

# 1 Electro-ejaculation in ruminant livestock: A welfare issue

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## 20 Introduction

21 The beef, mutton, wool and mohair industries generate billions of Rands annually. To obtain optimal  
22 production in the livestock, these industries rely largely on sound fertility in the animals, especially  
23 the male animals. Therefore, determination of breeding soundness is a prerequisite for acquiring  
24 and utilising the male animal. Examination for breeding soundness involves a complete assessment  
25 of sperm production and quality, which requires obtaining a semen sample. Semen collection can be  
26 achieved in various ways and Palmer et al. (2005) describe four ways which have varying degrees of  
27 success: electroejaculation (EE), artificial vagina (AV), aspiration of a sample from a recent  
28 ejaculation in the female animal and transrectal massaging of the accessory sex glands.

29 Transrectal massage prior to insertion of an electroejaculator probe provides sexual stimulation and  
30 encourages semen emission (Palmer, 2005). This technique requires a specific level of expertise and  
31 is applicable to bulls only. Aspiration of recent ejaculates in the vulva is an easy, non-invasive  
32 technique that does not cause discomfort to the animals. It requires that female animals in oestrus

33 are available in a small enclosure in order to obtain the sample directly after serving. This technique  
34 allows for the assessment and presence of live sperm only.

35 EE and AV are the most frequently used techniques (Damian and Ungerfeld, 2010; Palmer, 2005).  
36 According to Palmer (2005), the AV method of obtaining semen from domestic animals was in use  
37 long before the invention of the electroejaculator. The AV method is perceived to be a less painful  
38 and stressful alternative to EE, but requires training of the bull, buck or ram. The training is time-  
39 consuming and not all animals are suitable for training. During the training, the rams or bucks have  
40 to be isolated from the flock which is a separate stressor (Al-Qarawi & Ali, 2005). Female animals in  
41 oestrus or dummies are also required for AV. When a live animal is used as a “dummy”, the welfare  
42 concerns transfer from the male to the female animal.

43 The EE method in domestic animals has been in practice for decades (Dziuk et al., 1954; Marden,  
44 1954). The EE method is widely considered to be an effective method that does not require any  
45 training of animals or a female animal/dummy to be mounted, and which can be adapted to the  
46 handling facility. The procedure is, however, contentious since it is associated with discomfort to the  
47 animal during electrical stimulation. In recent years, welfare organisations have begun placing  
48 increasing pressure on producers to stop the use of EE. The Animal Welfare Requirements of the  
49 Responsible Wool Standard certification scheme, an independent, voluntary scheme, prohibits EE as  
50 a method for semen collection. In the United Kingdom, EE may only be carried out by a veterinary  
51 surgeon and the use of anaesthesia for the procedure is encouraged (Palmer, 2005). Some European  
52 countries have banned the use of EE. The Canadian Veterinary Medical Association advised that only  
53 experienced individuals should perform the procedure. In South Africa, EE is not regulated, but some  
54 superficial provision is made in the Animal Protection Act (Act No.71 of 1962) for unnecessary  
55 suffering. *“(f) uses on or attaches to any animal any equipment, appliance or vehicle which causes or*  
56 *will cause injury to such animal or which is loaded, used or attached in such a manner as will cause*  
57 *such animal to be injured or become diseased or to suffer unnecessarily.”* The EE procedure is,  
58 however, considered an acceptable procedure by most animal welfare organisations in South Africa,  
59 but it is becoming increasingly controversial. This article aims to review recent research concerning  
60 the effect of EE on rams, bucks and bulls.

## 61 **Electroejaculation in bulls, rams and bucks**

62 The use of EE as a method of obtaining a semen sample was reported as early as 1936 (Palmer,  
63 2005). Recent procedures and methods for EE have been refined with specially designed equipment.  
64 Modern electroejaculators for bulls achieve ejaculation with electrical impulses < 8 or 9 volts  
65 (Palmer, 2005) of sine-wave pulse at a frequency of 20-30 cycles. The modern electroejaculator

