

Animal &
Grassland Research
and Innovation
Programme



Examination of Bulls for Breeding Soundness

An Illustrated Guide

CONTENTS

Preface and Acknowledgements	3
1.0 Introduction	5
1.1 Facilities Required for Carrying Out Pre Breeding Examination (PBE)	5
1.2 Equipment Required for PBE	6-7
1.3 Preparation of Field Lab	7
2.0 Carrying Out a Bull PBE	8
2.1 SECTION 1 –PHYSICAL EXAMINATION	8-23
2.2 Musculoskeletal System	9-10
2.3 Examination of Scrotum and Contents	10-14
2.4 Scrotal Circumference	14-16
2.5 Examination of Sheath and Prepuce	16-18
2.6 Examination of Penis	18-21
2.7 Examination of Internal Accessory Sex Glands	21-23
3.0 SECTION 2- SEMEN EXAMINATION	24-43
3.1 Collection Methods	24-28
3.2 Assessment of Semen Sample	28-30
3.3 Sperm Morphology Assessment	31-32
3.4 Identification and Significance of Abnormal Sperm	33
3.5 Common Sperm Abnormalities	34-42
4.0 SECTION 3- ASSESSEMENT OF MATING ABILITY	44
5.0 SECTION 4- CLASSIFICATION	45
6.0 Summary and Further Reading	46
APPENDIX 1 - The BCVA Bull Pre-Breeding Certificate	48-49
APPENDIX 2 - PBE Recording Form	50-51
Overview of Pedigree Bull Production - Is it conducive to good fertility?	52
<i>Doreen Corridan</i>	
Problems with bulls in Artificial Insemination Centres	56
<i>Bernard Eivers & Ciara O'Meara</i>	
Identifying Problem Bulls at Farm Level	61
<i>Donal Murphy</i>	
Vetinary Bull Fertility Examination Forms	65





Preface

This guide, initially written by Colin Penny and published in 2010, has been updated as part of the International Bull Fertility Conference - Theory to Practice, held in Westport, Ireland from the 27th to the 30th May 2018. A further 3 chapters have been added dealing with management of bulls in AI studs, rearing of pedigree bulls, and practical aspects of and results from bull breeding soundness evaluations carried out in Ireland. With the introduction of electro-ejaculation (EEJ) as a technique for collecting semen samples from bulls, practitioners have been introduced to a previously neglected area of veterinary practice – routine examination of bulls to assess their fitness to breed efficiently. The completion of a routine examination of bulls prior to sale and prior to breeding is increasingly recognised as a crucial part of herd fertility management and can help avoid significant economic losses due to the use of infertile or subfertile bulls. In North America and Australasia breeding soundness evaluation of bulls has been common practice for decades and different countries have developed various standards and systems for reporting the findings.

The aim of this guide is to provide an illustrated framework to assist practitioners when carrying out breeding soundness examinations. The guide is set out in sections corresponding to the British Cattle Veterinary Association Bull Pre-Breeding Examination Certificate and it is hoped that this will be a useful resource for practitioners involved in the examination of bulls.

Acknowledgements

The organisers of International Bull Fertility Conference - Theory to Practice, acknowledges the willingness and generosity of Colin Penny to update his original publication and make it available to the Conference and the advice and input of several colleagues during the preparation of this manual in particular Prof. Michael McGowan, Prof. Albert Barth, Mr Alastair Smith, Mr Robert Anderson, Ms Lysan Eppink and others. Figures 44, 45, 48, 52 and 55 reproduced courtesy of Dr Eric Taylor. Without the enthusiasm and encouragement of Prof. Michael McGowan during the period he spent at the Royal Veterinary College London, it is unlikely that the process of examination of bulls for breeding soundness would have developed in the UK and Ireland. The conference organisers also acknowledge the contributions of Bernard Eivers, Ciara O'Meara, Doreen Corridan and Donal Murphy for their valuable contributions describing their experiences of dealing with practical aspects of bull fertility.

Michael G Diskin,

Chairperson, Organising Committee.

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Examination of Bulls for Breeding Soundness – An Illustrated Guide

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1. INTRODUCTION

Although the term breeding soundness evaluation is commonly used worldwide to describe the process of examination of breeding bulls the terminology used in this guide will be that used in the BCVA certificate – namely Pre-Breeding Examination (PBE). The aim of a PBE is to try and identify bulls that are potentially unfit for use as breeding bulls to avoid herd fertility losses. Various large studies worldwide have found up to 20% or more of bulls examined during routine screening failed PBE. These failures are for a variety of reasons including physical problems and poor semen quality. Very few bulls are infertile (incapable of impregnating) however many are subfertile. This raises the question of what we define as “normal fertility”. A useful definition of normal fertility has been suggested by Australian workers who state that “Fertile bulls can impregnate (pregnant at day 42 of gestation) by natural service at least 60% and 90% of 50 normal cycling, disease free females within 3 and 9 weeks respectively”. The same authors define sub-fertile bulls as “bulls that can achieve pregnancies by natural service, but not at the rate achieved by fertile bulls when opportunity exists”. Infertile bulls, which are rare, cannot achieve pregnancies.

