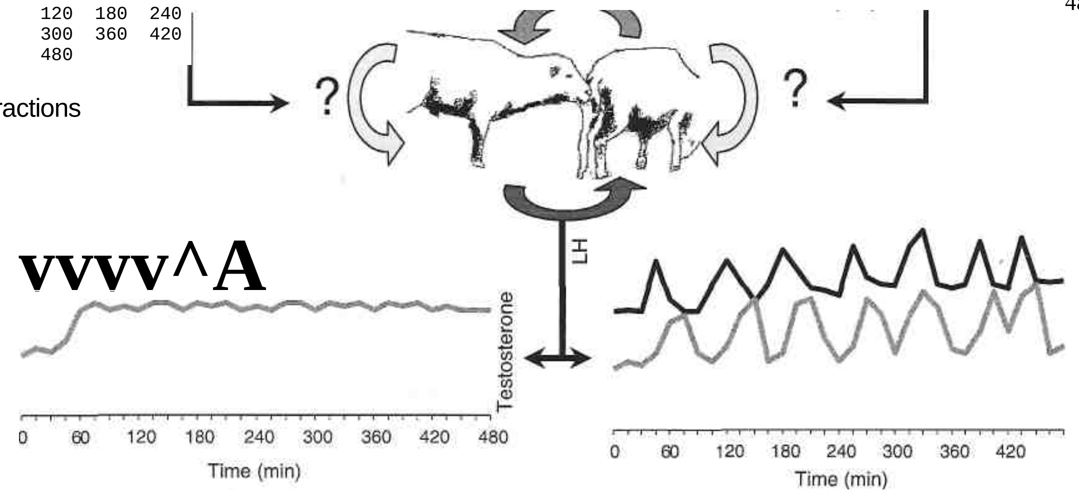


Fig. 1. Model of the endocrine response during the first hours after joining rams and ewes during the non-breeding season. The curved arrows indicate possible stimulus pathways implied in the response of anoestrous ewes to the introduction of rams. The intensity of grey is related to the iortance of each pathway; the darker the grey the more important the pathway. LH. luteinising hormone.

of delayed ovulations (4-6 days after the introduction of rams). We have also observed some other ovarian responses, such as short luteal phases caused by anovulatory follicles, or luteal phases of normal length provoked by luteinised follicles (Ungerfeld *et al.* 2002). Short luteal phases from anovulatory follicles have been described previously in prepubertal lambs (Berardinelli *et al.* 1980) and at the beginning . of the breeding season in adult ewes (Ravindra and Rawlings 1997). Luteinised follicles secreting progesterone for several days have been previously reported as a response to the ram effect (Knight *et al.* 1981) and have also been observed during the transition to the breeding season (Bartlewski *et al.* 1998) and after administration of gonadotrophin-releasmg hormone (GnRH) to anoestrous ewes (Rubianes *et al.* 1997). Recently we observed short luteal phases as an outcome of delayed ovulations (Ungerfeld *et al.* 2004).

The increase in LH pulsatility coincides with an increase in the number of large follicles and with an increase in the diameter of the largest follicle (Ungerfeld 2003). This should be expected because terminal preovulatory follicular growth is dependent on LH pulsatility (Baird and McNeilly 1981). Atkinson and Williamson (1985) reported an increase, after the introduction of rams to anoestrous ewes, in the number of large follicles, coinciding with a rapid increase in LH pulsatility. We observed that ewes responding with a luteal phase and ewes not responding had similar increases in the diameter of the largest follicle, coinciding with an increase in LH pulsatility (Ungerfeld 2003). This suggests that the type of luteal phase that a ewe displays after responding to the ram effect is not related to the maximum diameter attained by the largest follicle present after rams are introduced.

