



Sperm collection by electroejaculation in small ruminants: A review on welfare problems and alternative techniques

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ABSTRACT

There are different perspectives on whether there should be use of electroejaculation (EE) for semen collection because it can be stressful and painful for the males when this technique is imposed. In the present review it is examined 1) the effects of EE on animal welfare and semen quality in domestic and wild small ruminants, 2) benefits and limitations of administering anaesthetics and sedatives prior to EE, 3) advantages/disadvantages of transrectal ultrasonic-guided massage of the accessory sex glands (TUMASG) as an alternative to EE, and 4) benefits of administering hormones, such as oxytocin or PGF2 α analogues (which stimulate the contractility of the male accessory sex glands), prior to EE and TUMASG. In general, the administration of anaesthetics, sedatives or hormones reduces the pain and stress caused by EE, and can improve sperm quality, but results may vary depending on the species. The use of anaesthetics is, however, not devoid of risks and pre-EE administration of sedatives, or oxytocin or PGF2 α analogues, can aid sperm collection mitigate risks. The TUMASG is less stressful than EE, but its effectiveness varies greatly among species, and it can only be performed by trained personnel. Prior administration of the hormones may also result in a reduction in the period needed to induce ejaculation with use of TUMASG procedures.

1. Introduction

Artificial vaginas (AV) are widely used for semen collection from ruminants (Leboeuf et al., 2000). This is a practical method and use of this technique does not lead to alterations in semen quality compared to that when there is natural mating. When these techniques cannot be used or are not practical, semen is commonly collected by electroejaculation (EE).

The EE technique is frequently used to collect sperm from animals of seasonal breeding species outside of the normal breeding season (Lincoln and Davidson, 1977; Chemineau et al., 2008). The EE technique is also useful for collecting semen from animals not trained to ejaculate into an AV (training may require 1–2 weeks) (Wulster-Radcliffe et al., 2001), when attempting to collect semen from prepubertal males (Lacuesta et al., 2015), the males of wild species (Santiago-Moreno et al., 2009), or males that have been isolated from females (Giriboni et al., 2017). Although EE is an easy technique to use and is effective, it can be stressful and painful to the males on which the technique is being imposed (Stafford et al., 1996; Bath, 1998; Orihuela et al., 2009a).

To overcome the negative aspects with use of EE, anaesthetics, sedatives and hormones can be administered prior to initiation of

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Table 1
Electroejaculation protocols used with different ruminant species.

Species	Voltage	Number of electrical pulses	References
Domestic species			
Bull (<i>Bos taurus</i>)	10-57 V	16-28	Furman et al. (1975)
	1-20 V	20-25	Knight (1955)
	1-13 V	107	Whitlock et al. (2012)
Ram (<i>Ovis aries</i>)	8 V	1-5	Damián (2011)
	1-4 V	7	Ledesma et al. (2014)
	3-4 V	9-13	Abril-Sánchez et al. (2018)
Buck (<i>Capra hircus</i>)	4 V	21-25	Abril-Sánchez et al. (2018)
	4-5	28-40	Abril-Sánchez et al. (2018)
	4-5 V	39-46	Ungerfeld et al. (2018)
Camel (<i>Camelus dromedarius</i>)	10-40V	40	Tharwat et al. (2014)
Non-domestic species			
Mouflon (<i>Ovis musimon</i>)	0,1-0,3 mA	20	Ungerfeld et al. (2015)
Iberian Ibex (<i>Capra pyrenaica</i>)	0,1-0,3 mA	26-40	Santiago-Moreno et al. (2011); Ungerfeld et al. (2015)
Blanca-Celtibérica breed buck (<i>Capra hircus</i>)	1-5 V	4-16	Jiménez-Rabadán et al. (2012)
Caspus red deer (<i>Cervus elaphus maral</i>)	1-8 V	3-4	Cited by Garde et al. (2006), see Sipko et al. (1997)
Pampas deer (<i>Ozotoceros bezoarticus</i>)	1-5 V	10-50	Fumagalli et al. (2015)
	3-4 V	30-40	Fumagalli et al. (2015)
	2-4 V	20-30	Umaphathy et al. (2007)
Spotted deer (<i>Axis axis</i>)	2-6 V	90	Wahid Haron et al. (2000)
Java mouse deer (<i>Tragulus javanicus</i>)	2-5 V	10-40	Roth et al. (1999)
Oryx (<i>Oryx dammah</i>)	5-9 V	5	Holt et al. (1996)
Gazelle	<i>Gazelle dama mhorr</i>	1-5 V	5
	<i>Gazelle dorcas</i>		
	<i>Gazelle cuvieri</i>		