66 segmented probes are designed with three caudal electrodes, three middle and two probes at the  
67 cranial end. These segments can be activated independently as needed. This allows for electrical  
68 stimulation of desired nerves with little or no stimulation of non-target tissues. The purpose of the  
69 caudal segment is to produce an erection. The middle segment is then activated to induce  
70 ejaculation. Palmer (2005) describes the technique used in bulls that involve transrectal massage  
71 over the ampullae, seminal vesicles, prostate, pelvic urethra and inguinal rings for 10 to 60 seconds.  
72 The intention of the massage of those areas is to stimulate the bull sexually with subsequent  
73 relaxation of the anal sphincter prior to insertion of the electroejaculator probe. With the probe in  
74 place, electrical stimulation should be applied carefully while observing the response of the bull.  
75 Slight contraction of the muscles of the hind legs is an indication that the bull feels the stimulation.  
76 The stimulation should then be briefly stopped. Successive stimulations are then applied with a  
77 steady increase in voltage intensity, maintained for 1-2 seconds. This should be followed with 0.5 to  
78 1 seconds of no stimulation to rest. Stimulation continues through penile protrusion, erection and  
79 finally ejaculation whereby the semen is collected. (Furman et al., 1975). Comparing the aversive  
80 experience between bulls and rams or bucks towards EE is not justified. Semen collection by means  
81 of EE in bulls involves having the bull standing in a handling chute without physical constraints. The  
82 effect of isolation stress is thus not as relevant as would be the case in small stock that requires  
83 physical restraint in lateral recumbency.

84 Various commercially manufactured EE probes exist for use in rams or bucks. These probe types  
85 usually deliver stimulation of 10-15 volts of 30 – 50 sine or square waves. (Shipley et al., 2007). In  
86 contrast to bulls, EE procedures in rams or bucks require the animal to be constrained in lateral  
87 recumbency and physically held down by handlers. The penis is massaged out of the sheath, grasped  
88 behind the glans with sterile gauze and directed into the collection vial. Similar to cattle, gentle  
89 massaging of the accessory glands with either the fingers or the probe for 10 to 15 seconds will  
90 stimulate the ram sexually to ease semen collection. Electrical stimulation is applied with the probe  
91 for 3 to 5 seconds with a rest period of 5 to 15 seconds while applying gentle pressure downwards  
92 towards the pelvic floor. The movement of the probe during the on-off sequence stimulates the  
93 animal and helps with the collection. Individual rams or bucks may vary in their response to  
94 stimulation, but ejaculation usually occurs between 3 to 5 stimulations. The preparation of a ram or  
95 buck for EE semen collection is usually longer than the electrical stimulation itself and may  
96 contribute more to the discomfort the animal is experiencing than the electrical stimulus. It is  
97 strongly recommended that the equipment used for EE and the competency of the operators be  
98 regulated to align South Africa to international standards and to improve the welfare of the animals  
99 earmarked for EE procedures. Consultation with professional bodies such as the South African

100 Veterinary Council (SAVC), South African Veterinary Association (SAVA), South African Council for  
101 Natural Scientific Professions (SACNASP) and the Livestock Welfare Coordinating Committee (LWCC)  
102 would be of value in establishing such regulations.

### 103 Electroejaculation and pain

104 Based on animal behaviour in response to EE, the procedure has the potential to cause pain,  
105 discomfort and stress to the animal (Abril-Sánchez et al., 2017; Damian and Ungerfeld, 2010; Palmer,  
106 2005; Orihuela et al., 2009; Costa et al., 2018). There seems to be a consensus among scientists and  
107 researchers that EE has the potential to cause pain in bulls, bucks and rams. They do however differ  
108 in their description of the pain associated with EE. Palmer (2005) states that it is *considered* to be  
109 painful to bulls. Abril-Sánchez et al. (2018) describe EE in rams as stressful and *probably* painful.  
110 Baiee et al. (2017) and Damian and Ungerfeld (2010) state that EE in rams *may* be associated with  
111 pain. Abril-Sánchez et al. (2017) describe EE as a procedure that provokes muscular damage and  
112 *probably* pain.<sup>1</sup>

113 This reluctance to state conclusively that EE causes pain may be because animal and human  
114 experiences of pain, in response to a similar stimulus, may not be identical. Humans tend to report  
115 pain and seek help while animals will hide signs of pain because such signs may provoke an attack  
116 from predators or subordinate members of the group (Bayne, 2000). Humans can articulate a pain  
117 sensation. Ohl et al. (2001) reported humans who could not tolerate electrical stimulation during EE  
118 procedures due to pain. In contrast to human articulation, animals present with behavioural signs in  
119 response to pain stimuli. Ruminant animals are widely recognised as sentient beings and therefore  
120 it can be accepted that they will also experience pain and discomfort during EE. According to the  
121 Cambridge Declaration on Consciousness (Low et al., 2012):

122         The absence of a neocortex does not appear to preclude an organism from experiencing  
123         affective states. Convergent evidence indicates that non-human animals have the  
124         neuroanatomical, neurochemical, and neurophysiological substrates of conscious states  
125         along with the capacity to exhibit intentional behaviours. Consequently, the weight of  
126         evidence indicates that humans are not unique in possessing the neurological substrates  
127         that generate consciousness.