The onset of the follicular wave in anoestrous ewes may be manipulated with a single dose of oestradiol-17|3. The emergence of the following wave is synchronised ~3 days later (Ungerfeld *et al.* 2004). In a recent experiment, we observed that administration of oestradiol-17|3 either 3 or 5 days before the introduction of rams changes the timing of oestrus among ewes after the introduction of rams (Ungerfeld 2003; Fig. 2). Although the total number of ewes in oestrus was similar, the onset of oestrus was earlier in treated ewes *(P* < 0.001). This suggests that, as happens after a GnRH challenge (Rubianes *et al.* 1997), the ovarian response is related to the growth status of the largest follicular present when rams are introduced. However, this hypothesis could not be confirmed by ultrasound observations (Ungerfeld *et al.* 2004).

In summary, although most studies report classic ovarian responses, different ovarian response patterns may also be present after the introduction of rams.

Progestogen priming

The use of progestogen devices 12-14 days before the introduction of the rams to anoestrous ewes ensures that heat is displayed coincidently with the first ovulation, followed by a luteal phase of normal duration (Hunter *et al.* 1971).

As happens when oestrus is induced with equine chorionic gonadotrophin (eCG) (Ungerfeld and Rubianes 1999a, 2002), short-term (6-day) priming with sponges impregnated with commercial (60 mg) or lower doses (20 mg) of medroxyprogesterone acetate (MAP) is as effective for ram effect stimulation as traditional long-term (14-day) priming in terms of signs of oestrus, oestrus distribution and fertility (Ungerfeld *et al.* 2003). This is an important advantage and gives flexibility when working under field conditions. Used sponges are normally discarded, so decreasing the initial concentration in the sponges would be desirable to minimise environmental contamination with synthetic hormones. The results of using intravaginal devices containing progesterone, MAP or fluorogestone acetate (FGA) in 6-day priming are similar, with respect to the ram effect (Ungerfeld *et al.* 1999), as with oestrus synchronisation treatments (Walker *et al.* 1989; Ungerfeld and Rubianes 2002).

28 Time since the introduction of the rams (days)

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Fig. 2. Oestrous distribution of anestrous Corriedale ewes stimulated with the ram effect. Ewes remained untreated (a), or received 40 |xg of oestradiol-17P for 3 days *(b),* or 5 days (c) before rams were introduced.

Oestrous distribution and onset was significantly different between groups. Unpublished results.

It has also been observed that, in ewes in low body condition, progestogen priming increases the number of ewes in oestrus (Ungerfeld *et al.* 1999). However, it should be kept in mind that, as in cyclic ewes (Robinson *et al.* 1970), the fertility ofthe first oestrus after progestogen priming is lower than that ofthe first oestrus observed in unprimed ewes (Ungerfeld *et al.* 2003). The lower fertility after progestogen priming may be due to a failure in sperm transport (Pearce and Robinson 1985) and/or by the ovulation of an aged follicle as a consequence of low progestogen concentrations (for a review, see Rubianes and Menchaca 2003).

Administration of a single dose of 20 mg progesterone at the moment of introducing the rams prevents the occurrence of short luteal phases (Lindsay *et al.* 1984). A similar effect was also observed when progesterone was injected as early as 5 days before the introduction of the rams (Pearce *et al.* 1987). We recently observed that a single dose of MAP administered 3-5 days before the introduction of rams may substitute for a single dose of progesterone (Ungerfeld *et al.* 2003). Although there is a lack of information about the pharmacological characteristics of MAP (including its half-life), our observations agree with previous reports of a long half-life of the compound compared with progesterone (Shelton *et al.* 1967). Furthermore, when MAP was administered in the same way as progesterone (i.e. at the moment when rams were introduced) few ewes came into oestrus before Day 28 (Ungerfeld *et al.* 2003), but the number increased between Days 28 and 35 (Ungerfeld 2003). This suggests that the reproductive response was delayed, and not blocked, by MAP, another observation consistent with a long half-life.

For practical applications, fertility may be lower than in the next oestrous period, but the use of progestogen priming allows us to advance oestrus by 15 days in an important percentage of ewes. A simple and cheap way to synchronise oestrus is to administer a single dose of progesterone or MAP before introduction ofthe rams. If MAP is used it should not be administered at the moment ofthe introduction ofthe rams: oestrus will be synchronised and begin 17-18 days later.

