Guidelines for using the bull breeding soundness evaluation form

P.J. Chenoweth,a F.M. Hopkins,b J.C. Spitzer,c R.E. Larsend

aSchool of Animal and Veterinary Sciences, Charles Sturt University, Waga Waga, NSW, Australia; bCollege of Veterinary Medicine, University of Tennessee, Knoxville, TN, USA; cSpitzer Ranch, Fair Play, SC, USA; dSchool of Veterinary Medicine, St. George’s University, Grenada, West Indies

Introduction
The breeding soundness evaluation (BSE) is a relatively quick and economic procedure for screening bulls prior to sale or use. It is also a useful tool in infertility investigations. Proven benefits from its application include direct effects on herd fertility and indirect effects via genetic relationships with other fertility traits in both the male and female. Although the BSE classifies bulls into categories which generally perform as predicted, the examination is limited in its ability to consistently predict individual bull fertility. Reasons for this include:
   1. Fertility is complex and influenced by both male and female traits as well as by extraneous factors.
   2. The BSE is a relatively quick and simple screening procedure which does not attempt to comprehensively assess all aspects of male fertility
   3. Both knowledge and understanding keep increasing and changing.

Keywords: Breeding soundness evaluation, bull, semen quality, physical examination

General procedures
The primary mission of the bull is to efficiently impregnate all available females as early in the breeding season as possible. For this he needs good eyesight and musculo-skeletal conformation as well as the necessary reproductive equipment and sex drive to produce and deliver sufficient numbers of fertile spermatozoa when and where necessary.

The BSE consists of the following steps:
   1. Physical examination
   2. Reproductive examination (including scrotal circumference measurement)
   3. Collection and examination of semen

In addition, a libido/mating ability test may be included, as may special tests for diseases (e.g., campylobacterosis or trichomoniasis). These procedures may add predictive value to the assessment process and may be specifically indicated in some situations, but they are not generally part of the routine BSE.

Identification and ownership of bulls
Permanent and unique identification of each bull is important as well as the establishment of ownership. The form gives provision for breed, age, identification numbers (including brands, tattoos and eartags) and previous history. It is good procedure to identify all samples, slides and work-sheets with the bull identification number. This section includes owner information as well as case number and details of any previous tests.

Body condition and other body measurements
Body condition is relevant to reproductive assessment. Excessively fat or thin bulls can have semen, libido and mating ability problems. Poor body condition may also reflect health problems apart

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from poor nutrition. Provision is made for bulls to be categorized into general categories (thin, moderate, good, obese) or for scoring systems to be applied as developed for either beef (1-9) or dairy (1-5) cattle. Pelvic size in bulls is related to pelvic size in their daughters, an important consideration in dystocia management. Pelvic size measures (height, width, area) can be recorded on the form. The height and width are usually measured by manual insertion of a pelvimeter and multiplied together to give the approximate area.

While no specific slot was provided for bull weight, this measure can be inserted in any of the available blank areas on the form.

Structure, conformation and movement

The bull should be observed moving free of restraint in order to observe gait and movement. Once in the chute, closer attention can be paid to the limbs, joints and claws. The bull should have good, symmetrical musculo-skeletal conformation and he should walk smoothly. Asymmetrical or overgrown claws as well as swellings over the lower limb joints are common abnormalities.

NB: Bull examinations should be undertaken with due regard for the safety of both the examiner and the bull.

Physical and reproductive examination

A thorough physical examination is necessary since the bull must be able to see, eat and move about without discomfort to be a functional breeder. A systematic approach should be employed which emphasizes those aspects most important for breeding success.

The bull’s eyes should be examined for impaired vision. Common problems here include corneal defects, infectious keratoconjunctivitis (“pink eye”), scarring, dermoids and squamous cell carcinoma. Examination of the bull’s teeth allows the examiner to verify age and ensure that dentition is adequate for foraging. The bull’s feet are a common source of problems. Overgrown claws in need of trimming, malformed claws and interdigital fibromas are commonly identified problems. The sheath of the bull should be inspected and palpated. Problems here might include inflammatory exudates, trauma, fibrosis, abscessation and scarring. The penis, palpable within the sheath, should be symmetrical, freely moveable, and have no abnormal masses associated with it. Common problems include abscesses, adhesions, fibropapillomas and hematomas.