use of the technique, but this can affect semen quality. For example, the use of anaesthetics may interfere with the neuromuscular mechanisms that control erection and ejaculation (Santiago-Moreno et al., 2011), influencing the characteristics of the collected sperm. Alternatives to EE have also been developed, such as transrectal massage but the use of this approach is usually only viable with bulls (Palmer et al., 2004). Transrectal ultrasonic-guided massage of the accessory sex glands (TUMASG) has also, however, been used in domestic and wild small ruminants (Abril-Sánchez et al., 2017a; Santiago-Moreno et al., 2013; Ungerfeld et al., 2015).

This review was conducted to examine the negative effects of EE on the welfare of domestic and wild small ruminants, and to evaluate the advantages and disadvantages of the methods designed to reduce these problems.

2. Electroejaculation

The use of EE consists of the administration of low voltage, low current electrical pulses to the male rectum via a transrectal probe equipped with electrodes. This induces penis erection, semen emission, and eventually ejaculation (Brindley, 1981). The electrical pulses stimulate branches of the hypogastric plexus (Semans and Langworthy, 1938) surrounding the seminal vesicle, prostate gland, and ductus deferens (Thomas, 1983) inducing the emission of semen from the tail of the epididymis into the prostatic urethra. Use of this approach also results in stimulation of the pudendal nerve, which has branches that innervate the proximal region of the urethra, inducing contractions of the urethral muscles and ejaculation (Semans and Langworthy, 1938; Thomas, 1983). A number of electrical protocols are available for use with different ruminant species (Table 1), although electrical pulses are generally separated by intervening (“rest”) periods of 2–3 s where no stimulation is induced.

The use of this technique is a practical method for semen collection, as there is not the need to train males for use of the technique in ways that occurs with collection of semen using an AV, and it is effective and easy to use on farms with a large number of animals from which there is need for semen collection. In addition, many ruminant species have seasonal reproductive patterns, including fluctuations in the extent sexual behaviour is expressed (Lincoln and Davidson, 1977), with a decrease in reproductive functions and behaviours during non-breeding season (Chemineau et al., 2008). The use of EE, therefore, can occur throughout the year.

3. Welfare problems: EE is stressful and painful

Even though the use of EE is an effective semen collection method (Palmer, 2005), the stress and pain induced by EE have led to questioning of whether it should be used for semen collection in several countries (Falk et al., 2001). There are also concerns as whether the EE technique should be used because of animal welfare concerns when EE is imposed without anaesthesia. In several European countries, the use of EE for semen collection is banned as is considered to be an inhumane practice. In the European Union, the importation of semen is prohibited if the semen was collected by imposing EE (Falk et al., 2001). Currently, there are palliative protocols for animal management that induce minimal pain and stress that are consistent with animal welfare international legislation standards (UNESCO – Universal Declaration of Animal Rights 17-10-1978).

The use of EE induces typical stress responses, with physiological (increases of rectal temperature, respiratory and cardiac frequencies), endocrine (increase of serum cortisol concentration), biochemical (changes in haematocrit and haemoglobin

Table 2
Stress responses to electroejaculation reported in different ruminant species.

