JOURNAL OF VETERINARY ANDROLOGY ISSN 2542-3045

Review Article/Artículo de Revisión

MALE EFFECT: SUSTAINABILITY AND EFFECTIVENESS IN INDUCING ESTRUS IN GOATS

EFECTO MACHO: SOSTENIBILIDAD Y EFICIENCIA EN LA INDUCCION DEL CELO EN CABRAS

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ABSTRACT

The male effect is a strategy used in reproductive management of herds for the induction and synchronization of estrus, consisting of the reintroduction of males into a group of previously separated females. This interaction induces an increase in LH pulses followed by ovulation. In all species, there are communication mechanisms, many involving the use of chemoreceptor organs that enable the perception of pheromones, mediators in intraspecies interaction related to recognition for mating. Synchronization of female estrus by the male effect has many advantages reported by several authors, such as the reduction of costs, the absence of undesirable immune response by the use of chorionic gonadotropin, the decrease of hormonal residues in treated females, thus complying with ecological and sustainable production principles in animal production.

Keywords: biostimulation, ovulation, estrus, artificial insemination
JOURNAL OF VETERINARY ANDROLOGY (2016) 1(1):13-23

RESUMEN

El efecto macho es una estrategia usada en el manejo reproductivo de los rebaños para la inducción y sincronización del celo, consistiendo en la reintroducción de machos en un grupo de hembras previamente separadas. Esta interacción induce un incremento en los pulsos de LH seguidos por la ovulación. En todas las especies, existen unos mecanismos de comunicación, algunos involucran el uso de órganos quimiorreceptores que permiten la percepción de feromonas, mediadores en las interacciones intraespecíes relacionados con el reconocimiento para el apareamiento. La sincronización del celo en las hembras por el efecto macho tiene varias ventajas reportadas por diversos autores, tales como la reducción de costos, la ausencia de una respuesta inmune indeseada por el uso de gonadotropina coriónica, la disminución de residuos hormonales en las hembras tratadas, cumpliendo con los principios de una producción ecológica y sostenible en la producción animal.

Palabras clave: bioestimulación, ovulación, estro, inseminación artificial
JOURNAL OF VETERINARY ANDROLOGY (2016) 1(1):13-23

Received/Recibido: 23/06/2016; Accepted/Aceptado: 13/07/2106

INTRODUCCIÓN

Social relationships between animals of the same species, or even between different species, are able to affect the reproductive activity, either between males, females, or between males and females (Rosa & Bryant, 2002; Sampaio et al., 2012). In regions where the photoperiod has regulatory function on the reproductive activity of animals, socio-sexual interactions are able to change such situation, that is, reverse the condition of reproductive seasonality (Delgadillo et al., 2012).

The male effect is characterized as a sexual biostimulation promoted by multisensory contact (visual, olfactory, tactile and auditory) between males and females, able to induce and synchronize sexual activity of females, even in periods of anestrus, presenting a more efficient response when there is direct contact between the sexes. The olfactory factor is the main means of response to the male effect, but when comparing results of research involving only the olfactory pathway and the complete biostimulation, it is assumed then that, for a greater effectiveness of the male effect, it should be preserved the action of all the senses, visual, tactile, auditory, and olfactory (Walkden-Brown et al., 1993).

The introduction of rams or bucks into an effective flock of ewes or does previously isolated from males, before the normal mating season, was initially described by Underwood et al. (1944). This biostimulation is a low cost technique with several adventages such as accelerating the onset of ovulation (Souza et al., 1995) and estrous behavior, reducing the age at first kidding (Álvarez & Andrade, 2008), preventing the presence of hormone residues (Contreras-Solis et al., 2009), promoting a short time interval between the beginning of the experimental work and the onset of ovarian activity and estrus (Flores et al., 2000), enhancing the response to induction and synchronization of estrus using reduced amounts of hormones (Mellado et al., 2000) and increasing fertility (Horta & Gonçalves, 2006).

This review aims to show the physiological and endocrine mechanisms of the efficient technique of induction and synchronization of estrus by biostimulation using the male effect, which can be used alone, or in combination with various types of protocols, in natural or artificial breeding of small ruminants, in different climatic zones of animal production.