What makes a ewe respond or not respond to the introduction of rams?

The response of a ewe to the ram effect depends on the strength of the stimulus and the responsiveness of the ewe to the stimulus. There are ewes that will not respond, regardless ofthe strength ofthe stimulus (e.g. breeds with a strong seasonal pattern). Other ewes, by contrast, will respond to a very light stimulus (e.g. breeds with a light/shallow anoestrus close to the onset ofthe breeding season).

Factors associated with the stimulus

Initially, it was thought that the stimulus consisted only of a signal from rams to ewes. However, other social stimuli may be present simultaneously and may be necessary for obtaining a response in the ewes (Fig. 1). Oestrous ewes stimulate rams, in turn provoking them to strengthen the stimulus to anoe-strous ewes. Some experiments have shown that if there is close contact between them, oestrous ewes may stimulate ovulation in anoestrous ewes (Zarco *et al.* 1995). However, this has not been observed under other conditions and in other breeds of sheep (Knight 1985). In goats, dominant does have been reported to respond earlier to the introduction of bucks than subordinate does (Alvarez *et al.* 2003). Although behavioural dominance is not as strong among ewes, this and other aspects of social interactions, such as whether dominant interactions between rams affect the final response to the ram effect, remain to be elucidated. Male-male competition reportedly fails to stimulate rams during the breeding season (Price *et al.* 2001), but it remains to be determined whether it may have a stimulating effect outside the breeding season, when rams might not spontaneously display their maximum activity.

Many studies have investigated the female responses to the ram effect, but few have determined the importance of different ram characteristics in evoking those responses. In goats, the reproductive condition ofthe buck appears to be the limiting factor determining the response of anoestrous does to the male effect (Flores *et al.* 2000). The ram stimulates ewes through pheromones and visual, behavioural or tactile cues that act in a synergistic way. Several experiments have provided different, and sometimes contradictory, results with regard to the importance ofthe different signals (Knight and Lynch 1980a; Pearce and Oldham 1988; Cohen-Tannoudji *et al.* 1989), but this may be a consequence of using ewes in different states of responsiveness.

The role of pheromones

According to some reports, full contact is not necessary for ewes to respond to rams (Watson and Radford 1960). The scent of wool and wax from intact rams can be enough to trigger a response in terms of ovulation in ewes (Knight and Lynch 1980a). However, information about the importance of scent is contradictory. Morgan *et al.* (1972) observed that ewes with impaired smell did not respond to rams, but elsewhere a normal LH response was observed in ewes reported as being without vomeronasal (Cohen-Tannoudji *et al.* 1989) or olfactory (Cohen-Tannoudji *et al.* 1986) activity. Wool and wax have been reported to be the main source of the pheromones that are part ofthe ram effect (Knight and Lynch 1980a). Pheromones produced by the goat buck can also stimulate LH pulse frequency (Over *et al.* 1990) and ovulation (Knight *et al.* 1983) in anoestrous ewes, although bucks are less effective than rams (McMillan 1987). The pheromones secreted by the boar pig appear to be ineffective in ewes (Knight *etal.* 1983).

It has been demonstrated that pheromone production in rams is controlled by androgens (Fulkerson *et al.* 1981; Croker *et al.* 1982; Signoret *et al.* 1982). The pheromones are present in aqueous and petroleum-spirit extracts of wool and wax (Knight and Lynch 1980ft). They are produced by the skin, especially around the eyes. Cohen-Tannoudji *et al.* (1994) identified some of the components present in wool through a bioassay that measured the LH response of ewes. Using extracts from fleece and from the ante-orbital gland of rams, they established that several compounds are needed to obtain a maximum reproductive response in ewes. The acid fraction from the extract (without compound identification) plus a combination of 1,2-diols are mostly responsible for the pheromone component of the ram effect. The use of pheromones alone in anoestrous ewes has given controversial results: in one study pheromones did not induce any changes in LH or FSH secretion (Schneider and Rehbock 2003), but in others they resulted in ovulation (KaulfulB *et al.* 1997', 2002) or in an increase in pregnancy rates of inseminated ewes (Milovanov 1991).