Species	Type of response				References
	Physiological	Endocrine	Blood biochemistry	Haematological	
Goat buck (<i>Capra hircus</i>)	↑ RT, HR	↑ Cortisol	↑ Glycaemia, TP, Alb, Glb	↑ Hto, Hgb (after EE)	Abril-Sánchez et al. (2017a); Ortiz-de-Montellano et al. (2007)
Ram (<i>Ovis aries</i>)	↑ RT, HR, RR	↑ Cortisol	↑ Glycaemia, TP		Abril-Sánchez et al. (2017b); Damián and Ungerfeld (2011)
	↑ HR	↑ Cortisol ↑ Cortisol			Orihuela et al. (2009a, 2009b, 2010) Alomar et al. (2016); Stafford et al. (1996)
Bull (<i>Bos taurus</i>)		↑ Cortisol ↑ Progesterone ↑ Corticosteroids ↑ Progesterone ↓ Testosterone			Etson et al. (2004); Falk et al. (2001); Whitlock et al. (2012) Welsh and Johnson (1981)
Pampas deer (<i>Ozotoceros bezoarticus</i>)	↑ HR ↑ HR		↑ CK, ALP, AST ↑ CK		Mosure et al. (1998) Fumagalli et al. (2015)
Mouflon (<i>Ovis musimon</i>)	↑ HR	↑ Cortisol	↑ Glucaemia		Fumagalli et al. (2015) Ungerfeld et al. (2015)
Iberian Ibex (<i>Capra pyrenaica</i>)	↑ HR, RR	↑ Cortisol	↑ Glucaemia ↑ CK		Ungerfeld et al. (2015)
Camel (<i>Camelus dromedarius</i>)	↑ HR, RR ↑ PCO ₂	↑ Cortisol		↑ Ac. Lactic ↓ Blood pH ↓ VCM	Tharwat et al. (2014)

RT: rectal temperature; HR: heart rate; RR: respiratory rate; TP: total proteins; CK: creatine kinase; ALP: alkaline phosphatase; Alb: albumin; AST: aspartate aminotransferase; Glb: globulin; Hto: haematocrit; Hgb: haemoglobin; RBC: red blood cell; VCM: medium corpuscular volume of red blood cell; PCO₂: partial pressure of carbon dioxide. ↑: increase; ↓: decrease.

concentrations) and haematological alterations occurring (Damián and Ungerfeld, 2011; Abril-Sánchez et al., 2017a,b) (Table 2). The use of EE also induces increases in serum creatine kinase (CK), a consequence of muscle damage (Aktas et al., 1993); however, results of some studies indicate that the increase of CK may result when wild ruminants were already having muscle myopathy as a result of the stress of capture prior to the use of EE (Paterson, 2007). If EE induces a greater release than what was occurring from capture-induced muscle myopathy in wild animals welfare risks to the animals may be further increased. In addition, rams (Damián and Ungerfeld, 2011) and bulls (Falk et al., 2001; Whitlock et al., 2012) vocalise during the electrical stimuli, indicating the use of EE is painful (Damián and Ungerfeld, 2011; Palmer, 2005; Abril-Sánchez et al., 2017a). Pampas deer vocalise, however, when there is use of general anaesthesia and are imposed with EE (Fumagalli et al., 2015). Although semen collection with use of EE is an effective procedure (Palmer, 2005), the development of other alternatives is needed to address the animal welfare concerns that have arisen when there is imposing of the use of this technique.

During ejaculation, in male ruminants there are changes in serum cortisol (Borg et al., 1991), heart rate (Orihuela et al., 2016), respiratory rate (Slob et al., 1986) and haematological variables (Hatazoe et al., 2012) similar to those that occur when EE procedures are imposed. Abril-Sánchez et al. (2017b) reported that when the use of the EE technique is imposed on ewes the response is similar to those elicited in rams, indicating these are reliable indicators of the response to the procedure.

4. Quality of sperm collected by EE

In goats (Memon et al., 1986) and bulls (León et al., 1991), semen collected using EE has a lesser sperm concentration than that collected using an AV. In bulls, there is also a lesser sperm concentration in semen collected using EE than that in semen collected using the transrectal massage technique (Sarsaifi et al., 2015). In addition, the number of viable spermatozoa and mass motility are less, and the pH of the semen greater than that of semen collected with an AV (for goats see Greyling and Grobbelaar (1983) and Memon et al. (1986); for bulls see León et al. (1991)). Further, in both goats and bulls, the sperm of ejaculates collected using EE are less resilient to withstand cryopreservation than the sperm collected using an AV (for goats see Jiménez-Rabadán et al. (2012); for bulls see León et al. (1991)). In bulls, sperm of semen collected using EE are also less resistant to cryopreservation procedures than those collected using transrectal massage (Sarsaifi et al., 2015). In rams, however, sperm in semen collected using EE have more intact plasma membranes and have normal mitochondrial function when compared with sperm collected using an AV (Ledesma et al., 2014).