Scrotal circumference should be evaluated. Significant differences in the size and shape of either testicle should be cause for further evaluation; a disparity of more than 25% in either testicle should be regarded with suspicion. The most common cause of marked scrotal asymmetry is unilateral testicular hypoplasia although a variety of congenital, traumatic or infections conditions may be implicated. Testicles should be palpated for hardness and resiliency; a normal testicle should be firm and resilient (similar to a new tennis ball). The epididymides should be palpated. Problems here include inflammation, fibrosis, abscessation, hypoplasia or aplasia. To complete the examination of the external genitalia, the penis of the bull should be exteriorized. Commonly encountered problems include a persistent penile frenulum, hair rings, fibropapillomas, infections, trauma and scaring. Per-rectal examination of the bull is an essential step so that pelvic structures, particularly the ampullae and vesicular glands, can be evaluated. Infections, inflammation and adhesions can occur in both areas as can aplasia/hypoplasia.

Scrotal circumference

Scrotal circumference (SC) in bulls is related to sperm production, sperm quality and the age at puberty of related females. It is also fairly highly heritable, at least in beef bulls. The measurement of SC is simple and highly repeatable and does not require expensive or highly technical equipment to perform. For these reasons, it is an important component of the BSE.
To measure SC, a tape is applied snugly around the greatest circumference of both testicles which should be pushed to the bottom of the scrotum. A good procedure is to take two measurements to ensure accuracy, with two consecutive measurements not varying by more than 1.0 cm.

Scrotal circumference thresholds

<table>
<thead>
<tr>
<th>Age (months)</th>
<th>SC (cm)</th>
</tr>
</thead>
<tbody>
<tr>
<td>≤15</td>
<td>30</td>
</tr>
<tr>
<td>≥15 - ≤18</td>
<td>31</td>
</tr>
<tr>
<td>≥18 - ≤21</td>
<td>32</td>
</tr>
<tr>
<td>≥21 - ≤24</td>
<td>33</td>
</tr>
<tr>
<td>&gt;24</td>
<td>34</td>
</tr>
</tbody>
</table>

Semen collection

Semen can be collected from bulls by a variety of means including per-rectal massage, the use of an artificial vagina and by electroejaculation. The latter method is the one most commonly employed with range-type bulls.

Electroejaculators and probes

Commercially available electroejaculators are available with power being provided by AC current, by internal re-chargeable batteries, or by 12 volt automobile batteries. Electroejaculation requires stimulation of pelvic nerves controlling not only emission of semen into the penile urethra but also those controlling erection and ejaculation. Newer probe designs have full-length longitudinal electrodes which stimulate all functions simultaneously.

Preparation and stimulation

The bull’s rectum should be emptied of feces before the probe is inserted. The lubricated probe is inserted so the anal sphincter closes behind the main body of the unit. It is helpful to determine the lowest current level at which the animal first shows an obvious physical response. This initial response may be subtle, e.g., a twitch of the tail, a tightening of the anal sphincter, or a tensing of the gluteal muscles. This stimulus “threshold” provides a starting point for subsequent stimulations which should be conducted with a smooth routine to which the bull can easily adapt.

With *Bos taurus* breeds, a typical approach is to deliver a smooth increase in probe current from zero to the desired level over a duration of one to two seconds, followed by a more rapid reduction to zero current and a rest period of approximately one second before the next stimulation. Once the bull is settled into the routine, five to seven stimulations are given at each succeeding voltage step until erection and ejaculation occur. For machines that do not have separate voltage and current controls, the same stimulation pattern is employed except that the single control is used to generate incremental increases in probe voltage and current until ejaculation occurs. During the early stages of stimulation and erection, clear seminal plasma is often passed which is not generally collected. When the ejaculate turns cloudy, the subsequent jets of semen are collected. It is important to continue stimulation until the ejaculate starts...
to become clear again. Failure to proceed to this point can lead to errors of interpretation in the spermiogram as the initial portion of the ejaculate may contain large numbers of degenerating spermatozoa, especially in bulls which have been sexually quiescent for some time. For the same reason, if an ejaculate shows substandard motility in the absence of an obvious physical cause, the collection of a second sample within a short period of time (e.g. five to ten minutes) can often result in improvement.