Other stimulatory signals

Pearce and Oldham (1988) stimulated ewes with masks containing ram's wool (and therefore probably pheromones), but obtained the maximum reproductive response only with full contact between the rams and ewes, which suggests that behavioural and/or tactile signals are also needed in ewes in a deeper anoestrous state. Moreover, some authors have suggested that other sensory signals may completely replace the pheromone stimulus (Cohen-Tannoudji *etal.* 1986,1989). Perkins and Fitzgerald (1994) demonstrated the importance of the sexual behaviour of rams. A higher number of ewes ovulated when put together with rams expressing high libido compared with ewes put together with rams expressing low libido, although the testosterone levels of the rams appeared to be similar.

Breed and percentage of rams in the flock

There is little information about using rams from different breeds; most experiments have been performed in Dorset and Romney rams (Table 1). Regarding the percentage of rams in the flock, Lindsay *et al.* (1992) observed more ewes in oestrus when they used 3% or 6% of rams than with 1% of rams in the flock. Rodriguez Iglesias *et al.* (1997), by contrast, did not obtain a higher percentage of ewes in oestrus when they increased the percentage of rams from 8% to 16%.

Presence of ewes in oestrus

When rams are used as teasers, other social interactions are also involved and, in most published experiments, it is impossible to discriminate between components that are part of the ram effect and components that are not. Ewes in oestrus also influence the reproductive activity of rams, mainly by inducing an increase in LH pulses and testosterone levels during the first 4-8 h of contact (Yarney and Sanford 1983; Gonzalez *etal.* 1991; Fig. 1). Stimulation by oestrous females of anoestrous females has been widely demonstrated in goats (Restall

Table 1. Effectiveness of rams from different breeds as teasers to induce oestrus in anoestrous ewes

Ram breed		Reference
More effective	Less effective	
Dorset	Suffolk	Nugent *etal.* 1988
Dorset	Romney	Meyer 1979
Knight and Lynch 19806		
Tervit and Peterson 1978		
Tervit *etal.* 1977		
Knighted/. 1980		
Dorset	Romney X Finn	Meyer 1979
Poll Dorset	Coopworth	Scott and Johnstone 1994

et al. 1995) and some experiments suggest a similar effect in gilts (Pranier and Mounier 1991), cattle (Wright *et al.* 1994) and ewes (Oldham 1980). However, in Suffolk and Dorset ewes a luteal response of anoestrous ewes to oestrous ewes has been observed only where close contact between them allows considerable interaction (Zarco *etal.* 1995). In experiments with other breeds (e.g. Romney sheep) the presence of oestrous ewes as such did not induce ovulation in anoestrous ewes (Knight 1985).

The proportion of anoestrous ewes from some breeds (Romney, Corriedale) that ovulate after the introduction of rams has been reported to increase when oestrous ewes are introduced at the same time as rams in a process termed 'social facilitation' (Knight 1985; Rodriguez Iglesias *et al.* 1991). A similar response has been reported in rams that had been in contact with oestrous ewes for a short period before they were joined with ewes in anoestrus (Knight 1985). Rams that have been isolated from ewes and then placed with ewes in oestrus are more effective in stimulating ovulation in anoestrous ewes than are rams that have been in contact with ewes long before the procedure takes place (Knight *et al.* 1998). The increased response obtained with stimulated rams may be mediated by increase of their testosterone levels (Knight *etal.* 1998).