128 The U.S. Government Principles for Utilization and Care of Vertebrate Animals Used in Testing,  
129 Research, and Training (IRAC, 1985) states that "unless the contrary is established, investigators  
130 should consider that procedures that cause pain or distress in human beings may cause pain or

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<sup>1</sup> The author added emphasis to these quotes.

131 distress in other animals." (Bayne, 2000). One must, however, be cautious not to anthropomorphise  
132 and assign an animal's pain response equal weight to a human pain response. Humans are cautioned  
133 for anticipated pain, are thus more sensitised to it and may then exaggerate the sensation.

134 To complicate the matter more is the difficulty in defining pain. The Oxford Dictionary defines pain  
135 as a "highly unpleasant physical sensation caused by illness or injury." The Merriam-Webster  
136 dictionary describes it more elaborately as "a basic bodily sensation induced by a noxious stimulus,  
137 received by naked nerve endings, characterised by physical discomfort such as pricking, throbbing,  
138 or aching, and typically leading to evasive action." Baillière's Comprehensive Veterinary Dictionary  
139 defines pain in animals as "a feeling of distress, suffering or agony, caused by the stimulation of  
140 specialised nerve endings." The behaviour of an animal in response to painful stimuli is obvious and  
141 recognisable and does not need a definition to identify it. Prunier et al. (2013) describe four types of  
142 pain indicators in mammals: behavioural, physiological, lesional and production related. This article  
143 will discuss the behavioural and physiological indicators of pain.

#### 144 *Behaviour as an indicator of pain*

145 Vocalisation is widely recognised as a more accurate indicator of pain during EE when compared to  
146 other behavioural responses (Falk et al. 2001). It is an immediate response to pain and indicates the  
147 stage in the procedure where the electrical stimulus is most painful. Vocal behaviour differs between  
148 species, for example, the mere restraint of a goat will result in the animal being vocal, which is  
149 normal behaviour for a goat. In contrast to this, abusive restraint in a sheep will not elicit a vocal  
150 response. This does not mean that vocalisation is not an indicator of pain. Vocalisation during  
151 electrical stimulation and being silent in-between stimulations can be considered an indicator of  
152 pain.

153 Grandin (2010) refers to unpublished data (Voisinet and Grandin, 1997), where vocalisation  
154 responses in bulls in three treatments (restraint, restraint with low voltage EE and restraint with high  
155 voltage EE) differed significantly. Whitlock et al. (2012) published similar results with significantly  
156 more vocalisations in bulls during electrical stimulation compared to the control group with no  
157 stimulation. If vocalisation can thus be accepted as an indicator of pain, then these results support  
158 the argument that EE is a painful procedure. However, according to Bayne (2000), vocalisation alone  
159 is not a sufficiently sensitive measure in the assessment of pain.

160 Molony and Kent (1997) describe more types of responses to pain, besides vocalisation. One of  
161 these is the modification of the animal's behaviour by learning, which enables the animal to avoid a  
162 recurrent experience. In Molony and Kent's (1997) experiment, sheep were moved through a  
163 handling chute to a pen for different treatments:

- 164 (i) partial shearing
- 165 (ii) EE
- 166 (iii) free movement

167 The aversive behaviour to the respective treatments was reflected in the time it took the sheep to  
168 move through the chute later on in the trial. Transit times for both EE and partial shearing were  
169 significantly higher than for the free movement pen. Personal experience by the author has  
170 witnessed similar behaviour of Zulu sheep rams where semen was collected by EE twice a week.  
171 When the sheep recognised the semen collection team, they refused to move into or through the  
172 handling chute. The same rams would, however, move freely through the handling chute when the  
173 regular handler who fed the sheep was at the chute. This time-averse relationship, known as  
174 “conditioned suppression” (Rushen, 1986) is indicative of pain associated procedures. This is,  
175 however, not the same in all animal species. Bulls subjected to EE twice a week for several weeks  
176 developed little aversion to the procedure. Bulls who were lining up in the chute and exhibited  
177 preputial and even penile protrusion, seemingly in anticipation of EE (Palmer, 2005). This lack of  
178 aversive behaviour may be interpreted as EE not inflicting pain in bulls. The degree of aversion is  
179 however influenced by the intensity and duration of the electric shock. Aversive behaviour was more  
180 evident if the electric stimulation was intense or of a long duration (Rushen, 1986). The lack of  
181 aversive behaviour may imply that the EE procedure can be performed without causing major  
182 discomfort or pain in bulls, provided that the intensity and duration of the electrical stimulus are  
183 tolerable for the bulls. This further supports the motivation to use proper standardised equipment  
184 and competent operators to perform the technique.