PHEROMONES AND PHYSIOLOGICAL CHANGES CAUSED BY THE MALE EFFECT

The introduction of a sexually active breeder in a flock of goats is able to stimulate the sexual activity of females (Avdi et al., 2004), the presence causes changes in female physiology, and this acute response is called male effect (Rosa & Bryant, 2002), and seems to depend mainly on olfactory signals originating from pheromones produced by the males, through stimulation of androgens (Gelez & Fabre-Nys, 2004), in association with behavioral stimuli generated mainly during courtship activity (Rosa & Bryant, 2002).

The characteristic scent from the buck comes from the sebaceous glands, where it is produced, and is directly related to the concentration of testosterone (Iwata et al., 2000). These substances are detected by the olfactory receptor neurons (ORN), capable of detecting volatile odors, or vomeronasal receptor neurons (VRN), which detect pheromone molecules (Murata et al., 2009). The initial interaction between males and females occurs by action and recognition of pheromones, which are chemicals transported by air and excreted through feces or urine or by skin glands, perceived by the olfactory system, and capable of causing behavioral and endocrine alterations (Rekwot et al., 2001), as part of a chemical communication (Castañeda et al., 2007). In the male, during the detection of olfactory signals, chemical communication is evidenced by the Flehmen response. It is through these pheromone stimuli, the sight of the male and the physical contact with the male that promote the restoration of hormonal activity in the female, with consequent manifestation of estrus.

The male effect is capable of altering the female physiology (Abi Salloum & Claus, 2005). The presence of the male is perceived through the recognition of pheromones by the olfactory system, which transmits a signal to the medial nucleus of the amygdala, and then is relayed to the hypothalamus, which is the generator of GnRH pulses (Figure 1; Murata et al., 2009). Consequently, there is an increase in the frequency and amplitude of pulsatile LH secretion, increasing from 2 to 3 times the number of pulses (Chemineau et al., 1986; Alvarez et al., 2009). Thus, exposure of females to sexually active males promotes a rapid activation of LH secretion and a reduction in negative estradiol feedback on the pituitary hypothalamic axis, culminating in the preovulatory LH surge (Signoret 1980), generating an acute response (short-term response) followed by a chronic response (response long-term), when there is continuity of this interaction (Chanvallon et al., 2010). Sakamoto et al. (2013) suggest that the main target of pheromones, in goats, are the kisspeptin/neurokinin B neurons found in the arcuate nucleus.

Murata et al. (2014) identified a volatile molecule, 4-ethyloctanal, as the pheromone of the buck responsible for the “male effect” in goat reproduction. This was the first report of the identification of a pheromone in mammals with clear evidence of the neurophysiological action in the central regulation of reproduction.