Rodriguez Iglesias *et al.* (1991) suggested that the continuous presence of oestrous ewes is important because it provides visual cues to anoestrous ewes of rams displaying sexual behaviour. The number of ewes that ovulate increases with increasing sexual performance (Perkins and Fitzgerald 1994) and testosterone levels in rams (Rosa *etal.* 2000). We recently observed that rams respond to the introduction of oestrous ewes with rapid increases in LH and testosterone levels and that the concentrations of both hormones remain high for several days, whereas the ewes that are in contact with the rams are in oestrus (Ungerfeld and Silva 2004). The high proportion of anoestrous ewes that ovulate when oestrous ewes are present may in part be explained by increasing testosterone levels of the rams. The increase in testosterone concentrations may stimulate pheromone production and perhaps libido and this may be part of the mechanism of social facilitation.

Other factors associated with rams

Walkden-Brown *et al.* (1993) found that bucks that had a high level of nutrition were more effective in stimulating anoestrous does to ovulate than were bucks with a low level of nutrition. However, in rams, nutritional status does not seem to influence serving capacity or the ability to induce ovulation, at least not with regard to the nutritional differences evaluated by Fisher *et al.* (1994).

Factors associated with receptivity

Depth of anoestrus

Seasonal anoestrus is associated with a decrease in LH pulsatility (for a review, see Gallegos-Sanchez *et al.* 1998) and an absence of preovulatory surges of FSH and LH. The low LH pulsatility is due to two inhibitory mechanisms: (7) an increased negative feedback effect of oestradiol on the hypothalamus; and (2) a direct effect of photoperiod on the hypothalamo-hypophyseal system controlling LH secretion (Goodman and Karsch 1981).

Thomas and colleagues (1984) observed that some breeds are less sensitive to negative feedback of oestradiol than others. With ram effect stimulation, more ewes from a less seasonal breed (e.g. Dorset) than from a more seasonal breed (e.g. Hampshire) have been reported to ovulate and conceive (Nugent *et al.* 1988). However, ewes from more seasonal breeds may not necessarily respond to the ram effect by ovulating, even if they display an increase in LH pulsatility (Minton *et al.* 1991).

There is little information about the physiological mechanisms that determine whether a ewe will respond to the ram effect or not. Although more work is needed to determine what' anoestrus depth' means, LH pulsatility has been consistently proposed as one parameter by which to assess it (Martin *et al.* 1985; Ungerfeld 2003; Table 2). Also, we observed that FSH concentrations before the introduction of rams are significantly higher in ewes that subsequently show a luteal phase than in those that do not (Fig. 3). We also observed that anoestrous progestogen-primed ewes, which responded to the ram effect with ovulation plus oestrus, had higher FSH and oestradiol-17(3 concentrations before the introduction of rams than did ewes that had an ovulation without oestrus or that did not ovulate (Ungerfeld 2003). However, specific values from any hormone that may be used to characterise the depth of anoestrous of an individual female should be considered only against data from a specific flock, because basal hormone concentrations may differ with factors such as breed, the stage of the anoestrous season, or the nutritional status of the animals.

It has been reported that the percentage of ewes that respond to the ram effect is related to the percentage in the flock that are ovulating spontaneously (Lindsay and Signoret 1980) although other observations could not confirm this relation (Latimori 1987). The response is also related to the stage of the anoestrous season: ewes are more receptive to the ram stimulus when rams are introduced close to the spontaneous onset of the breeding season (Oldham *et al.* 1984; Cushwae?a/. 1992).

Table 2. Luteinising hormone (LH) pulses per 8h in ewes that respond and ewes that do not respond to the ram effect

Time of joining	Pulse numbed		Pulse number[6]	
	OE	NOE	LP	NLP
Before	1.4	1.0	1.2 ±0.3	0.0 ±0.0
After	4.3	2.3	3.1 ±0.9	2.0 ±0.7

[A]Romanov and [B]Corriedale ewes were stimulated during the mid-non breeding season; whereas Romanov ewes were stimulated only with rams, Corriedale ewes were stimulated with rams and oestrous ewes. OE = Ovulating ewes (n = 7); NOE = non-ovulating ewes (n=4); LP = ewes responding with a luteal phase *(n* — 7); NLP = ewes without a luteal phase *(n — A).* Recalculated from Martin *et al.* (1985). [B]Ungerfeld (2003).