Semen collected using EE have a greater proportion of seminal plasma (SP) than semen collected using an AV (Ledesma et al., 2014) which may be a consequence of overstimulation produced by electrical pulses applied during EE. Maxwell and Johnson, (1999) and Maxwell et al. (2007) reported that the constituents of SP preserve the integrity of plasma membrane, which might explain the larger number of spermatozoa with intact plasma membrane when semen is collected using EE. In addition, in both goats (Memon et al., 1986) and bulls (León et al., 1991), the ejaculates obtained using EE are of larger volume than those collected when using an AV, probably due to the greater contractions of the accessory sex glands induced using the electrical stimuli. In rams, stimulation

using EE of the accessory glands results in differences in the composition of the seminal plasma compared to that of AV-collected sperm, including the protein profile and the concentration of several electrolytes (Marco-Jiménez et al., 2008; Ledesma et al., 2014). Thus, both the welfare of semen donor animals and the characteristics of the semen obtained should be carefully considered before deciding which semen collection method should be used.

5. Reducing the animal welfare problems associated with EE

5.1. Use of anaesthesia or sedation

In some countries, the use of general anaesthesia is required to reduce the stress and pain associated with EE (Falk et al., 2001), but little information is available about any of the other advantages or disadvantages of use of these techniques for EE. Certainly, in bulls, the use of epidural anaesthesia to inhibit the neural transmissions for pain, reduces the number of vocalisations, prevents the increase in cortisol concentrations, and impedes any increase in heart rate in response to use of EE. All of these outcomes indicate the use of anaesthetics when imposing EE procedures results in a reduction in pain and stress (Mosure et al., 1998; Falk et al., 2001). In rams, however, the use of epidural anaesthesia can induce a stress response before initiation of the use of EE with administration inducing transient paraplegia, which is evident by the animal's response to the aesthetic treatment (Damián, 2011). General anaesthesia induced by ketamine plus xylazine mitigates the increase in heart rate and cortisol concentrations in rams (Orihuela et al., 2009a) and goat bucks (Abril-Sánchez et al., 2018) during EE. With use of anaesthetics there is also a partial attenuation of any increase in rectal temperature and changes in haematological and biochemical indicators of stress in goat bucks (Abril-Sánchez et al., 2018). The use of anaesthesia with xylazine plus ketamine also improves the quality of sperm when there is use of EE (Abril-Sánchez et al., 2018), probably due to the muscle contractions of the epididymis, accessory sex glands and deferent ducts that are induced by the α -adrenergic agonist (Knight, 1974) which facilitate the semen output and result in greater sperm quality in the ejaculate. Anaesthesia cannot be used, however, without risks (Brodgelt et al., 2008; Bille et al., 2012), and ruminants need to be fasted for 12–24 h to reduce the possibility of regurgitation and tympanism when there is use of anaesthetics before using EE for semen collection (Taylor, 1991; Carroll and Hartsfield, 1996).

Xylazine is the α -adrenergic agonist most widely used to anaesthetise domestic ruminants, but more potent agonists, such as detomidine or medetomidine, are required for wild ruminants (Santiago-Moreno et al., 2010). Because wild animals have to be darted for anaesthesia, only drugs that can be administered in small volumes can be used (Santiago-Moreno et al., 2010) for this purpose. Detomidine is, therefore, preferred to medetomidine for this type of anaesthesia (Santiago-Moreno et al., 2011). The use of benzodiazepines, such as zolazepam, in combination with α -adrenergic agonists and dissociative anaesthetics (ketamine and tiletamine) is effective for anaesthetising wild aoudads (*Ammotragus lervia*) (Santiago-Moreno et al., 2013), ibexes (*Iberian ibex*) (Santiago-Moreno et al., 2011), mouflons (*Ovis musimon*) and chamois (*Rupicapra pyrenaica*) (Pradieu et al., 2016). With these drugs, the muscles are adequately relaxed, and changes in rectal temperature are attenuated, helping to prevent capture myopathy (Santiago-Moreno et al., 2010). It should be remembered, however, that the use of different anaesthetics induces differences in responses in different species.