The purpose of a PBE is to help identify bulls that are infertile or potentially subfertile and a bull certified as “Suitable for Breeding” whilst not guaranteed to be of normal fertility should be of considerably less risk than a bull that has not been examined prior to breeding. The process of PBE is not an exact science however there is no doubt that if all bulls were subjected to a PBE as described in this manual, significant economic losses in beef and dairy herds could be avoided due to bull infertility/subfertility.

1.1 FACILITIES REQUIRED FOR CARRYING OUT PBE

To safely carry out PBE of bulls on farm it is essential that owners provide adequate facilities to safely restrain bulls for the process. Ideally a crush of the type shown in Fig 1 below should be used however as long as the bull can be securely restrained to allow access to the sheath and scrotum then a variety of crushes or races can be used (Fig 2). A rump bar is recommended to reduce risk of being kicked when measuring and palpating the testicles. A non-slip floor surface such as rubber mat will prevent bulls from losing their footing during EEJ. If bulls are restrained in a neck yoke, care should be taken to ensure the cervical spine is not being excessively squeezed which could lead to ataxia when released from the crush. The bull should have the ability to move backwards and forward during EEJ and not be pushed too far forward by a rump bar. In some crushes if the neck yoke is unsuitable for catching bulls it may be preferable to simply halter the bull loosely at the front of the crate during collection.



Figure 1



Figure 2

1.2 EQUIPMENT REQUIRED FOR PBE

Scrotal Tape

A purpose made scrotal measuring tape should be used to ensure accuracy and consistency. A metal tape and plastic tape (Reliabull™) are shown in Fig 3. The Reliabull tape has a device to ensure the same pressure is applied each time when measuring but in general the tape should be pulled tight around the widest part of the scrotum until the skin is indented (Fig 4).



Figure 3



Figure 4

Electro-ejaculator (EEJ) machine or Artificial Vagina (AV)

The EEJ machine used by the author is the Lane Pulsator IV (Lane Manufacturing, Denver) and its use will be described in a later section (Fig 5). When carrying out routine PBE semen is collected using EEJ in most cases however, the use of an AV (Fig 6) will also be described in the semen collection section.



Figure 5



Figure 6

Microscope

For examination of semen a good quality, well serviced microscope is required ideally with x10, x20, x40 and x100 (oil) lenses fitted. Phase contrast condenser with phase lenses x20, x40 make assessment of motility much easier than with standard bright field microscopy lenses. A phase contrast x100 oil lens allows morphology examination using wet mounts which can be advantageous.

Heated Stage

A heated stage is essential to ensure semen motility is not compromised by cooling of samples during examination. Even pre-warmed slides will chill rapidly when placed on a normal microscope stage especially in cold ambient or draughty conditions.

Warm Box and Laboratory Materials

A portable incubator (see Fig 8) acts as a useful container for carrying the miscellaneous equipment required for semen examination. A checklist of things that should be carried in the warm box is:

- ◆ Microscope slides (plain and frosted ends)
- ◆ Cover slips – large size
- ◆ Permanent marker pen (fine tip)
- ◆ Slide carrier boxes
- ◆ Nigrosin-eosin stain
- ◆ Methylene blue stain
- ◆ Micro-pipettes or insulin injection syringes
- ◆ Small vials of sterile saline
- ◆ Semen collection tubes with screw tops



Figure 7

Recording Forms

Standard recording forms should be used to record the findings of each stage of the examination. An example of a suitable form is shown in appendix 2. The information recorded on the forms can then be used to create a PBE Certificate if required. Recording forms should be filed for future reference.

1.3 PREPARATION OF FIELD LAB AREA

The field lab should be set up as close to the bull examination area as possible avoiding a site exposed to draughts, dust and severe cold. All that is needed is a bench with a power point preferably in a sheltered shed or covered area. Setting up a microscope in a draughty exposed area is not ideal as even with a heated stage, cold draught/wind will rapidly cool the equipment and dust will spoil the smear preparation. The warm box should contain all the equipment required for the examination and handling of the semen and prior to going to collect from the bulls a slide should be placed on the heated stage and the microscope prepared for the gross motility examination that will be carried out first (Fig 8).



Figure 8

2. CARRYING OUT A BULL PRE-BREEDING EXAMINATION

The following description of PBE will follow the order as set out in the BCVA Bull PBE Certificate (see appendix 1)

Identification/History

The bull should be identified by recording the official ear tag and age. Studies have shown that most bulls of *Bos taurus* breeds will have reached puberty and normal sperm production by 16 months of age. If bulls are being examined at <16 months of age then the failure rate of PBE will be higher due to immaturity affecting semen quality. In general, the author prefers to wait until bulls are at least 18 months old before carrying out PBE for certification purposes.