With *Bos indicus* bulls this stimulation routine is often unsuccessful in eliciting both erection and ejaculation. A more gentle stimulation pattern incorporating longer rest periods can sometimes improve ejaculation success; however this still may not be accompanied by erection so that semen is voided through the preputial lumen. The voiding of semen within the prepuce is a problem as it can increase semen contamination with preputial debris. Success in exteriorizing the penis during electroejaculation can sometimes be obtained by pushing on the sigmoid flexure with a clenched fist from the rear of the bull; this technique can also be successful with young bulls. Inability to exteriorize the penis means that this organ cannot be properly examined. *Unless the penis and inner prepuce are extended and examined, the bull cannot be properly classified.*

Variations on these stimulus patterns occur with different machines and operators. With difficult bulls some experimentation might well be necessary. In all situations, however, the welfare of the animal is paramount and stimulation should be discontinued if either undue stress is being caused or physical injury to the bull might occur.

**Collection devices**

Semen is collected into a prewarmed insulated or jacketed tube through a funnel or cone. All surfaces coming into contact with semen should be warm, dry, and free of spermatoxic agents. Because “cold shock” causes irreversible damage to spermatozoa, efforts to maintain semen at 30-35 °C until the “on-site” evaluation procedures are complete is an important consideration for successful semen assessment.

**Semen evaluation**

**Initial impressions**

Volume, density, and gross characteristics of the ejaculate are not “front-line” BSE assessments because they have not been shown to be related to fertility. Space on the score sheet is, however, provided for recording such information. Likewise, the assessment of spermatozoa concentration is not a routine part of the BSE; measurement of SC provides a better estimate of sperm production in range-type bulls which are subject to infrequent examinations. However, informal recording of such information may help to monitor the success of semen collection and to interpret gross motility estimation.

Other gross characteristics which may be noted include evidence of contamination, hemorrhage or inflammatory material. If the ejaculate contains sufficient purulent material for this to be obvious to the naked eye, then the bull should not be classified as satisfactory at least until a benign cause is found. Debris or contamination from the sheath may be regarded as being less serous unless it represents active infection.

**Motility**

Motility should be assessed microscopically. Two methods of assessing sperm motility are traditionally employed: gross motility (or mass activity) and individual motility (or percent progressive motility). It is good procedure to use both methods as they can differ somewhat in interpretation and precision. With all motility estimates, it is important to protect semen against adverse effects (e.g. cold shock) and do the estimation as soon as possible after semen collection.

Gross motility, or the amount of swirling (or wave motion) present in an undiluted semen sample, is a function of both sperm concentration and individual motility. Under field conditions, gross motility is typically assessed by placing a drop of raw semen on a warmed slide and observing it at 100...
magnifications (10x eyepiece and 10x objective). With the condenser properly adjusted, mass action or “swirl” can be observed in samples which have adequate numbers of motile spermatozoa. The rankings for this estimate are as follows:

<table>
<thead>
<tr>
<th>Mass Activity (Gross Motility)</th>
<th>Rating</th>
</tr>
</thead>
<tbody>
<tr>
<td>Rapid swirling</td>
<td>Very Good (VG)</td>
</tr>
<tr>
<td>Slower swirling</td>
<td>Good (G)</td>
</tr>
<tr>
<td>Generalized oscillation</td>
<td>Fair (F)</td>
</tr>
<tr>
<td>Sporadic oscillation</td>
<td>Poor (P)</td>
</tr>
</tbody>
</table>

The minimum recommended threshold for gross motility is fair (F)