Sedation attenuates the stress responses induced by EE in goat bucks to an extent similar to that which occurs with use of general anaesthesia, avoiding the risk associated with this latter practice (Abril-Sánchez et al., 2018). Animals also recover in a shorter period of time from sedation than from anaesthesia, and the quality of sperm in the ejaculates collected with use of EE from animals that have been sedated is similar to that of obtained from animals that have been anaesthetised (Abril-Sánchez et al., 2018).

5.2. TUMASG: an alternative to EE

The use of the TUMASG semen collection technique first occurred in elephants (Schmitt and Hildebrandt, 1998) and with this technique there is use of ultrasonography to observe the ampulla and monitor the ejaculatory response (Hildebrandt et al., 1998). The TUMASG technique has been used to collect semen from humans (Fahmy et al., 1999) and bulls, although it usually requires the additional use of some electrical pulses (Palmer et al., 2004). The TUMASG technique is useful for collecting semen from wild ruminants because there are few or no electrical pulses required when imposing the use of this technique (for information regarding the aoudad see Santiago-Moreno et al. (2013); for ibex and mouflon see Ungerfeld et al. (2015)). The TUMASG technique has also been used in domestic ruminants as an alternative to EE. With use of this approach for semen collection, there is the requirement for fewer electrical stimuli and there is, therefore, mitigation of animal welfare concerns (Abril-Sánchez et al., 2017a).

The procedure involves massaging the ampulla of the ductus deferens with the ultrasonic probe, alternating with vigorous manual massages of the bulbourethral gland, pressing it against the symphysis pubis. Simultaneously, the penile, perineal and pelvic parts of the urethra are massaged to facilitate the transport of the ejaculatory fluids. The ampulla of the ductus deferens is monitored by ultrasonography to assess when it is empty, thus avoiding unnecessary electrical stimuli as a result of the procedure being halted when no more semen is available for collection. The content of the ampulla of the ductus deferens appears anechoic and has a tubular appearance before TUMASG, but following ejaculation it becomes more echogenic, smaller in size, and serpentine in shape (Santiago-Moreno et al., 2013). The positioning of the probe close to the accessory glands may also stimulate the adrenergic nerves, favouring ejaculation similar to what occurs with use for EE, but with more moderate stimuli.

In wild species, the use of TUMASG for semen collection also requires the use of general anaesthesia, although there is need for fewer electrical pulses to induce ejaculation, therefore, there is a reduction in the risk of capture myopathy in ibexes (Ungerfeld et al., 2015). In anaesthetised mouflons, the use of TUMASG allows for there to be a shorter time period for semen collection compared to use of EE (Ungerfeld et al., 2015), however, the time period needs for semen collection using this approach is greater for non-

Table 3

Number of electrical stimuli (range) and intensity (mA or V) required to induce ejaculation using transrectal ultrasonic-guided massage of the accessory sex glands in different ruminant species, with or without anaesthesia; Electrical stimuli lasted 5–8 s and are provided using an electroejaculator with intermittent periods of non-stimulation for echo-guided transrectal massage.