A brief history should be taken to ensure there has been no recent illness, lameness or veterinary treatment that could influence semen quality. The reason for PBE should be recorded and details of any previous breeding history.

Disease Testing

The BCVA Bull PBE Certificate does not include any testing for disease status as in the UK this is often certified separately by health declarations required at bull sales. It is the purchaser's responsibility along with their veterinary surgeon to ensure any new bulls entering the herd are tested/vaccinated as required to comply with their farm health plan to ensure the bull does not constitute a biosecurity risk to the farm.

2.1 SECTION 1 – PHYSICAL EXAMINATION

The aim of this part of the PBE is to identify any physical abnormalities that could compromise the bull's fertility or give rise to heritable defects in its progeny.

Body Condition Score

This is assessed on a five point scale with 1 being very thin and 5 obese. Bulls that are in BCS 2 or less should be deemed unsatisfactory in this section as they are unlikely to be able to perform adequately during an intensive breeding period. Bulls that are obese may suffer from poor semen quality due to deposition of fat in the scrotum leading to thermo-regulation problems and this will become apparent in the next stage of the examination when semen is assessed.

General Clinical Examination

A PBE is not intended to be a full clinical examination however for certification or insurance purposes the heart and lungs should be auscultated to eliminate the presence of any abnormalities. The eyes should be examined for lesions that could compromise vision such as cataract, corneal opacity, or carcinoma. The incisor/dental pad alignment should be checked for evidence of any significant pro/brachygnathia.

2.2 MUSCULOSKELETAL SYSTEM

Many bulls are culled prematurely due to problems with the musculoskeletal system so it is critical that problems with the feet or legs are identified if possible at PBE to prevent these bulls being selected as sires. Bulls should be observed walking on a smooth level surface to check for evidence of lameness and the limbs carefully inspected for any conformational defects. Although some conformational defects may not be causing lameness on the day of examination, many have a hereditary component or will lead to lameness at a later date and thus will render bulls unsuitable for certification. Some examples of common musculoskeletal problems that can be identified during bull PBE are shown below.

Post-Legged

Post-legged conformation (Fig 9) is associated with an increased risk of lameness due to hock, stifle or hip joint pathology and young bulls showing this conformation should be avoided. Bulls with extreme post-legged conformation should not be certified as suitable for breeding and the reason for failing the physical examination should be noted in the comments section.



Figure 9



Figure 10

Sickle Hock

The opposite extreme – sickle hock (Fig 10), will lead to collapsed heels and overgrown claws with a tendency for foot lameness and thus bulls with extreme sickle hock are unsuitable for certification.

Valgus Deformity of Forelimbs

Young bulls with obvious valgus deformity will have a tendency to develop abnormal claw overgrowth of the fore-feet and have ongoing problems (Figs 11-12).



Figure 11



Figure 12

Swollen Hocks (tarsitis)

Swollen hocks due to excessive synovial fluid are not uncommon in intensively reared young bulls less than 2 years old. Distension of the hock joint may be caused by osteochondrosis which if mild may not be causing any lameness (Fig 13). Progression to osteochondritis dissecans (OCD) is possible in some bulls however if distended hock joints are present in the absence of lameness then the bull could pass the musculoskeletal examination but the swelling should be noted in the comments section. Bulls with swollen hocks due to OCD are likely to be lame and will be deemed unsuitable for breeding (Fig 14).



Figure 13



Figure 14

Corkscrew Claw and Interdigital Fibroma (corns)

Both of these foot conditions (Figs 15-17) can cause lameness and ongoing problems during a bull's lifetime. Due to the hereditary nature of these problems then bulls should not be certified as suitable for breeding if they are evident even though they may not be interfering with the ability to breed on the day of examination. The reason for being classified as unsuitable for breeding can be detailed on the comments section of the PBE certificate allowing potential purchasers to make their own judgement on whether they still choose to purchase a bull with the condition noted.



Figure 15



Figure 16



Figure 17

2.3 EXAMINATION OF SCROTUM AND CONTENTS

Examination of Scrotal Shape

Before handling the scrotum and testicles a visual appraisal of the scrotal shape should be carried out. In a cold environment this may be difficult as the dartos muscle of the scrotal wall and the cremaster muscles will pull the testicles closer to the body wall. In warm conditions with the dartos and cremaster relaxed the scrotal shape can be assessed more easily.

The three common descriptors for scrotal shape are

Straight (slab) sided scrotum (Fig 18) – may be associated with small testicles and excessive fat in neck of scrotum

Normal scrotum (Fig 19) – a pendulous scrotum with a well defined neck is ideal for thermoregulation of testicles

Wedge-shaped scrotum (Fig 20) – this is associated with smaller testicles and excess fat in the neck which will be detrimental to thermoregulation and semen quality.



Figure 18



Figure 19



Figure 20

There is a clear relationship between scrotal conformation and semen quality and in one retrospective study of 958 bulls in Canada 72% of bulls with pendulous scrotums had satisfactory semen quality whereas only 27% of bulls with straight-sided and 1% of bulls with wedge-shaped scrotums had satisfactory semen quality.