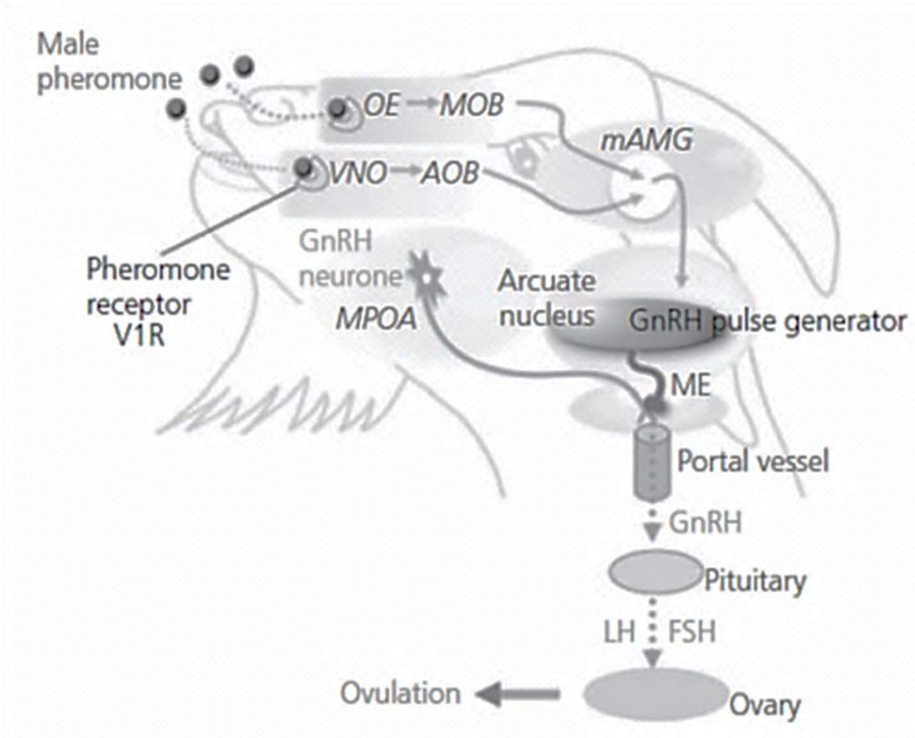

Figure 1. Perception-action mechanism of pheromones of the buck on the hypothalamic-pituitary-gonadal axis of does. OE: olfactory epithelium; MOB: main olfactory bulb; VNO: vomeronasal organ; AOB: auxiliary olfactory bulb; mAMG: medial amygdala. (Adapted from Murata et al., 2009).

MALE EFFECT AND INCREASED FREQUENCY OF GNRH/LH PULSES

In sheep and goats, exposure of females to sexually active males during anestrous periods results in the activation and secretion of luteinizing hormone (LH), the substance responsible for ovulation (Gelez & Fabre-Nys, 2004). This rapid response, generating a higher frequency of LH pulses and concentration, increases significantly after exposure to male and tends to increase progressively over time (Martin et al., 1986; Alvarez et al., 2009). This increase in LH concentration seems to be widespread, thus, it is verified an increase in all respects, either at the basal level or in the medium and maximum concentration of progesterone (Claus et al., 1990; Chanvallon et al., 2010). Bucks are capable of inducing estrus in cyclic females, but this factor is regulated by progesterone, thus does in either early or late luteal phase are susceptible to these alterations, but in females which have progesterone plasma levels higher than 5 ng/mL, the male effect may have a low efficacy. This fact is different in ewes, as these animals for having a shorter estrous cycle have insufficient levels of progesterone to block the male effect (Hawken et al., 2009). Therefore, endocrine action in response to the male differs according to the stage of the estrous cycle of the female, and the level of progesterone is a limiting factor to change the endocrine pattern of ewes.

After the introduction of male, follicle growth is pronounced, reaching 1.5 mm/day, and this development is higher for small follicles, which evidences the male role in ovulation induction (Delgadillo et al., 2011). According to Alvarez et al. (2009), does subjected to the male effect showed LH surge from 80 to 317 minutes, with 5.7 to 8 pulses within 39 hours and ovulation between 8 to 10 days after the introduction of the male. The first estrus induced by the male presence may be accompanied by ovulation. The ovulation rate of estrus induced by the male effect is similar (1.8 ± 0.2, 2.0 ± 0.9) (Delgadillo et al., 2011) for the 1st and 2nd estrus, respectively. Thus, a female naturally or artificially inseminated at the first estrus can become fertile.

With the introduction of the male, there is a progressive hormonal adjustment, promoting stabilization of physiology and adequate ovarian functionality. Thus, it is commonly observed short estrous cycles at the beginning of biostimulation (Chemineau et al., 1986; Sutherland, 1987). These short cycles depend on the stage of development in which the follicle is at the time of introduction of the male (Gonzalez-Bulnes et al., 2006). This physiological regulation means that females are more likely to conceive on the next estrus, when compared to the physiological state existing at the first estrus, since the ovarian function is re-established to normality (Mellado et al., 1994). Flores et al. (2000) achieved a pregnancy rate of 95% at the second estrus, and only 10% at the first estrus.