Specie	Number of electrical stimuli	Electrical intensity	References
Goat (<i>Capra hircus</i>)	1–2	3 V	Abril-Sánchez et al. (2017a), Ungerfeld et al. 2016
Aoudad (<i>Ammotragus lervia</i>)	0–3	0.2 mA	(Santiago-Moreno et al., 2013)
Chamois (<i>Rupicapra pyrenaica</i>)	0	–	Unpublished data
Mouflon (<i>Ovis musimon</i>)	0–2	0.2 mA	Ungerfeld et al. (2016, 2015)
Iberian Ibex (<i>Capra pyrenaica</i>)	0–4	0.2 mA	Ungerfeld et al. (2015)
African Pygmy goat (<i>Capra hircus</i>)	1–4	0.2–0.5 mA	Unpublished data
Red deer (<i>Cervus elaphus</i>)	1–2	0.4 V	Unpublished data
Fallow deer (<i>Dama dama</i>)	2–5	0.2–0.3 mA	Unpublished data
Père David's deer (<i>Elaphurus davidianus</i>)	2	0.2–0.3 mA	Unpublished data
Reeves' muntjac (<i>Muntiacus reevesi</i>)	4–10	2–4.5 V	Unpublished data
Roe deer (<i>Capreolus capreolus</i>)	2–9	0.2–0.45 V	Unpublished data
Dorcas gazelle (<i>Gazella dorcas</i>)	1–9	2–5 V	Unpublished data
Sitatunga (<i>Tragelaphus spekii gratus</i>)	5–11	0.2–10 V	Unpublished data

anaesthetised domestic goats (Abril-Sánchez et al., 2017a). An important limitation of TUMASG is that it must be performed by experienced staff.

The need for electrical pulses with use of TUMASG depends on the semen donor species (Table 3). Aoudads and chamois usually ejaculate without the use of electrical stimulation (Santiago-Moreno et al., 2013), mouflons require only one or two electrical stimuli (with some requiring none), and ibexes require zero to four electrical pulses (Ungerfeld et al., 2015, 2016) to induce ejaculation. Goats require at least one electrical stimulus (Abril-Sánchez et al., 2017a; Ungerfeld et al., 2016) to induce EE. Compared with the number of electrical pulses required for use of EE (Santiago-Moreno et al., 2011; Abril-Sánchez et al., 2017a), these are important reductions with use of TUMASG.

Sperm collected with use of TUMASG differ from sperm collected using EE. In mouflons, the use of EE results in obtaining sperm with greater values for motility variables and similarly with the ibex there is sperm with greater progressive motility collected with use of EE than when there is use of TUMASG. This is probably because with EE there is a greater stimuli due to electrical pulses for semen collection (Ungerfeld et al., 2015). In non-anaesthetised goat bucks, the use of TUMASG, however, does not affect sperm characteristics compared to EE (Abril-Sánchez et al., 2017a). This may be related to species-dependent sensitivity, but also to the differential effects anaesthesia among species.

5.3. Hormonal treatments that enhance EE- and TUMASG-associated ejaculation

In rabbits (Todd and Lightman, 1986) and humans (Murphy et al., 1987), there is an increase in serum oxytocin concentration during natural ejaculation. In bulls, the use of EE induces an increase in oxytocin concentrations in blood (Schams et al., 1982). Increases in oxytocin and vasopressin during ejaculation stimulate epididymal contractility in rats (Studdard et al., 2002) and rams (Nicholson et al., 1999), and of the prostate in most mammals that have been studied (Bodanzky et al., 1992). There are receptors for oxytocin in several regions of the male reproductive tract, including the epididymis, tunica albuginea, ductus deferens and seminal vesicle (Filippi et al., 2002). In rats, the administration of oxytocin may induce ejaculation (Watcho et al., 2014). This hormone has regulatory actions on the ejaculatory process by having effects on the peripheral, central, and probably spinal neural transmissions (see reviews by Corona et al., 2012, 2013). The administration of oxytocin shortly before the use of EE for semen collection results in an increase in the number of sperm in bull (Berndtson and Igboeli, 1988) and ram (Voglmayr, 1975) ejaculates. Administration of oxytocin will also result in an increase in the number of sperm collected from rabbit bucks when administered before there is use of an AV for semen collection (Sharma and Hays, 1976).