Examination of the Scrotum and Contents

Careful palpation of the scrotum and its contents is critical and many different abnormalities can be detected at this stage of the examination that may lead to a bull failing the physical examination section.

The scrotal skin should be smooth and elastic and the testicles should move freely within it. Chronic thickening of the skin at the base of the scrotum (Figs 21-22) may be caused by trauma or mange and if inflamed may cause heating of the testicles or a tendency for them to be held up high in the scrotum. Acute photosensitisation (Fig 23) can cause oedema and swelling of the scrotum and contents.



Figure 21



Figure 22



Figure 23

The neck of the scrotum should be palpated from the body wall down to the top of the testicles (Fig 24). Thickening of the neck and spermatic cords may be due to conditions such as scrotal hernia (Fig 25), varicoceles, abscesses or simply excessive fat.



Figure 24



Figure 25



Figure 26

Each testicle should be palpated in turn and any abnormalities noted (Fig 26 – normal anatomy). Testicular tone should be noted. If excessively soft (may be evidence of degeneration) or firm/painful (evidence of orchitis) but judgement of tone is subjective and individual bulls' testicular tone will vary depending on time of year, plane of nutrition and general health. Abnormalities in testicular tone that are significant (ie. due to degeneration) will normally be confirmed when semen is examined. The testicles should be symmetrical and any significant difference in shape or size should be noted.

Orchitis

Inflammation of the testicles (orchitis) can be unilateral or bilateral. In the author's experience it is unusual for cases of acute orchitis to be detected by owners and often when detected during PBE the condition is chronic. Acute orchitis with or without periorchitis will present with hot swollen testicle(s), with concurrent hydrocele and oedema making palpation difficult (Fig 27). Until the initial swelling has subsided it is hard to establish if orchitis or acute epididymitis or both are present. Treatment with hydrotherapy, antibiotics and non-steroidal anti-inflammatories is indicated however prognosis is poor. Once the acute swelling has regressed re-examination will allow more specific diagnosis. Chronic orchitis will lead to enlargement and change of testicular shape (Fig 28). Often the affected testicle may appear rounded and tense (Fig 29). There may be pain on palpation in some cases.



Figure 27



Figure 28



Figure 29

Testicular hypoplasia

Testicular hypoplasia can be unilateral or bilateral as in the bull in Fig 30 which had a SC of 26cm at four years of age. Differences in testicular size <20% may be hard to identify, so mild unilateral hypoplasia may not always be diagnosed. Significant unilateral hypoplasia will clearly lead to a fail in the physical examination section and cases of bilateral hypoplasia will normally lead to substandard SC measurement and semen quality. It is important that young bulls with testicular hypoplasia are not selected for breeding due to the hereditary nature of the condition and minimum SC standards must reflect this. Testicular hypoplasia is a developmental problem so affected bulls are likely to have history of infertility and small SC since puberty whereas testicular degeneration (Fig 31) may follow a history of previous normal fertility.



Figure 30



Figure 31

Testicular degeneration

Degeneration can be unilateral (Fig 31) or bilateral and can be caused by many factors including overheating, stress (which reduces LH and testosterone levels), toxic, genetic or nutritional problems. Loss of testicular tone and reduction in size is due to degeneration of the spermatogenic cells within the tubules and depending on cause can be temporary or permanent. Chronic degeneration can progress to atrophy with fibrosis causing the affected testicle(s) to feel firm.

Ultrasound examination of testicles may be useful when differences in testicular size or tone are detected. Differences in degree of echogenicity of the testicular stroma or areas of calcification/fibrosis may indicate chronic degeneration. Figures 32 and 33 are the left and right testicles of the bull pictured in Fig 28 and show diffuse increased echogenicity in the right testicular stroma. This bull had a history of poor fertility and was producing semen with substandard motility and morphology. The lesions seen in Fig 34 indicate areas of calcification or fibrosis suggesting chronic degeneration. It is worth noting however that older bulls may have areas of scattered calcification within the testicular stroma but still be capable of producing semen that is satisfactory.

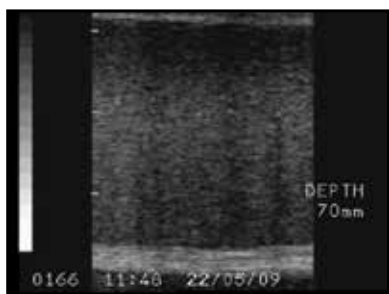


Figure 32



Figure 33

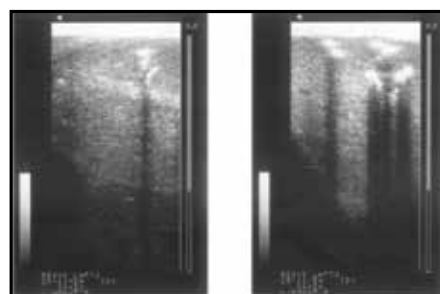


Figure 34

Epididymitis

The head (caput), tail (cauda) and body of the epididymis should be palpated carefully. The caput should be obvious and in some bulls feels quite firm. The cauda should be prominent and turgid in bulls with normal sperm production. An empty, small or flaccid cauda may be an indication of blockage/segmental aplasia of the ducts or simply poor sperm production (testicular hypoplasia/degeneration).