Individual progressive motility of spermatozoa is assessed under a brightfield or phase contrast microscope preferably equipped with a warm stage or other means of preventing cold shock of spermatozoa. Coverslipped specimens are usually examined at a total magnification of 400x. In dense samples (milky or creamy), the sample should be diluted for proper observation of individual spermatozoa. Sodium citrate or skim milk based semen extenders are serviceable diluents; physiological saline (PSS) may be used although readings should not be delayed when it is used. The percentage of active, progressively motile cells is estimated. This procedure takes more practice than does the gross motility estimation, but is probably more accurate in experienced hands. Individual motility ratings are as follows:

<table>
<thead>
<tr>
<th>Percent progressive motility</th>
<th>Rating</th>
</tr>
</thead>
<tbody>
<tr>
<td>≥70%</td>
<td>Very good (VG)</td>
</tr>
<tr>
<td>50-69%</td>
<td>Good (G)</td>
</tr>
<tr>
<td>30-49%</td>
<td>Fair (F)</td>
</tr>
<tr>
<td>≤30%</td>
<td>Poor (P)</td>
</tr>
</tbody>
</table>

The minimum recommended threshold for individual motility is 30%

Observation of the semen sample at 400x can also help to identify the presence of abnormal numbers of other cells (e.g. squamous epithelial cells, inflammatory cells or spheroids) within the sample. The identification of aberrant cellular material can benefit from the staining of the semen smear with modified Wright Giemsa stain, new methylene blue, or other differential blood cell stain, while bacteria are best categorized using a Gram stain.

Morphology of spermatozoa

Morphology of spermatozoa (differential counts of normal and abnormal cells) is assessed either by phase contrast microscopy (by using preparations “fixed” in formol-buffered saline or PSS-glutaraldehyde) or by using brightfield microscopy of stained smears. Common stains used for this purpose include nigrosin-eosin, William’s stain, modified Giemsa, and even India ink. The Society for Theriogenology recommends nigrosin-eosin for its combination of ease and utility. Although this stain is a “supra-vital” stain (i.e., sperm which are “alive” at staining will not absorb the stain while those that are “dead” will partially or completely absorb the red eosin color), here it used for its ability to depict sperm morphology only.

With nigrosin-eosin staining of spermatozoa, the most common method is to mix a fraction of a drop of semen with a drop of background or “negative” stain and spread the mixture over the surface of a
glass slide which is allowed to air-dry. Care should be taken during the smearing process to avoid trauma
to sperm. It is also helpful to vary the thickness of the smear to provide a variety of background densities
to the smear from which an area can be picked for best microscopic examination.

Brightfield microscopy of stained smears is best done at 1,000x with an oil immersion lens. At
least 100 spermatozoa should be observed in different fields and classified for normality or abnormality. Normal sperm should be at least 70% of those counted for the bull to pass the BSE (see below).

In previous BSE systems, sperm abnormalities were classified as being either “primary” or
“secondary” with the underlying assumptions being that primary abnormals (considered to be caused
during spermatogenesis) were more serious than secondaries (caused subsequent to sperm released into
the extragonadal system). More recent knowledge has cast doubt upon these assumptions. In the
meantime a system of using “major” and “minor” abnormalities was created to more accurately reflect
sperm abnormalities for which fertility data were available. It was apparent that the lists of sperm
abnormalities in routine use for both systems were essentially indistinguishable (except perhaps for
proximal cytoplasmic droplets). Thus, as the primary/secondary scheme is widely used at present, this
was retained as the reference point in the present system (see Appendix 1 for categories of abnormalities).
Although total abnormalities only are employed as the threshold in the new BSE system, “primary” and
“secondary” abnormalities can be collated to arrive at this number. The recording of specific
abnormalities (or their category as primary or secondary) can also be useful for the monitoring of bulls
and their progress.

The minimum recommended threshold for sperm morphology is 70% normal spermatozoa

Evaluation categories

Satisfactory
Bulls which equal or surpass the minimum thresholds for SC, sperm motility and sperm
morphology, and which do not show genetic, infectious or other problems or faults which could
compromise breeding or fertility.

Unsatisfactory
Bulls which are below one or more thresholds and which are unlikely to ever improve their status.
Also, bulls which show genetic faults or irrevocable physical problems (including infectious disease)
which would compromise breeding or fertility are included.

