

# RUM\_009 Semen collection by electro-ejaculation in the bull

## I. OBJECTIVE

To describe a safe and reliable protocol for semen collection by electroejaculation in the bull.

### **II. DEFINITIONS**

**Competent** - "the consistent application of knowledge and skill to the standard of performance required regarding the care and use of animals. It embodies the ability to transfer and apply knowledge and skill to new situations and environments." (as per, Australian code for the care and use of animals for scientific purposes, 2013)

### **III. COMMENTS / RECOMMENDATIONS**

- This procedure should be performed:
  - by a registered veterinarian, or under their directed supervision,
  - no greater than once a week for teaching purposes (in an individual animal),
  - as part of a holistic assessment of the Breeding Soundness Examination (BSE)
- Where appropriate, less invasive alternatives should first be considered, e.g. bulls that are responsive to and will serve an artificial vagina, or bulls that are trained to /will ejaculate from transrectal palpation, with focus on massage of the ampullae.
- If evidence of rectal bleeding is observed, an animal's use in that session should be considered complete.
- As routine, any health concerns (such as rectal bleeding or excessive straining) must be managed as per veterinary advice.
- Relative to animal ethics applications, when using this SOP, the following should be described in the individual ethics application: duration and frequency of animal use, and any variation to this SOP.

### **IV. EQUIPMENT**

- Obstetric lubricant
- Transrectal palpation glove
- Electro-ejaculator, and appropriately sized diameter rectal probe (e.g. 50, 75 or 90mm)
- Semen collection tubes
- Collection funnel, attached to an extended handle
- Scissors, or battery-powered clippers to trim preputial hair
- Suitable infrastructure for animal restraint (e.g. cattle crush with option for head bale and rump support)

### V. PREPARATION

- Ensure the animal is appropriately restrained for the procedure; generally the minimum requirements will include a cattle crush with a rump support bar (or "kick gate") is required. The use of straps to hold the animal upright during the electroejaculation procedure is not acceptable.
- Ensure the electro-ejaculator and all associated equipment is set up and ready to go (this includes ensuring the electro-ejaculator is sufficiently charged, and the batteries are operating effectively)

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## **VI. PROCEDURE**

#### Rectal palpation in preparation for semen collection

- 1. Remove any bulky items (e.g. watches and jewellery) and ensure fingernails are clipped to an appropriate length (on the arm that will be used for the palpation). This will help to reduce the risk of trauma to the rectal mucosa.
- 2. Apply a rectal glove and ample lubricant to the palpating-arm.
- 3. Gently, and initially only to the level of the wrist, insert the gloved and well-lubricated palpating arm into the rectum.
- 4. Using the non-palpating hand, the tail may be clasped to position it out of the way.
- 5. Observing contractions and the animal's response, gradually and gently extend the arm further into the rectum. Do not "push through" the peristaltic contractions but rather let them slide over your arm.
- 6. Assess the volume of faeces within the rectum. Excessive volumes of faeces should be gently directed to the anus and expressed without completely withdrawing the arm. Withdrawing the arm may allow aspiration of air into the rectum ("air sucking"), and subsequent rectal "ballooning".
- 7. If the rectum contains excessive volumes of air (ballooning of rectum) this may be reduced by grasping the most caudal contracted peristaltic ring and gently pulling it caudally. This step may only be performed by a competent and experienced operator.
- 8. Identify the relevant internal reproductive anatomy (prostate, seminal vesicles and ampulla).
- 9. Perform transrectal massage of the ampullae for 10 60 seconds; this massage is intended to sexually excite the bull, "milk" spermatozoa and seminal fluid into the urethra and cause relaxation of the anal sphincter prior to probe insertion. This is believed to facilitate semen emission during electro-ejaculation and potentially reduce the need for unnecessary electrical over-stimulation. Rectal massage may also relax the retractor muscles of the penis resulting in extrusion of the glans penile from the prepuce a useful technique for assessing the penis.