Epididymitis is not uncommon in bulls and is normally detected during PBE without a history of clinical signs related to the acute stage of disease. Epididymitis may be found along with seminal vesiculitis particularly in young bulls (see later). Epididymitis affecting the caput will lead to discomfort on palpation with varying degrees of swelling/enlargement (Fig 35-36, compare with normal anatomy Fig 37). Significant, non painful enlargement may be found with cases of sperm granuloma/spermatocoele where blockage of the efferent ducts will lead to a small flaccid cauda epididymis on the same side. Enlargement of the cauda caused by epididymitis can be unilateral (Fig 38) or bilateral. It is important to note that some bulls with unilateral chronic epididymitis may produce a perfectly acceptable ejaculate that passes semen examination standards however, these bulls are unsuitable for certification as they are only producing sperm from one testicle.



Figure 35



Figure 36



Figure 37



Figure 38

2.4 SCROTAL CIRCUMFERENCE (SC)

Scrotal circumference is a critical component of PBE of bulls as the SC measurement is highly correlated to paired testes weight, daily sperm production and semen quality. The SC measurement at 1-2 yrs of age in bulls is moderately to highly heritable. Bulls with above average SC reach puberty earlier and this trait can be passed to female offspring. Selection of young bulls with above average SC will improve the potential fertility of their female offspring which is clearly beneficial in sires used to breed replacement heifers. Bulls with below target SC as yearlings will still have small testicles by 2 year old so culling decisions can be made when measuring SC in young bulls. As SC is directly related to the volume of seminiferous tubules available to produce sperm it is critical that bulls expected to sire large breeding groups (eg 40-50 cows) in restricted mating periods have adequate SC.

Overfeeding young bulls will not improve testicular size but may falsely increase SC due to obesity and fat in scrotum. Subsequent reduction in SC may be due to a combination of fat loss and / or degeneration leading to poor semen quality. Rearing of pedigree bulls on moderate levels of nutrition from weaning to 15 months of age would no doubt reduce the incidence of subsequent laminitis and musculoskeletal problems and improve semen quality in young bulls for their first breeding season.

Technique for SC Measurement

With a ramp bar in place the practitioner should approach the bull from behind moving the hands over the rump and then down to the thigh region to ensure the bull is aware of the examiner. The testicles must be gently pulled ventrally into the base of the scrotal sac if pulled up and usually bulls will relax and allow this after a period of massage/palpation. If the bull resents initial palpation or kicks out (which is unusual) then mild sedation may be required to ensure safety. Sedation may be contra-indicated for the next stage of the examination (EEJ or AV collection) so another option is to leave the SC measurement and palpation until after EEJ when the bull will often be calmer and be easier to examine.

The testes are cradled in the scrotum by one hand held at the neck of the scrotum with thumb and forefinger held either side of the neck of the scrotum (Fig 39). The thumb should not be pushed in between the spermatic cords as this can push the testes apart thus increasing the SC falsely. The tape is placed around the widest part of the scrotum and pulled snugly until the skin is indented (Figs 40-41). The procedure should be repeatable and accurate within 0.5cm with different operators using the same tape.



Figure 39



Figure 40



Figure 41

Factors Influencing SC

SC is influenced significantly by breed (*Bos indicus* breeds tend to be smaller than *Bos Taurus*) and breeds historically selected for dual-purpose or milk traits such as Simmental, Charolais and Angus reach puberty earlier and tend to have greater average SC than breeds such as Belgian Blue, Limousin and Blonde d'Aquitaine which have been selected for beef traits.

Nutrition can also have an effect on SC in young bulls. Various studies have shown bulls fed high energy rations between weaning and 12-15 months of age will tend to have greater SC than bulls fed less intensively. In these studies the greater SC was attributable to scrotal fat rather than greater testicular volume. Also bulls fed excessive energy levels at this age also tend to have poorer sperm quality than bulls fed moderate /forage based diets. The effects on sperm quality are probably due to the impairment of thermoregulation by excessive fat in the neck of the scrotum. Other studies have suggested that high energy diets fed up to 12 months of age will not be detrimental to sperm quality provided that the diet after 12 months of age is moderated to prevent fattening.

It is clear that excessive high energy cereal based fattening diets fed to bulls between weaning and 12-18 months of age are likely to lead to obesity, lowered semen quality and temporary degeneration when rapid weight loss occurs at the start of breeding. In addition to the effects on semen quality the feeding of intensive cereal based diets will also increase the risk of laminitis and musculoskeletal problems.