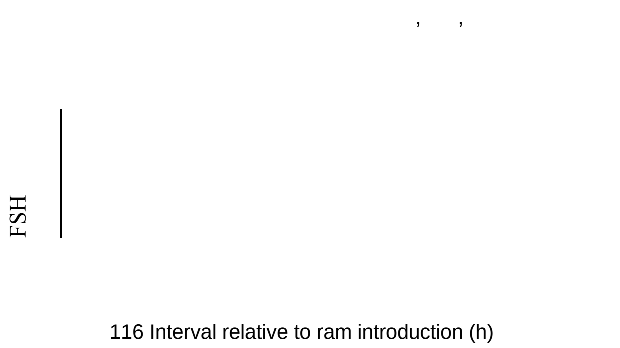

Fig. 3. Concentrations of follicle-stimulating hormone (FSH; mean ± s.e.m.) relative to ram introduction (arrow). Eleven Corriedale ewes were stimulated during the mid-non breeding season with rams and oestrous ewes. Ewes were classified as with (-A-; *n* — 7) or without (-■-; *n* = 4) luteal phases after the introduction of the rams. Profiles of FSH concentrations in ewes responding with luteal phases were significantly higher in both periods, before and after the introduction of the rams (reproduced from Ungerfeld 2003).

Some investigators have administered melatonin - either by implants, with food, or by daily injections - to ewes to reduce the depth of anoestrus before introducing the rams. In most trials the result was an increase in lambing rate (Rekik *et al.* 1991; Croker *et al.* 1992; Kusakari and Ohara 1996). In some trials the response depended on the breed of the ewe (Gomez Brunet *et al.* 1995). An increase in the proportion of ewes coming into oestrus (Kaya *et al.* 1998), an earlier conception (Croker *et al.* 1992) or an increase in the conception rate (Gomez Brunet *et al.* 1995; Kusakari and Ohara 1996) were also observed in some experiments.

In summary, two aspects of the phenomenon should be considered carefully when the ram effect is applied. First, the rams that are to be used should be selected with respect to more than just their reproductive history. Likewise, the characteristics of the ewes - breed, nutritional status, interval since last lactation, etc - should be taken into account if we expect to obtain a good lambing percentage.

The ram effect in other phases of ewe reproduction

The ram effect has also been used successfully to induce cyclic activity in prepubertal lambs and post-partum ewes. Some experiments have also been performed to determine possible applications of the ram effect in cyclic ewes.

Puberty

The introduction of rams to prepubertal lambs during the non-breeding season has been reported to result in an increase in LH pulsatility; however, ovulation has been said to occur only when rams are introduced shortly before the onset of the breeding season (Hanrahan and O'Riordan 1990; Al-Mauly *et al.* 1991). The percentage of 7- to 10-month-old lambs ovulating ranged from 30% to 60% depending on breed and season (Oldham and Gray 1984). In 14-month-old Corriedale lambs that had not yet been cyclic during the previous breeding season, we recently observed that more than 60% came into oestrus after the introduction of rams halfway through the non-breeding season (R. Ungerfeld *et al,* unpublished observations). Dyrmundsson and Lees (1972) observed that the mtroduction of rams to lambs during the transition period from the non-breeding season to the breeding season did not affect the time of onset of mating activity, but did give a better synchronisation of receptivity. Garcia and Perez (1999) and Murtagh and colleagues (1984) reported that the percentage of lambs that responded to the introduction of rams increased in lambs preconditioned by previous exposure to rams.

Post-partum period

There is little information about the use of the ram effect to induce oestrus during the post-partum period. However, there are studies showing that the interval from parturition to conception may be reduced if rams are introduced to post-partum ewes in autumn (Wright *et al.* 1989) and spring (Ungerfeld *et al.* 2001). Introduction of rams appears to have no effect on uterine involution of the ewe (Godfrey *et al.* 1998).