185 From the available literature, there seems to be a more pronounced aversive experience noticed in  
186 rams in contrast to bulls. As mentioned earlier, there is a significant difference in the execution of  
187 the procedure between the two species. Restraint of bulls for EE merely involves the bulls standing  
188 in a chute. Even though the technique in both species involves a disturbance of the homeostasis, it is  
189 worse for rams that need to be restrained physically in lateral recumbency in contrast to bulls  
190 allowed to be free standing. More research needs to be conducted on the differences in the  
191 anatomy, physiology and psychological factors of ejaculation between cattle and small stock to  
192 understand the difference in aversive behaviour for the procedure.

### 193 *Physiological indicators of pain*

194 According to the Merck Veterinary Manual:

195 Pain is a complex, multidimensional experience with sensory and affective elements. All  
196 mammals process the neuroanatomic and neuropharmacological components involved in

197 transduction, transmission, and perception of noxious stimuli; therefore, it is expected that  
198 animals experience pain even if they cannot exactly perceive or communicate it in the same  
199 way people do.

200 According to Prunier et al. (2013), the nociceptive withdrawal reflex is the simplest form of  
201 avoidance and defensive behaviour. Defensive behaviour such as leg and body movements (as if the  
202 animal was trying to escape the painful stimuli) were observed during EE in rams and bucks  
203 restrained in lateral recumbency even before electrical stimuli. It can thus not be considered  
204 nociceptive withdrawal reflex behaviour. Any sheep or goat will attempt to escape restraint in a  
205 normal position and even more so in an abnormal position. All the current methods available to  
206 determine pain in animals are prone to errors of under- or overestimation. The way in which the  
207 animal is coping with the pain further complicates an accurate estimation of pain in the animal.

208 Physiologic parameters such as changes in rectal temperature, heart rate and respiratory rate  
209 (Boussena et al., 2013) indicate responses to acute noxious (painful) stimuli, but moderate stress  
210 levels due to handling and restraint have a similar effect on those parameters. Boussena et al.  
211 (2013) report significant changes in rectal temperature, heart rate and to a lesser degree respiratory  
212 rate, in rams during EE procedures. The increase in heart rate described by Boussena et al. (2013)  
213 that occurred during EE is ascribed to a combination of pain and muscle contractions. However, an  
214 increase in heart rate as an indication of pain is contradicted by Mosure et al (1988) with the  
215 argument that normal sexual activity also leads to an increase in heart rate. The heart rate in bulls  
216 was evaluated after six different treatments for EE in an experiment by Mosure et al (1988):

- 217 (i) transrectal massaging of the seminal vesicles
- 218 (ii) conventional EE
- 219 (iii) intra-rectal lidocaine prior to EE
- 220 (iv) intra-venous xylazine prior to EE
- 221 (v) epidural lidocaine prior to EE
- 222 (vi) epidural xylazine prior to EE.

223 During the experiment, heart rates were recorded prior to treatment, immediately after treatment  
224 and every two minutes for up to ten minutes after EE. All six treatments presented with increased  
225 heart rates, with no significant difference among the groups. In all the treatments, the heart rates  
226 returned to the baseline within two minutes of cessation of electrical stimulus. The rapid return of  
227 the heart rate to the baseline suggests that pain associated with EE was of very short duration and  
228 furthermore that heart rate cannot be utilised as a reliable measure of pain in bulls. Pain is a  
229 powerful stressor that stimulates the release of hormones from the hypothalamus-pituitary-adrenal

230 axis (HPA-Axis) (Prunier et al., 2013). Many studies have examined cortisol concentrations in blood  
231 samples after painful interventions such as castration in calves and lambs (Cohen et al., 1990; Lester  
232 et al., 1991). Elevated cortisol concentrations are, however, an unreliable indicator of pain during EE,  
233 because it is elicited by activities requiring energy mobilisation, including normal sexual activity  
234 (Moberg, 2000). According to Palmer (2005), early attempts to use cortisol to measure the response  
235 to pain associated with EE have been unreliable. Routine handling procedures involving no pain and  
236 very little stress, such as drenching and subcutaneous vaccination have resulted in significantly  
237 elevated blood cortisol concentration for up to 60 minutes (Kruger et al., 2016). Profiles of mean  
238 cortisol concentration were similar between groups of bulls when a rectal probe was inserted  
239 without electrical stimulation and when a rectal probe was inserted with electrical stimulation  
240 applied (Welsh and Johnson, 1981). These results indicate that activation of the HPA-Axis can result  
241 from the handling of the animal and not necessarily from a painful stimulus.