FACTORS AFFECTING THE RESPONSE TO THE MALE EFFECT

The response to the male effect as for the velocity, synchrony intensity and percentage of ovulating females, is dependent on environmental, social, physiological and genetic factors (Walkden-Brown 1993; Whitley and Jackson 2004; Alvarez et al., 2009), which are related to males or females. It is noteworthy the physiological state of the male (Véliz et al., 2006), body condition (Mellado et al., 1994), phase of the estrous cycle (Hawken et al., 2009) and sexual experience (Gelez & Fabre-Nys, 2004; Horta & Cavaco Gonçalves, 2006).

The sexual status of the male is extremely important to obtain a favorable result when working with biostimulation, because it performs sexual behaviors more frequently (Véliz et al., 2002), which favors the quality of the stimulus, an important factor for a successful male effect (Véliz et al., 2006a). The previous sexual experience is a social factor widely discussed among researchers. According to Bench et al. (2001), prior contact of males with females can provide experience to the male regarding copulation or can help in disinhibition of inexperienced males. Sexually active bucks are capable of mitigating various negative effects related to the effectiveness of biostimulation, either in experimental or field conditions (Avdi et al., 2004). Flores et al. (2000) observed that bucks treated with melatonin were able to induce estrus in seasonally anestrous does (100% estrus females with ovulation) in a short period of time with increased intensity of sexual behavior. Martínez-Alfaro et al. (2014) confirmed the importance of social interaction between males and females after observing that sexually active bucks, when sedated, displayed fewer reproductive behaviors, which resulted in ovulatory failure in females. Véliz et al. (2002) studied the combination of sexually active males with introduction of estrous females at the start of treatment, and observed an efficient response to the male effect (94.7% estrous females in estrus) and concentrated induction, because 79% does demonstrated estrus in the first three days.

Nutrition and health of animals involved in biostimulation by the male effect are of fundamental importance with direct influence on the results (Scaramuzzi & Martin, 2008). Body weight is directly related to reproductive response of the female when subjected to the male effect (Rivas-Muñoz et al., 2010), even when using sexually active males (Véliz et al., 2006b); this limiting factor continues to have negative effect on the response to the male effect, resulting in abnormalities related to estrous cycle, ovulation rate, in addition to increasing the number of does showing anovulatory estrus (Santiago-Miramontes et al., 2009). Véliz et al. (2006b) found that Mexican goats with body weight higher than 34 kg showed 83% estrus in the first five days in contact with males, while females weighing less than 34 kg, showed only 47% ($P < 0.006$). In turn, Mellado et al. (1994) observed that, in extensive conditions, does with good body condition score (BCS: 7 on a 1-9 range), when compared to the low score (BCS: 2, 1-9 range), tend to respond to the male effect earlier. Fitz-Rodríguez et al. (2009) evaluated the response of female goats on pasture to the male effect and reported that supplementation for seven days, starting at the introduction of males, resulted in a greater number of ovulating females, whereas supplementation for 14 days resulted in a higher number of pregnant goats. Thus, animals selected for the implementation of the male effect must have optimal body weight, related to parameters, such as breed, physiology, age, among others.

The experience of the female which will be submitted to the male effect can be a regulatory factor in the response to the male effect (Gelez & Fabre-Nys, 2006; Véliz et al., 2009). Chanvallon et al. (2010) found that multiparous goats had shorter latency between the introduction of the male and onset of physiological changes, when compared to nulliparous females, which showed physiological change 45 min after the multiparous. However, the latency between the introduction of the male and LH surge was not affected by the female experience. In turn, Walkden-Brown et al. (1993) reported that when exposed to buck fleece, multiparous does showed better response to treatment than nulliparous females. However, Fernández et al. (2011) and Sampaio et al. (2012) verified no physiological changes (alterations in LH), neither on the fertility of nulliparous compared to multiparous goats after the introduction of the male. Previous experience of does to the male effect can result in a greater ability to recognize and direct the system from the moment the females realize the change in the environment through the male introduction, and thus they may be able to respond to the male effect with greater intensity and promptness.