Classification deferred
Any bull which does not fit into the above categories and which could benefit from a retest.
Provision is provided for the scheduling of a retest and this is recommended. This category would
include bulls with an “immature” semen profile as well as any bull whose semen is substandard but
considered to be capable of improvement. Also in this category are bulls from which a satisfactory
ejaculate could not be obtained for reasons unknown as well as bulls with treatable problems such as
seminal vesiculitis or footrot. In general, if any doubt exists about a bull fitting into either the satisfactory
or unsatisfactory categories, he should be considered as a candidate for a retest and placed into the
“classification deferred” category.

Veterinarians are encouraged to work with their clients to accept higher standards for bulls than
the “minimum acceptable” standards employed in this BSE system

† It is not uncommon for yearling bulls to exhibit higher levels of certain types of spermatozoal abnormalities which are
associated with immaturity. Such bulls will usually require a second examination before being classified as a satisfactory
potential breeder.
Selected references


Appendix 1

Categories of sperm abnormalities

<table>
<thead>
<tr>
<th>Primary abnormalities</th>
<th>Secondary abnormalities</th>
</tr>
</thead>
<tbody>
<tr>
<td>Underdeveloped</td>
<td>Small normal heads</td>
</tr>
<tr>
<td>Double forms</td>
<td>Giant and short broad heads</td>
</tr>
<tr>
<td>Acrosome defects (e.g. knobbled)</td>
<td>Free normal heads</td>
</tr>
<tr>
<td>Crater-diadem defect</td>
<td>Detached, folded, loose acrosome membranes</td>
</tr>
<tr>
<td>Pear-shape head</td>
<td>Abaxial midpiece</td>
</tr>
<tr>
<td>Abnormal head contour</td>
<td>Distal droplet</td>
</tr>
<tr>
<td>Small and free abnormal heads</td>
<td>Simple bent tail</td>
</tr>
<tr>
<td>Proximal droplet</td>
<td>Terminal coiled tail</td>
</tr>
<tr>
<td>Double bent and coiled tail</td>
<td></td>
</tr>
<tr>
<td>Accessory tail</td>
<td></td>
</tr>
</tbody>
</table>
Appendix 2

**Bull Breeding Soundness Evaluation**

Guidelines Established by Society for Theriogenology
P.O. Box 3007 • Montgomery, AL 36109
Phone 334-205-4666 • Fax 334-270-3399 • www.theriogen.org

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**PHYSICAL EXAMINATION**

<table>
<thead>
<tr>
<th>Physical Examination</th>
<th>Score</th>
</tr>
</thead>
<tbody>
<tr>
<td>Body Condition Score</td>
<td>1, 2, 3, 4, 5, 6, 7, 8, 9</td>
</tr>
<tr>
<td>Pubic Hair</td>
<td>Weak, None, Abnormal</td>
</tr>
<tr>
<td>Testes, Spermatic Cord</td>
<td>Normal, Abnormal</td>
</tr>
<tr>
<td>Scrotal Circumference (cm)</td>
<td></td>
</tr>
</tbody>
</table>

**SEMINAL EXAMINATION**

<table>
<thead>
<tr>
<th>Semen Characteristics</th>
<th>Ejaculate 1</th>
<th>Ejaculate 2</th>
</tr>
</thead>
<tbody>
<tr>
<td>Motility</td>
<td>Good, Normal (%)</td>
<td></td>
</tr>
<tr>
<td>% Normal Cells</td>
<td></td>
<td></td>
</tr>
<tr>
<td>% Primary Abnormalities</td>
<td></td>
<td></td>
</tr>
<tr>
<td>% Secondary Abnormalities</td>
<td></td>
<td></td>
</tr>
<tr>
<td>WBC, RBC, Other</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

**CLASSIFICATION**

Interpretation of data resulting from this examination would indicate that on this date, this bull is:

- Satisfactory potential breeder
- Unsatisfactory potential breeder
- Classification Deferred

Re-examination recommended: ____________

Signed: ____________________________

Clinic: ____________________________

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Bull breeding soundness evaluation form available to members of the Society for Theriogenology, P.O. Box 3007, Montgomery, AL 36109.