242 Orihuela et al. (2009) report contradictory results in a similar study with sheep. In the study  
243 involving rams that had previously undergone EE, the effect of EE on blood cortisol was evaluated.  
244 Two treatments were applied: first with only a rectal probe inserted without electrical stimulus and  
245 second, a rectal probe inserted with electrical stimulus. An untreated control group was also  
246 incorporated into the study. Blood cortisol concentration was significantly higher in the group of  
247 rams where EE was applied compared to the group that experienced probe insertion without  
248 electrical stimulus. There was no difference in blood cortisol concentrations between the probe  
249 without stimulus group and the untreated control group. The sheep in the study were not restrained  
250 in lateral recumbency and the procedure was conducted in the rams' home pen. With the lack of  
251 such external stress factors, the results of this study do indicate EE to be a painful procedure with  
252 the potential to cause elevated cortisol concentration. A study by Ortiz de Montellano et al. (2007) in  
253 bucks reported similar results. Increased cortisol concentration can thus be considered a result of  
254 painful stimuli in sheep and goats but not necessarily in cattle. Similar results between cattle and  
255 sheep were reported concerning aversive behaviour where cattle did not present with "conditioned  
256 suppression" (Rushen, 1986) but sheep did (Molony and Kent, 1997; Palmer, 2005).

257 Although there are variations in the results of trials conducted with EE, regarding the perception of  
258 noxious stimuli, or how animals experience and respond to pain and how to estimate the intensity of  
259 pain, there is evidence to indicate that EE is a painful procedure. The reaction of conscious animals  
260 (rams, bucks and bulls) to electrical stimulation presents with intense muscle contraction, struggling  
261 and vocalisation, all of which are indicative of discomfort (Palmer, 2005). This behavioural data, in  
262 particular vocalisation and learned aversive behavior, supports the notion of a painful procedure. In  
263 addition, epidural anaesthetic has proven to reduce pain responses to EE (Etson et al. 2004).



264 Contributory factors to the pain response associated with EE have been identified, such as the  
265 duration and intensity of the electrical shock (Falk et al., 2001) and operator technique (Palmer,  
266 2005).

## 267 Electroejaculation and stress

268 Selye (1973) states that “everybody knows what stress is but nobody really knows.” Defining stress is  
269 no less complex than defining pain because it is such a highly subjective phenomenon that defies  
270 definition. Where pain elicits an immediate response such as muscle contractions and vocalisation,  
271 stress does not present with immediate clinical symptoms. A short and concise description of stress  
272 is the non-specific response of the body to any demand made upon it (Selye, 1973). Selye (1973)  
273 states very clearly that stress is not anxiety. Stress associated with EE has been extensively studied  
274 (Abril-Sánchez et al., 2017; Palmer, 2005; Stafford, 1995; Ungerfeld et al., 2016; Costa et al., 2018).  
275 As stated earlier, the stress experienced by an animal during EE cannot be ascribed to EE alone.  
276 Zulkifli (2013) states that any disruption of an animal’s homeostasis that requires a response to  
277 maintain its psycho-physiological integrity has the potential to elicit a stress response in the animal.  
278 EE qualifies as a disturbance in the homeostasis of an animal, as it is an artificial procedure designed  
279 by humans and may thus cause stress in the animal on which it is performed (Stafford et al., 1996;  
280 Orihuela et al., 2009). However, the steering of the animals to the handling facility, confinement,  
281 handling and restraint all disturb the homeostasis of the animal and would also have the potential to  
282 elicit a stress response. While this is true, animals become habituated to repeated stimuli of the  
283 same stressors (Grissom and Bhatnagar, 2009), for example weighing of animals. According to  
284 Thompson and Spencer (1966), habituation is a form of simple non-associative learning in which the  
285 magnitude of the response to a specific stimulus decreases with repeated exposure to that stimulus.  
286 Damian and Ungerfeld (2010) have characterised the stress response to EE in frequently  
287 electroejaculated rams (every 15 days since 45 days of age) and concluded that although rams had  
288 been subjected to frequent EE, they constantly suffered from stress. The study characterised stress  
289 by describing changes in heart and respiratory rates and increased cortisol concentrations. The  
290 results suggest that rams did not habituate to frequent EE.

291 Page (2017) coined the term “anticipation stress”, where the Hypothalamus- Pituitary- Adrenal- Axis  
292 (HPA – Axis) is activated as soon as the animals are steered towards the handling chute for a  
293 procedure. Kruger et al. (2015) reported elevated cortisol concentrations in goats at zero minutes  
294 after a “low stressor” procedure such as deworming, confirming the theory of anticipation stress.  
295 Stress is thus a continuum of experience that presents in different degrees according to the  
296 procedure performed. Short-term mild stress results in short term physiological responses on the

297 part of some animals with slight or no behavioural adjustment. Moderate stressors evoke  
298 behavioural adjustment with physiological recovery and adaptation by the animal. Severe stressors  
299 or distress is a state where the animal is unable to adapt to the stressor and the animal may even  
300 exhibit maladaptive behaviour (Bayne, 2000).