The social relations existing in a flock also act assiduously in response to biostimulation, since there is a positive relationship between the success rate, which ranks animals socially (Mendl et al., 1992), and the association index, concerning the interaction between the female and the male (Martin & Bateson, 2009). Alvarez et al. (2003) reported that the greater the intensity of contact, the more efficient the male effect, and, moreover, observed that among ovulating females, the number of days between the onset of the male effect and ovulation, as well as the interval between the introduction of males and conception, was lower for dominant than for submissive does. This fact is in line with Flores et al. (2000), who verified a relationship between the number of sexual behavior and high estrus induction index. In Cashmere female goats, Alvarez et al. (2007) stated that social categories assume importance in the estrus manifestation, and dominant females expressed more quickly estrous behavior in relation to subordinate females and that, in the study, the weight difference was not relevant to the displayed behavior.

The temperament of females, the influence of emotional reactivity on the reproductive success of sheep began to be investigated over the past two decades by Blache & Bickell (2010), who suggested that the temperament is one of the requirements in selection of animals. In Merino sheep, Lima (2006) noted that females with calm temperament responded better to biostimulation by ram than nervous ewes. Silva et al. (2012) consider that the observation of this parameter influences the responses of reproductive behavior.

EFFICIENCY OF MALE EFFECT ON INDUCTION AND SYNCHRONIZATION OF ESTRUS

The male effect has been used in various situations, either during the period of seasonal anestrus or in the breeding season. The sole and exclusive use of male effect, when performed correctly and seeking to minimize the various factors that act negatively on the response, is effective and reaffirms its power in the induction and synchronization of estrus, without the use of exogenous hormones (Mellado et al., 2000). Salles et al. (2008) and Salles et al. (2010) used the male effect after successive years in Saanen goats, reared in the tropical climate of northeastern Brazil, and reported increasing values over the years, and over 80% kidding and 1.7 prolificacy. Results of male effect efficiency can be seen in Table 1, which highlights the region in which it was performed and fertility rates obtained by different researchers.

Besides the exclusive use, the male effect can be used combined with other methods, thereby increasing the efficiency of treatment, inasmuch as the biostimulation is capable of potentiating the protocol response (Mellado et al., 2000). Demonstrated by Celi et al. (2013), the use of melatonin associated with the male effect, resulted in a higher fertility (91.7%), and higher percentage of females demonstrating estrus with ovulation (95.8%) during the winter solstice in temperate climate. The association between the application of PGF2α and the male effect, resulted in 100% estrus, reduced the time interval between the application of hormone and the time of ovulation (49.5 ± 3.4), significant result demonstrated by Contreras-Solis et al. (2009).

Synchronization protocols using progesterone (intramuscularly) and the male effect have been effective and relevant. Studying Murciano-Granadina goats, Díaz Delfa et al. (2002) used the male effect combined with progesterone or alone, and found similar rates of estrus induction (87.3 vs 79.6%, respectively), but achieved greater synchronicity and prolificacy with the use of progesterone. This greater synchronization is confirmed by Véliz et al. (2009), who observed that does treated with progesterone showed higher percentage of estrus in the first five days, and only one peak estrus, unlike the non-treated goats, which presented two peak estrus.

By subjecting prepubertal goats to estrus induction and synchronization protocols, Mellado et al. (2000), reported that the male effect is capable of amplifying the response to treatment, when compared the protocols Male Effect, SMB® (Synchro Mate B®) + PMSG and SMB®+PMSG+EM (16.6%, 66.6% vs. 91.6% estrus, respectively). Maia Júnior et al. (2009) observed that when synchronizing estrus of dairy goats with CIDR® using the male effect replacing eCG, reached values of 90% estrus, 61.1% pregnancy and 1.7 prolificacy. These values were similar to those obtained using CIDR® + eCG. In turn, Rodrigues et al. (2015) replaced the eCG with the male effect in protocols reusing CIDR® (1st or 2nd reuse) in dairy goats, and reported no decrease in the protocol effectiveness, either nulliparous or multiparous goat.

These findings indicate that the male effect, even when not used exclusively, either by experimental questions or inefficiency to the goal desired, can be employed along with other protocols, thus preventing the excessive use of exogenous hormones, which favors productive life and welfare of the animal, by the use of sustainably viable technology, which will result in a smaller amount of hormonal residues in animals and the environment, culminating in a lower environmental impact and more sustainable production.