In autumn-lambing Merino ewes it was observed that the percentage of ewes ovulating during the first 4 days after the introduction of the rams increases progressively from 21 to 45 days post-partum (Geytenbeek *et al.* 1984). However, the overall proportion of ewes that exhibited oestrus at Day 82 post partum was similar despite the post-partum time (14 to 45 days), showing that the final reproductive result is unrelated to the moment when the rams are introduced during this post-partum period.

In one study performed during the non-breeding season, post-partum Corriedale ewes and ewes that had lambed several months previously responded equally to the ram effect, with a similar number of ewes in oestrus (Table 3). However,

Table 3. Frequency of ewes in oestrus (OE) and conception rates (CR; pregnant ewes/oestrous ewes) on Days 1 to 5 and Days 17 to 30 (periods during which ewes came into oestrus)
On Day 0 (mid non-breeding season), rams were introduced to previously isolated suckling post-partum Corriedale ewes (PP) and to Corriedale ewes whose lambs had been weaned at least 6 months before the onset of the experiment (C)

	Days 1-5		Days 17-30		Total	
	OE (%)	CR (%)	OE (%)	CR (%)	OE (%)	CR (%)
PP	15/75 (20.0)	0/15 (Of	49/75 (65.3)	29/49 (59.2)	64/75 (85.3)	29/64 (45.3)[c]
C	12/59 (20.3)	5/12 (41.7)[b]	43/59 (72.9)	30/43 (69.8)	55/59 (93.2)	35/55 (63.6)[d]

[a] v. [b]: $P< 0.01$; [z] $v.$ [A]: $P< 0.05$.

Table 4. Frequency of ewes in oestrus and conception rates (pregnant ewes/oestrous ewes) of post-partum suckling ewes stimulated with the ram effect
During the mid non-breeding season rams were introduced to previously isolated Corriedale ewes that had lambed 5, 6, 7 or 8 weeks before

Time post-partum (weeks)	OE (%)	CR (%)
5	7/14 (50.0)	0/7 (0)
6	10/24(41.7)	1/10(10)
7	11/24(45.8)	4/11(36.4)
8	5/11 (45.5)	2/5 (40)

OE, ewes in oestrus; CR, conception rates.

in agreement with Wright *et al.* (1990), the conception rate was lower in the post-partum ewes, probably as a consequence of suckling and low body condition score.

The response of ewes to the ram effect during the postpartum period is time-dependent. In ewes that had lambed during the non-breeding season, Khaldi (1984) observed that the percentage of ewes that ovulated after the introduction of rams was higher at 75 days than at 15, 30, 45 or 60 days after parturition. We studied suckling Corriedale ewes and did not find significant differences in oestrous response after introducing the rams at 5, 6, 7 or 8 weeks after parturition (Table 4). However, conception rates in ewes stimulated 7-8 weeks after lambing were higher than in ewes stimulated at 5-6 weeks (37.5% v. 5.9%; $P< 0.05$) (L. Silva and R. Ungerfeld, unpublished observations).

Cappai *et al.* (1984) reported that the response of postpartum Sarda ewes to the ram effect was related to the milk yield. A high milk yield reduced ovulation rate and delayed the LH surge. Prolactin concentrations, which are high in lactating ewes (Gomez Brunet and Lopez Sebastian 1991), increase the negative feedback of oestrogens on tonic LH secretion (Kann *et al.* 1976). However, similar to findings in rebreeding of post-partum cattle (Williams and Ray 1980), Poindron and colleagues (1980) observed that prolactin secretion was not related to the response of post-partum ewes to the ram effect.

Cyclic ewes

It has been observed that permanent contact of cyclic ewes with rams increases the length of the breeding season by inducing both an earlier onset (Eldon 1993; O'Callaghan *et al.* 1994) and a later end (O'Callaghan *et al.* 1994) of the season.