301 According to Whitlock et al. (2012) increased vocalisation in bulls after the first electrical stimulus  
302 resulted in a peak in plasma cortisol concentration after ten minutes. The mean plasma cortisol and  
303 progesterone concentration following EE were higher in EE bulls compared to control bulls with a  
304 rectal probe inserted with no electrical stimulus or no manipulation. The cortisol concentrations in  
305 the EE bulls returned to the basal level concentrations after 45 minutes. This correlation between  
306 the electrical stimulation of EE and cortisol concentration motivates the argument suggesting that EE  
307 causes stress. There was, however, no difference in the mean plasma substance P concentration  
308 between EE bulls and control bulls. Substance P is a neurotransmitter that stimulates pain receptors  
309 in the body. If substance P levels are elevated, the perception of pain may be exaggerated (Lubbers  
310 et al., 2009). This lack of difference between the EE bulls and control bulls indicates a lack of pain  
311 associated with nociception and may explain the lack of aversive behaviour in bulls exposed to  
312 repeated EE procedures as stated by Palmer (2005).

313 In a similar study by Costa et al (2018) with rams, the serum cortisol concentration peaked 20  
314 minutes after the application of an electrical stimulus and only returned to the basal level  
315 concentration after 60 minutes. The total plasma protein concentration increased significantly from  
316 the beginning of the electrical stimulus up to 120 minutes, after which it returned to baseline  
317 concentrations. Costa et al. (2018) suggest, based on the results of previous studies on rams  
318 (Orihuela et al., 2009), the use of anaesthesia prior to EE in order to reduce the observed pain  
319 symptoms.

320 As mentioned earlier, any disturbance of the homeostasis has the potential to stimulate the HPA-  
321 Axis with subsequent release of stress hormones such as cortisol. Cortisol is a gluco-corticoid  
322 hormone that may compromise the immune system and is implicated as contributing to the early  
323 stages of the pathogenesis of pneumonia caused by *Mannheimia haemolytica* (Pillai et al., 2018;  
324 Taylor et al., 2010; Rice et al., 2007). Almost all studies on EE associate the procedure with stress.  
325 There are no practical alternatives for semen collection other than AV or EE. Refinement of the  
326 equipment and technique should be investigated and regulated as a practical solution.

## 327 Alternatives to EE

328 The animal welfare implications associated with EE are a concern raised by retailers and consumers.  
329 Therefore, alternatives to EE or refining of the EE technique, especially in small stock, should be  
330 explored and developed, not only for practical application, but also to carry the approval of animal  
331 welfare organizations. (Costa et al., 2018; Damian and Ungerfeld, 2010; Falk et al., 2001; Eton et al.,  
332 2004).

### 333 *Artificial vagina*

334 The use of artificial vagina (AV) as a method to collect semen existed long before the invention of  
335 the electroejaculator (Salisbury et al., 1978). The procedure requires the active participation of the  
336 male animal and closely approximates a natural breeding situation with the advantage that it also  
337 allows for libido and mating ability evaluation. The procedure, however, has the disadvantage that  
338 the bull, buck or ram needs to be trained. This is time-consuming and animals unaccustomed to  
339 handling do not train well to the procedure (Palmer, 2005). The reticence of a bull to mount a cow in  
340 the presence of humans led to the development of the internal artificial vagina (Barth et al., 2004).  
341 The internal AV requires the restraining of a cow and the bull introduced to serve her. The success  
342 rate of obtaining semen with this technique is however not as high as that achieved with EE (Barth et  
343 al., 2004). This technique is also only applicable to cattle and not small stock, for which alternatives  
344 or refinement is required. The artificial vagina technique used in small stock involves additional  
345 potential stressors as mentioned before i.e. isolation, restraint and human interaction. Restraint is a  
346 distinct disturbance of the homeostasis and has the potential to cause stress and discomfort to the  
347 animal. The intention of relieving stress to the ram or buck with the use of AV is not a practical  
348 solution because the stressor is transferred to the ewe or doe.