Table 1. Efficiency of male effect in inducing estrus in dairy goats

Author	Local	Breed	Estrus	Pregnancy	Kidding	Prolificacy
Mellado et al., 2000	Mexico	Crossbred	92	83	-	1.8
Véliz et al. 2009	Mexico	Saanen	80	60	-	-
Rivas-Muñoz et al., 2010	Mexico	Local Goats	96	77	-	1.7
Salles et al., 2010	Brazil	Saanen	-	-	80	1.8
Vitaliano, 2011	Brazil	Saanen	100	95	80	1.8
Fernández et al., 2011	Mexico	Local Goats	97	85	60	1.4
Bedos et al., 2012	Mexico	Local Goats	92	72	64	1.7
Sampaio et al., 2012	Brazil	Saanen	47	67	40	-
Zarazaga et al., 2012	Spain	Mediterranean	100	81	-	1.4
Celi et al., 2013	Spain	Payoya	100	65	-	1.27
Loya-Carrera et al., 2014	Mexico	Local Goats	92	83	61	1.3
Gallego-Calvo et al., 2014	Spain	Blanca Andaluza	76	62	62	1.44
Ángel-García et al., 2015	Mexico	Crossbred	87	83	27	1.9

MALE EFFECT INTERSPECIES

The use of the male effect interspecies, i.e., the insertion of ram in a flock of does, or vice versa, is a proven technique for the induction and synchronization of estrus (Vitaliano et al., 2012). This form of implementation is partly because of the similar composition of pheromones involved in the male effect in goats and sheep (Ichimaru et al., 2008). Knight et al. (1983) observed that the introduction of a buck in a flock of anoestrus ewes was able to induce estrus with efficiency similar to a ram. Over et al. (1990) observed that the scent of buck result in physiological changes (increase in LH secretion) in sheep.

Sampaio et al. (2012) evaluated the estrous response of dairy goats with different birth orders and observed that nulliparous females were more responsive to the male effect performed by the ram (83%) by than multiparous goats (81%). Moreover, the interval between the introduction of the male and estrus was similar regardless of the species (buck: 23 days, ram: 27 days). However, goats that had estrus induced by ram had 73% pregnancy rate, while goats induced by buck had 67% pregnancy. This may be related to the quality of the stimulus by the ram due to greater social interaction, as seen by Vitaliano et al. (2012), who assessed the presence of a ram in a herd of dairy goats and initially observed disinterest of the ram in females as well as a greater number of aggressive behavior towards the buck, but, at the end of the mating season, the ram displayed a higher number of reproductive behavior relative to the buck (Fig 2), proving to be very effective in inducing estrus in these does, ram: 95%; buck: 100% (Vitaliano, 2011).

USE OF THE MALE EFFECT IN ARTIFICIAL INSEMINATION PROTOCOLS

Artificial insemination is a widespread reproductive biotechnique worldwide and applied to reproductive management of goats (Martínez-Rojero et al., 2006). When using insemination, this is preceded by hormonal protocols, from the most simplified to the most sophisticated (Wildeus 1999). The male effect is an alternative to reduce the widespread use of hormones, because of both the low cost and the adequacy to organic production conditions, meeting the emerging requirements from consumer market, seeking for products from sustainable production systems (Martin & Kadokawa, 2006; Pellicer-Rubio et al., 2007). Physiologically, the use of biostimulation for the purpose of estrus induction combined with artificial insemination does not affect the physiology of cyclical events (Durrant, 2009).

In the literature, most studies involving the male effect and reproductive biotechniques comprises the use of exogenous hormones, and the male effect is often used only to start the reproductive activity of females (López-Sebastian et al., 2007; Pellicer-Rubio et al., 2008). In this way, there are few studies using only the male effect as the primary means of estrus induction and synchronization of goats in artificial insemination protocol. One of the main limitations for its use is the variation in LH surge and consequently the time of ovulation (Pellicer-Rubio et al., 2007).