#### Electroejaculation and semen collection

- 10. Remove your arm from the bull's rectum, and while doing so insert the largest rectal probe that can be comfortably accommodated (usually 75mm or 90mm diameter). The probe should be inserted with ample lubricant applied, the longitudinal electrodes on the probe facing ventrally, and the electro-ejaculation machine turned off.
- 11. In preparation for semen collection hairs and dirt around the preputial orifice can be trimmed and wiped clean, respectively. A non-spermicidal semen collection tube should be connected the rubber collection cone at this time in readiness to capture seminal emissions it is important that the collection tube remains at body temperature and free of particulate matter and water (condensation).
- 12. With an assistant holding the rectal probe in place (horizontally) and the operator positioned alongside the bull, monitoring the animal's behaviour, electro-stimulation may commence. It is important to start at the lowest power setting on the machine.
- 13. While there is a great deal of individual variation in the sensitivity (and response) to electrostimulation, stimulus should typically be delivered in a rhythmic series of gradually increasing intensity, carefully observing, and adapting to, the bull's reaction. Generally, the stimulus is turned on for ~2 seconds, then off for 1-2 seconds (Bos taurus cattle), or stimulus is on for ~2 seconds, then off for 3-5 seconds (Bos indicus cattle).
- 14. It is important to note that many bulls go through a period of agitation in the early stages of the procedure. These animals generally need to be stimulated more slowly and gradually until the operator is comfortable that the bull is responding predictably (in terms of muscular contraction to electrical stimulus being provided).
- 15. After some initial stimulation the animal's behaviour will become somewhat predicable and "controlled". This is recognised by a characteristic movement of the bull, gently rocking back and forth, in response to the

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stimulus. At this point the operator may open the lower side door of the crush (if present) to gain access to the penis for the collection of semen.

- 16. Once the operator has "control" of the bull's muscular response, they can continue to increase the level of the stimulus until the bull protrudes the penis and/or commences to ejaculate seminal fluid typically the bull will ejaculate a clear pre-seminal fluid initially (this may be discarded from the analysis, unless there is concern that the bull has an underlying pathology).
- 17. Following this, the level of stimulus may be increased more rapidly to the point of ejaculation which is typically indicated by an emission of a white (milky to creamy) coloured ejaculate.
- 18. If the bull fails to ejaculate on the first attempt repeat transrectal massage of the ampullae may be performed to obtain a diluted, less concentrated semen sample for analysis. A repeat attempt at electroejaculation may also be made after a 5 to 15-minute interval.
- 19. Normally one sperm rich ejaculate of 1-2 mL is sufficient for semen analysis but if the bull is being used for custom collection for chilled storage or cryopreservation, then further volume or a repeat of the procedure may be required.
- 20. In some cases, if the bull has not ejaculated for a prolonged period, it may be necessary to collect a number of ejaculates to expel sperm from the reproductive tract that may have degenerated in the cauda epididymis or ampullae. This can be judged readily by the operator via analysis of the semen sample using a microscope set up at the "crush-side".
- 21. Once semen collection has been completed, the machine should be turned off and the rectal probe gently removed from the animal.
- 22. Before cleaning the probe, for use with the next animal, inspect it for any signs of blood. If blood is observed veterinary advise should be sought to guide management (note: scant amounts of blood may not require any intervention, other than veterinary examination and assessment).
- 23. Make a clear record of the procedure, your assessment, and note any other relevant findings.
- 24. Release the animal from the restraint and monitor it for signs adverse events (e.g. weakness, lethargy, bleeding from the rectum). If an unexpected adverse event(s) occurs take immediate action as outlined on the <u>animal ethics webpage</u>.

Version #	Reviewing AEC (note: all other relevant AECs ratify the approval)	AEC Review Date	Approval To Date
1	PCA	20/07/2022	20/07/2025

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