### 349 *Transrectal massage*

350 Transrectal massaging whereby the thumb and little finger are held in direct apposition with the  
351 ampulla, and gentle motion anterior to the caudal is applied has had success in obtaining a semen  
352 sample. In a trial by Palmer (2005), semen was obtained in 280 of 288 attempts compared to 285 of  
353 288 attempts for EE. The semen samples obtained this way had significantly fewer motile ( $63.5 \pm$   
354  $24.0\%$  versus  $74.5 \pm 16.8\%$ ) and live ( $78 \pm 14.8\%$  versus  $83.1 \pm 11.7\%$ ) sperm than those obtained by  
355 EE. This technique is only applicable to bulls with a docile nature and the quality of the sperm is not  
356 as good as compared to EE and is thus not a practical alternative to EE

### 357 *Drugs to facilitate semen collection by EE*

358 In the continued exploration of alternatives to EE, two experiments investigated the effect of  
359 oxytocin (a peptide hormone associated with sexual reproduction) and cloprostenol (a synthetic

360 analogue of prostaglandin), on the time and number of EE stimuli required to cause semen emission  
361 (Palmer et al., 2004). Cloprostenol had no effect on the time to semen emission, sperm output or  
362 the number of electroejaculator stimuli required (Palmer et al., 2004). Oxytocin decreased the time  
363 to semen emission and tended to decrease the number of EE stimuli to semen emission but it did  
364 not increase the number of sperm in the ejaculate (Palmer et al., 2004). The results of oxytocin show  
365 some potential in refining the EE technique, but it is a scheduled drug for use by veterinarians only.  
366 If the EE procedure can be regulated to allow properly trained persons only, then the use of Oxytocin  
367 may be explored further.

### 368 *Anaesthesia/sedatives*

369 The use of anaesthetics and sedatives in an attempt to reduce the discomfort and stress associated  
370 with EE has been extensively studied for decades (Mosure et al., 1998; Etson et al., 2004; Falk et al.,  
371 2001; Palmer, 2005; Stafford 1995; Pagliosa et al., 2015). According to Palmer (2005), lidocaine  
372 epidural anaesthesia had no adverse effects on penile protrusion or semen emission with less  
373 elevation in serum cortisol and progesterone. The heart rate of bulls receiving lidocaine epidural  
374 anaesthesia prior to EE was also lower than untreated bulls. The reduction in pain attributable to  
375 lidocaine epidural anaesthesia was, however, not significant (Mosure et al., 1998; Falk et al., 2001;  
376 Etson et al., 2004). Caudal epidural anaesthesia requires appropriate clinical training and practical  
377 experience far more advanced than that required for EE, and may only be performed by  
378 veterinarians. The harm versus benefit of lidocaine epidural anaesthesia in an attempt to relieve  
379 discomfort during EE is questionable, with the inconvenience associated with administering the  
380 anaesthetic outweighing the benefits of the procedure (Palmer, 2005). Abril-Sánchez et al (2018)  
381 studied the use of anaesthetics and sedatives to reduce the potential of stress associated with EE.  
382 The use of sedatives is regarded as having a lower risk than anaesthetics.

383 Goat bucks were divided into three treatment groups:

- 384 (i) EE in untreated control
- 385 (ii) EE under sedation
- 386 (iii) EE under general anaesthesia

387 They were compared to one another regarding their physiological and behavioural responses  
388 associated with stress and pain, as well as the semen quality of the three treatments. The results  
389 showed that the untreated bucks vocalised more frequently when compared to sedated or  
390 anaesthetised bucks. The rectal temperature, heart rate, total protein, albumin and haemoglobin  
391 concentrations were higher in the untreated bucks compared to the sedated or anaesthetised bucks.  
392 The serum cortisol concentration increased after EE with no differences between procedures. The

393 study concluded that general anaesthesia and sedation decrease stress and probably also pain  
394 during EE with an added advantage of improved quality of semen in sedated bucks (Abril-Sánchez et  
395 al., 2018). Similar to the caudal epidural anaesthesia technique, the sedation and general  
396 anaesthesia of animals requires specific skills, expertise and training and can only be performed by  
397 registered veterinary professionals. The use of anesthetics or sedatives in an attempt to alleviate the  
398 pain or stress involved with EE is impractical and does not warrant any further investigation.

### 399 *Modified EE technique*

400 Baiee et al. (2017) explored a modified EE technique with the aim to reduce the discomfort  
401 associated with the normal EE procedure. The trial involved three stages of graduated electrical  
402 stimulation. The results of the modified technique showed that intensive muscle spasms, struggling  
403 and arched backline posture were reduced, as well as the time of penile protrusion and semen  
404 emission compared to the normal EE method. The time to collect the sample was the same for both  
405 methods and there was no difference in the semen parameters such as sperm volume, motility,  
406 morphology, viability and concentration. The authors concluded that the modified EE procedure  
407 with the gradation of electrical stimulation into three stages could reduce the discomfort associated  
408 with the normal EE method. The procedure as tested by Baiee et al. (2017) is a practical and drug  
409 free refinement of the EE technique. It does require proper training of the operators and the use of  
410 standardized equipment, further emphasizing the need for regulation of EE equipment and training  
411 of operators. The modified technique was performed on bulls only and further studies should be  
412 explored on small stock too.