Once sexual maturity is reached some variation in SC will occur related to seasonality and plane of nutrition but these changes should not normally exceed 1-2 cm. In a Canadian study the SC of 251 bulls were measured at the time of sale and again at pre-breeding checks and SC reductions of around 2 cm were found in many bulls. Losses in SC of 2-4 cm between point of sale and start of mating are likely to indicate testicular degeneration rather than simply bodyweight loss. Significant reductions in SC of 2-4 cm or more can occur over a period of weeks when testicular degeneration occurs and recovery, if it occurs, may take several months.

Minimum Standards for Scrotal Circumference

The standards used for assessing whether bulls have adequate SC vary around the world however the Society for Theriogenology (SFT) standards (table 1) have been used in the USA as the basis for breeding soundness standards for many years.

Table 1 : SFT standards for SC

Age in months	12-15	>15 < 18	>18 < 21	>21 < 24	>24
Minimum SC	30cm	31cm	32cm	33cm	34 cm

These measurements should be achieved by most normal bulls of *Bos taurus* breeds. However, as there is significant breed variation in age at puberty and SC, if they were applied to all breeds for certification purpose there is no doubt that certain breeds such as Belgian/British Blue, Limousin and Blonde d'Aquitaine would have a proportion of bulls that failed to reach these minimum standards by age. For this reason many countries have adapted the SFT guidelines for age and breed and for certification purposes it seems reasonable to use published breed standards as criteria for certification.

In the UK BCVA Bull PBE certificate bulls can pass SC standards for certification if they achieve the relevant breed standards for age. For most breeds this will be similar or indeed greater than the SFT standards but for others, SC standards may be lower (eg British Blue, Limousin).

2.5 EXAMINATION OF SHEATH AND PREPUCE

Having completed the examination of the scrotum and contents the sheath, prepuce and penis can be inspected (Fig 42, a normal bull). The sheath should be examined and palpated for any abnormal swellings which may be present. Lesions that may cause swelling in the sheath would include:



Figure 42

Penile Rupture/Haematoma

Rupture of the tunica albuginea commonly occurs around the dorsal aspect of the sigmoid flexure leading to haematoma formation as blood leaks from the corpus cavernosum. This may be caused by sudden movement of the cow during intromission or when young bulls are mounting each other.

There will be a variable sized, painful swelling at the base of the sheath anterior to the scrotum (Figs 43-44). Initially the swelling may be diffuse and extensive and later, more localised. The prepuce and/or tip of penis may be prolapsed. Ultrasound will confirm the diagnosis of haematoma. Chronic cases may have progressed to abscess formation by the time of presentation. In acute cases, surgery to drain very large haematomas can be attempted within 7 days of occurrence. In more chronic cases or with smaller haematomas then medical treatment is more common and should include hydrotherapy, systemic antibiotics and at least 2 months of sexual rest. Complications are common and include recurrence of rupture, abscessation with adhesion formation, development of venous shunts and desensitisation due to dorsal nerve damage.



Figure 43



Figure 44

Preputial Laceration/Avulsion

Traumatic tearing of the prepuce most commonly occurs ventrally where it joins the free portion of the penis (Fig 45). In acute cases there will be diffuse painful swelling of the sheath just caudal to the preputial orifice (Fig 46). There may be bloody discharge and partial prolapse of the tip of the penis or prepuce. In chronic/neglected cases the damaged prepuce may become infected and contraction of the damaged tissue may cause paraphimosis and “club penis” (Fig 47) due to constriction at the junction of the penis and prepuce. More commonly the injury may lead to structure formation or adhesions within the sheath leading to an inability to extrude the penis and total infertility. Chronic infected case will have a diffuse painful swelling extending for a variable way along the sheath.



Figure 45



Figure 46



Figure 47

Surgical repair of preputial tears is rarely possible as many cases are not presented until some time after the initial injury. Topical antibiotic/anti-inflammatory applied regularly or flushing the sheath with mild antiseptic solution will help reduce the risk of infection and adhesions/stricture forming during healing. After 2 months rest the bull can be test mated to assess healing and see if normal intromission is possible.

Prolapse of the Prepuce

A sporadic problem associated traditionally with mature Hereford or Angus bulls although any breed can be affected. Failure of the retractor preputiae muscle allows preputial epithelium to protrude beyond the preputial orifice (Fig 48) possibly leading to traumatic damage. Preputial epithelium may be prolapsed permanently leading to secondary trauma and infection (Figs 49-50). This can cause infertility as stenosis of the prepuce develops preventing full extrusion of the penis (phimosis).



Figure 48



Figure 49



Figure 50

In mild cases hydrotherapy and treatment with antibiotics and anti-inflammatory systemically/locally may be sufficient however if the prepuce is chronically inflamed and stenosis has occurred then surgical resection of the prepuce or culling may be required.