Restall (1988) combined the male effect with artificial insemination during fall, and achieved more than 80% fertility when does were inseminated from 6 to 10 days after the beginning of induction. Nevertheless, when inseminated between the first and the fifth day after the initiation of the male effect, the fertility of these females was lower than 52%. Moore and Hall (1991) induced estrus with the male effect and inseminated does following the Trimberg method, and obtained 56% pregnancy, being higher when insemination was performed between 6-10 days after the start of the male effect. Likewise, Viana Neto et al. (2015) inseminated dairy goats 24 hours after estrus detection and reached 51% pregnancy and 47% kidding, and these authors also observed that the second estrus is the most likely to result in pregnancy. This may be the result of physiological regulation, which acts on the reproductive function, after the introduction of the male. Pellicer-Rubio et al. (2016) verified that the insemination 24 hours after the LH surge results in a higher kidding rate (70%) and two subsequent inseminations, on the 6^{th} and 7^{th} day after the introduction of the male, exhibited fertility similar to the protocol using hormones.

Some authors report that a good portion of the females when exposed to the male effect have short intervals between the 1^{st} and 2^{nd} estrus, by action of progesterone, due to the short cycle and may result in greater fertility (Restall, 1988; Pellicer-Rubio et al., 2016). Further, the second estrus of does, when driven by the male effect, is related to a greater amount of estrus followed by an increased ovulation rate (Chemineau, 1983; Delgadillo et al., 2011).

Those fertility and kidding rates are not high but satisfactory, and they may be related to changes in the time of the LH surge, which appears quite variable when employing the male effect, and thus represents a barrier to the association between the male effect and artificial insemination (Pellicer-Rubio et al., 2007). Thus, the use of the male effect as estrus induction and synchronization technique in goats for an artificial insemination program is a viable alternative, but it is extremely important to be attentive to the factors that interfere with both the male effect efficiency, and time of ovulation, which is essential for a good pregnancy outcome.

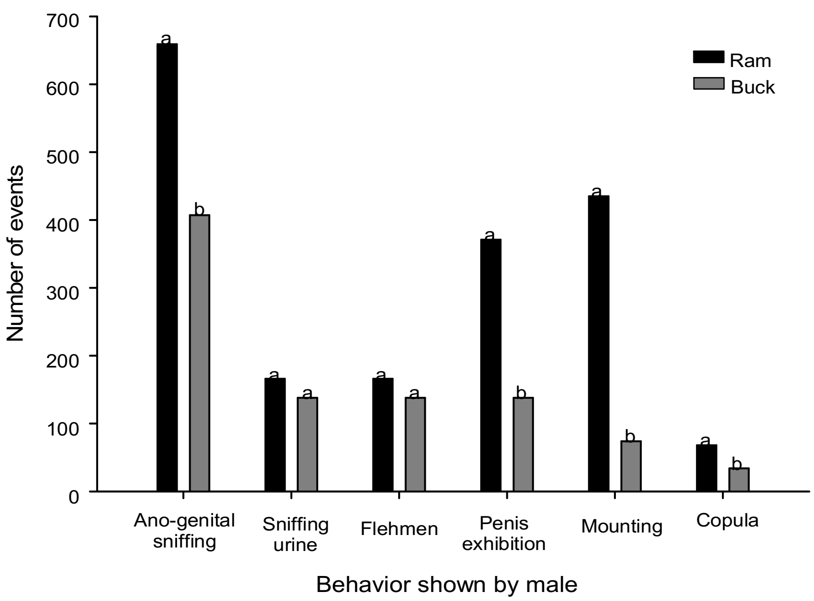

Figure 2. Frequency of behavioral activities in males in presence of goats (Adapted from Vitaliano et al., 2012).

CONCLUSION

Induction and synchronization of female estrus by the male effect is a relevant technique in the reproductive management of flocks, because the advantages indicated by several authors include direct reduction of costs, prevention of unwanted immune responses through the use of chorionic gonadotropin, reduction of hormonal residues in treated females and hence in the environment, thus complying with ecological and animal welfare principles in ethical and sustainable animal production.

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