One of the actions of PGF_{2α} is stimulation of the contraction of the smooth muscles involved in ejaculation (Cosentino et al., 1984). There certainly are actions of PGF_{2α} in the ejaculatory processes of stallions (McDonnell, 1995). With PGF_{2α} administration, there is also an increase in the number of sperm cells collected when using an AV with rabbits, bulls (Hafs et al., 1974), rams (Knight, 1974), buffalo bulls (Capitan et al., 1990) and boars (Hemsworth et al., 1977) - although there are some reports in which there are no positive effects of oxytocin administration when used in bulls (Berndtson et al., 1979) and boars (Levis and Reicks, 2005). It appears, however, that these hormones when administered generally lead to a shortening in the duration of EE or TUMASG for induction of ejaculation. There is, therefore, a reduction as a result of use of these hormones in those male responses with use of EE that are associated with animal welfare concerns. Certainly, oxytocin administration has been reported to shorten the time before ejaculation induction in bulls subjected to EE (Palmer et al., 2004). The combined administration of a PGF_{2α} analogue (cloprostenol) and oxytocin (250 µg and 10 UI intramuscularly, respectively) before collecting semen with use of EE results in a shortening in the duration of the procedure and reduction in the need to use the number of electrical stimuli required when there is use of EE for semen collection in non-anaesthetised goat bucks (Ungerfeld et al., 2018). With this approach for semen collection in goats, there is no detrimental effect on the quality of semen collected. The administration of oxytocin or PGF_{2α} separately, however, did not have the beneficial effects as compared to administration of the two hormones in combination. Administering oxytocin to non-anaesthetised

goat bucks before the use of TUMASG for semen collection allowed for shortening of this procedure for semen collection, and there was a trend towards reducing the number of electrical pulses needed before ejaculation occurred (Ungerfeld et al., 2016). When EE is imposed for semen collection, this may induce an increase in oxytocin secretion (Schams et al., 1982), perhaps explaining the lack of positive effects of oxytocin when administered alone before the use of EE for semen collection. Similarly, the use of TUMASG for semen collection requires the massage of the accessory sex glands, which may also induce endogenous increases in oxytocin release (Peeters et al., 1983), although probably to a lesser extent than those induced with use of EE. In another study, oxytocin administration had no effect in anaesthetised mouflons, perhaps because anaesthetised animals are less responsive to hormonal stimuli (Ungerfeld et al., 2016).

Although the use of these hormones may be practical and useful, there is little information on protocols for use when collecting semen. The proper timing has not been ascertained for hormone administration relative to the timing when there is imposing of EE for semen collection so that there is minimisation of the time and the number of electrical pulses needed. Furthermore, there are no studies on the possible advantages/disadvantages of these treatments with respect to sperm cryopreservation. In general, the treatment with both hormones is innocuous with no obvious detrimental effects as a result of these administrations before semen collection. These hormonal treatments may, therefore, safely reduce the stress and pain of semen donor animals when there is imposing of EE and other approaches for semen collections.

6. Conclusions

The use of EE for semen collection is stressful and painful, but the use of sedation or general anaesthesia can reduce these problems, and might lead to improvements in semen quality. The use of hormones, such as oxytocin combined with PFF_{2α}, may reduce the negative effects associated with EE, however, outcomes may vary between species. Further, the administration of oxytocin in combination or without being combined with use of analogues of PGF_{2α}, or the use of sedatives, may lead to improvements for sperm collection with use of EE without increasing the risk that results with use of general anaesthesia. The use of TUMASG requires fewer electrical stimuli for ejaculation to occur than with use of EE and in some cases there is no need for use of electrical pulses for induction of ejaculation. The period for induction of ejaculation may also be shorter with the use of these hormones. For there to be proper use of the TUMASG procedure, there is a requirement for properly trained personnel.

Based on the findings from studies cited in this review article, EE protocols may be enhanced, however, there also are alternative protocols for semen collection in some situations that may be considered for substitution of the use of EE for semen collection. There is, however, need for further research, because there is need for more knowledge about how males of different species respond to ejaculatory induction techniques in development of protocols for semen collection with or without administration of hormones or sedative drugs. The physiological basis of the differences observed in different species in response to ejaculatory induction need to be understood, protocols involving the use of anaesthetics, sedatives, or combinations of these hormones, both with and without hormonal pre-treatments, need to be standardised, and the characteristics of sperm collected using different techniques need to be determined. Importantly, if procedures designed to reduce the pain and stress experienced by males when there is ejaculatory induction imposed are to be widely used, the effects of these treatments on sperm cryo-resistance during storage must be known.

Authorship

All authors contributed to the drafting of this review and approved it before submission.

Conflict of interest

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