Pearce and Oldham (1983), using ovariectomised, progesterone-treated ewes during the breeding season, observed an increase in LH pulse frequency after ram introduction, which was not affected by progesterone treatment. Ngere and Dzakuma (1975) observed a marked synchrony of oestrus among cyclic ewes after the introduction of rams. Cyclic ewes with an intravaginal sponge containing MAP also respond to ram introduction, with an increase in LH secretion (A.C.O. Evans, personal communication). Similarly, in cyclic ewes stimulated with rams, an earlier lambing period has been observed (Ann Lai 1988), as has an advance in the onset of oestrus, the LH surge and ovulation (Evans *et al.* 2002). Lucidi *et al.* (2001) recently reported an increase in pregnancy rate when rams were introduced to cyclic ewes. We observed that the ram effect, when used in combination with traditional synchronisation treatments at the beginning of the breeding season, shortens the interval to oestrus and improves synchronisation (Ungerfeld and Rubianes 1999ft). However, we could not obtain a similar response when the stimulus was provided in the mid-breeding season, which is when ewes spontaneously display their maximum reproductive activity (Ungerfeld 2003).

Although few experiments have been performed in cyclic ewes, results suggest that the effect of ram introduction should be studied to determine possible advantages of its incorporation in reproductive management of these ewes.

Some practical considerations

The ram effect has been widely used throughout the world (Martin *et al.* 1986) as an easy and cheap management tool that may provide good results in productive systems. However, unless the following factors are taken into consideration before using it, the ram effect may be incorrectly used and completely ineffective:

- Ewes should be completely isolated from rams. Although some authors report shorter periods as being sufficient, or even no isolation when rams are changed (Pearce and Oldham 1988), to ensure that it will work under all conditions (breed, landscape conditions, etc) ewes should be isolated for 1 month.
- Isolation means that ewes cannot see, hear or smell rams during that period. A minimum distance of 1 km between rams and ewes is recommended. Ideally ewes should be kept in a pen with no males near them. Also, if the pen is near the boundaries of the farm, the neighbours' pen should not contain males during this period.
- Rams and ewes should be examined before beginning the period of isolation. Examining them during the isolation period may, depending on the conditions of the farm, result in rams and ewes being nearer than the minimum recommended distance, particularly if the examination entails moving the animals from their pen.
- Breeding soundness of rams should be examined at least 45-60 days before they are used and should include both a physical test and a test of the ram's reproductive soundness. Rams should be selected only after performance of a physical evaluation of their feet and legs, body condition, vision and any defect that might impair a ram's ability to breed. Also, the scrotum and testicles should be measured and palpated, and the penis should be physically examined. The body condition of the ewes should be examined early enough, especially if nutritional management (i.e. flushing) is planned.
- If there are doubts about the stimulus that may be obtained with the rams to be used, it is recommended to incorporate ewes in oestrus in the procedure. These ewes may be hormonally induced (progestogen + oestrogens or eCG) or may come from a less seasonal breed that is cyclic during that period of the year.

Future considerations

The recent increase in the sheep population in undeveloped countries (FAO 2003) demands an increase in knowledge of the mechanisms that control reproduction and the use of scientific information in management. Although experiments on the ram effect have been performed in more than 45 breeds (Ungerfeld 2003), more than half of this information was obtained in Merino and Romney ewes. This is related to the fact that more than 40% of the published papers come from Australia and New Zealand, where these breeds predominate. Although more than 70% of the world's sheep are farmed in Latin America, Africa and Asia, only 33% of the information was obtained in countries from these continents. Little is known about possible differences in the response of other breeds and about whether the breed or the strength of its seasonal pattern influences, not only the percentage, but also the characteristics of the response to the introduction of rams.

Another issue, which has not been well addressed during recent years, is how insensitivity to the stimulus can be managed. Which desirable characteristics in rams are used to stimulate ewes? If we can manage the strength of the stimulus through selection of rams or through the way we manage them, can we obtain ovulations in more seasonal breeds?

The ram effect, as a cheap and easy technique, may be included in management strategies, allowing farmers to obtain out-of-season lambing, advance puberty onset or develop accelerated reproductive programmes. However, more research should be done to determine how to obtain the best results from the ram effect.

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