2.6 EXAMINATION OF THE PENIS

There are many problems that can effect the bull's penis that may be detected during routine PBE. A bull should not be certified suitable for breeding unless the penis has been visualised during the PBE as problems such as papillomata or persistent frenulum could be missed. If semen is collected using EEJ it is critical to remember that even if full erection is achieved during the procedure there are still problems such as deviations that cannot be detected reliably unless the bull is observed serving a female. Initial examination of the penis can be carried out during the physical examination by palpation through the sheath. Lesions such as penile haematomata or large papillomata may be detected at this stage however proper examination requires protrusion of the penis which will mean this part of the examination is done during semen collection (Fig 51). Many bulls will partly protrude the penis during rectal stimulation prior to EEJ allowing inspection of the tip of the penis. An assistant can carefully grasp the free end of the penis with a gauze swap and pull it forward allowing further examination if required.



Figure 51

The more common abnormalities of the penis that may be found are listed below:

Penile Deviations

The most common deviation of the penis in bulls is premature spiral deviation or “corkscrew” penis (Figs 52-53). This condition is normally acquired following a history of normal breeding performance and it is normally first detected during the breeding season when cows are seen to be returning despite repeated attempts at service by the bull. It must be remembered that spiral deviation of the penis is a normal phenomenon after intromission during the ejaculatory thrust within the vagina. Premature spiral deviation is seen as the bull is searching for the vulva and thus prevents normal intromission in many attempts at service leading to variable degrees of subfertility or total infertility. The progression of the condition can be gradual but most bulls develop the problem between 2-6 years of age with polled breeds more susceptible. As the condition is caused by failure of the dorsal apical ligament, surgical techniques have been described that involve anchor suturing the apical ligament to the tunica albuginea. The poor success rate of surgery means it is rarely attempted and most bulls are culled after diagnosis. As the condition has a heritable component this must be considered during pedigree breeding programmes.



Figure 52



Figure 53



Figure 54

NB. Occasionally, normal bulls will corkscrew during EEJ but this is not grounds for failing a PBE. Bulls noted to spiral during EEJ should be recorded in the comments section with advice to observe mating closely to ensure normal intromission is occurring.

Other penile deviations are occasionally observed such as ventral deviation (Fig 54) when the penis curves downward preventing normal intromission.

Traumatic injury to free end of penis

This may occur along with preputial injury or in isolation. Penile lacerations may heal well by second intention and treatment is rarely required. Occasionally problems may be caused by damage extending into the urethra or damage to the dorsal nerve leading to desensitisation of the tip of the penis. Hair rings can cause necrosis of the distal penis if neglected (Figs 55-56).



Figure 55



Figure 56

Balanoposthitis

Infection of the preputial and penile epithelium can cause temporary infertility due to reluctance to serve or occasionally more permanent problems due to adhesion formation leading to inability to fully extrude the penis. Specific agents causing balanoposthitis include *Bovine herpesvirus -1* (BHV1) and *Mycoplasma/ureaplasma* spp. Lesions may not be noted in the acute stage of the disease and may be found in the healing stage at routine examination (Fig 57). Vaccination of breeding bulls with IBR virus vaccine should be routine unless bulls are potentially for export or semen collection. Taking naïve pedigree bulls to shows is high risk and the author has seen bulls that have seroconverted to IBR with subsequent testicular degeneration leading to prolonged subfertility. Mixed ureaplasma/mycoplasma infections can cause a granular balanoposthitis the significance of which is unclear. Infection of the preputial cavity with *Campylobacter fetus* var. *venerealis* will normally be subclinical however isolation of this agent from preputial swabs/washings clearly is highly significant as a venereal disease.



Figure 57

Transmissible Viral Fibropapillomatosis (warts)

Caused by bovine papillomavirus and normally only seen in young bulls (< 3yo) fibropapillomas can be venereally transmitted to females (Fig 58). Fibropapillomas can be single pedunculated masses or large broad-based cauliflower-like lesions on the glans penis and prepuce which may prevent intromission due to the size of the lesion and associated discomfort (Figs 59-60). Secondary superficial bacterial infection of papillomas is common and may lead to bleeding/discharge from prepuce. Treatment is not normally indicated except in severe cases when autogenous vaccines and/or de-bulking the lesions with surgery are options. Care must be taken when attempting surgery that the dorsal nerve of the penis or urethral opening are not damaged. Cryosurgery is an alternative to surgical debridement. Spontaneous regression occurs in most cases however bulls with fibropapillomas found at routine PBE should not be certified as suitable for breeding and the reason for failing the physical examination section should be noted in the comments section of the PBE certificate.



Figure 58



Figure 59



Figure 60

Persistent Penile Frenulum

A rare congenital problem identified in young bulls caused by failure of complete separation of the fused penis and prepuce at puberty. A band of tissue extends from the ventral prepuce to the tip of the penis causing deviation of the tip of the penis during attempted service (Fig 61). Clinical examination during attempted service or when stimulated with an electroejaulator will demonstrate the lesion (Fig 62). Simple surgical excision of the band of tissue is successful but as the condition can be inherited, treated bulls should not be used for pedigree breeding. Occasionally, a flap of skin is observed on the ventral penis at the junction of the prepuce (Fig 63) which can be mistaken for a papilloma but is probably a remnant of the frenulum. Unlike papillomas, these lesions are harmless and do not interfere with service.