### 413 *Equipment & Operator factors affecting pain response during EE*

414 Etson et al. (2004) investigated the influence of probe type used during EE, with and without  
415 epidural lidocaine anaesthesia, on blood cortisol and progesterone concentrations in bulls. The  
416 results indicated the use of segmented over non-segmented probes was not effective in reducing  
417 pain during EE (Etson et al., 2004). The rest interval between stimuli influenced the level of pain  
418 response observed (Stafford 1995). A ten second stimulus rest interval significantly reduced the  
419 physical response of rams to EE compared with a stimulus rest interval of five seconds (Stafford,  
420 1995). Palmer (2005) recommends applying stimuli based on close monitoring of bull's behaviour  
421 during EE. This involves removal of the stimulus for 0.5 - 1 seconds once the bull starts reacting by  
422 struggling, muscle spasms and arched back. The voltage is then steadily increased with each  
423 successive stimulation of one to two seconds. The correct placement and orientation of the probe  
424 are also important to minimise the pain response to EE (Stafford, 1995).

425 The variations and differences reported between studies on pain and stress indicators caused by EE  
426 may stem from variations in operator technique. Regulation with regard to standardised equipment  
427 and properly trained operators is practically implementable and if the technique is applied correctly,  
428 it may cause a reduction in pain responsive behaviour associated with EE.

429

### 430 ***Electro ejaculation and Role Players in South Africa***

431 Animal and animal product production in South Africa must conform to international standards with  
432 regard to animal welfare if it wants to be competitive on the international market. To achieve this it  
433 is necessary for South Africa to introduce specific laws and regulations to the use of semen collection  
434 techniques. Since EE involves animals, veterinary skills are required to ensure suitability, proper  
435 health, restraint and monitoring animal responses for signs of discomfort. The South African  
436 Veterinary Council (SAVC) should thus introduce such laws and regulations in consultation with all  
437 relevant role players such as South African Veterinary Association (SAVA), Ruminant Veterinary  
438 Association of South Africa (RUVASA), National Society for the Prevention of Cruelty to Animals  
439 (NSPCA), South African Council Natural Scientific Professions (SACNASP), South African Veterinary  
440 Semen and Embryo Group (SAVSEG), South African Semen and Embryo Group (SASEG) and the  
441 Livestock Welfare Coordinating Committee (LWCC). Current artificial practitioners, with or without  
442 SAVC registration, should either register as a veterinarian (Assisted Reproduction Professional) or as  
443 a para-veterinary professional, (Assisted Reproduction Operator), with a regulatory body such as the  
444 SAVC. Training facilities for any of the assisted reproduction procedure courses must also have an  
445 accredited syllabus to be registered with and quality assured audited by SAVC.

### 446 **Conclusion**

447 Semen collection is a procedure practiced worldwide to obtain semen samples. Various techniques  
448 exist for obtaining semen samples with varying degrees of discomfort to the animal, which means  
449 that the procedures must conform to animal welfare standards.

450 Research results have proven that EE is the preferred technique for obtaining a semen sample. It is a  
451 quick and effective method that can be applied to animals without prior training, only involves the  
452 male animal and provides a semen sample of good quality. It is an aversive procedure with the  
453 potential to cause pain and stress in the animal. However, if the technique, especially the electrical  
454 stimulation, is refined and correctly applied by trained operators and conducted with proper  
455 equipment, there should be less pain and discomfort associated with EE. Despite the concerns  
456 raised by animal welfare groups, EE should remain as an ethically acceptable technique for obtaining

457 a single semen sample and regular semen samples, provided that the procedure is regulated  
458 essentially as a veterinary procedure only to be performed by properly trained operators.

459 The most common and practical alternative to EE is AV. The procedure is not painful and may be less  
460 stressful to the trained male animal. The technique, however, requires restraint of the female  
461 animal and the welfare concerns shift from the male to the female animal and it is thus not a  
462 practical solution. With trained male animals and a female animal habituated to act as a “dummy”  
463 for the AV technique, the technique could be considered the preferred option for regular semen  
464 collection. Similar to the EE technique, the AV technique should also be regulated to be performed  
465 by properly trained operators only.

466 Other alternatives such as the use of sedatives or anaesthetics are not recommended for farm  
467 animals under normal conditions because of various risks involved. They are also impractical and do  
468 not justify any further investigation.

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