Figure 61



Figure 62

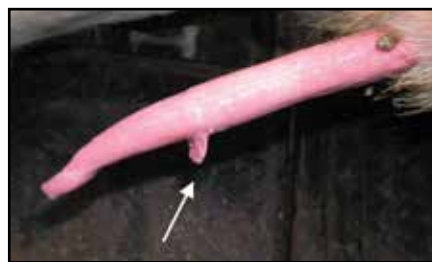


Figure 63

Failure of Erection

Bulls can develop erection failure or partial erection causing failure of intromission. Erection failure may be suspected during EEJ but must be confirmed during natural service observation. The most common cause of erection failure when libido is normal is the presence of vascular shunts between the corpus cavernosum and other structures preventing normal intracavernosal blood pressure required to achieve or maintain erection. Occlusions or shunts may be a sequelae to penile trauma/haematoma. Definitive diagnosis requires contrast radiography of the penis.

2.7 EXAMINATION OF THE INTERNAL ACCESSORY SEX GLANDS

After the physical examination of the external genitalia the next stage in the physical examination of the reproductive tract is rectal palpation to check for abnormalities of the accessory sex glands (Fig 65). The accessory sex glands of the bull include the prostate, bulbourethral (Cowper's) and vesicular glands (Fig 64). The bulbourethral glands are embedded in the urethralis muscle near the anal region and are not palpable. On the floor of the pelvis working from caudal to cranial the pelvic urethra surrounded by the firm urethralis muscle is felt and the prostate is located as a firm transverse ring-like band of tissue either side of which lie the paired seminal vesicles. The vesicular glands should be uniform in size, lobulated, turgid and mobile and non painful on palpation. They enlarge with age of bull. The ampullae of the ductus deferens are narrow tubular structures that lie cranial to the prostate and continue into the ductus deferens which leave the abdomen via the inguinal rings. While infection of any of the accessory sex glands is possible the most common abnormality detected during rectal examination is seminal vesiculitis (vesicular adenitis).

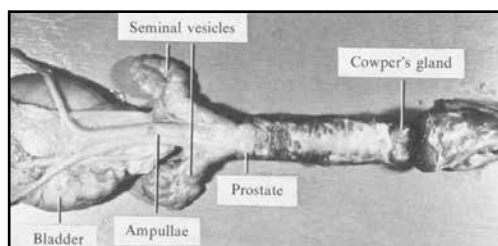


Figure 64



Figure 65

Seminal Vesiculitis

Infection of the seminal vesicles is not uncommon in young peripubertal bulls and can occur alone or in association with epididymitis, orchitis or infection of other accessory glands. Older bulls can be affected with a chronic form of the disease. The aetiology of seminal vesiculitis is unclear and it has been associated with various pathogens including, viruses, *Chlamydia*, *Mycoplasma* spp, *Mycobacteria* and *Trueperella pyogenes*. Some cases are associated with segmental aplasia of the glands. Infection may be ascending from epididymitis/cystitis or due to bacteraemic spread from lesions elsewhere in the body eg. rumenitis, liver abscess or bronchopneumonia. In most cases the bull will show no overt clinical signs but occasionally in acute cases there may be signs of pyrexia, abdominal pain and reluctance to serve. Pain may be associated with defecation and rectal examination. Conditions associated with abdominal/pelvic pain such as urolithiasis, peritonitis or infection of other accessory glands could produce similar signs.

The condition is normally diagnosed during routine breeding soundness examination when rectal palpation may reveal initially swollen/painful then firm/hard vesicles. Cases can be unilateral or more rarely bilateral. Some chronic cases may develop abscessation with localised adhesions and peritonitis (Fig 66). Ultrasound examination may reveal abscessation and dilation of the glands. Examination of an ejaculate will normally reveal semen with leucocytes, poor motility and a high percentage of secondary defects such as detached heads and midpiece reflex defect. Flakes of pus may be evident floating in the ejaculate in severe cases (Fig 67) or as a deposit when the collection tube is left to settle for a short time (Fig 68).



Figure 66



Figure 67



Figure 68

Leucocytes are often detected when carrying out progressive motility examination with phase contrast microscopy (see p 41) and staining air-dried semen smears with giemsa or methylene blue will confirm large numbers of leucocytes in the ejaculate which grossly may contain blood or floccules of pus. Leucocytes will not take up nigrosin-eosin stain and appear as round white structures with a diameter 1-2 times the length of a sperm head on morphology smears. Normal bull semen should not contain leucocytes however if semen is collected by EEJ and bulls ejaculate in the sheath the source of leucocytes in the ejaculate could be from a balanoposthitis. In older bulls with chronic sclerosing type vesiculitis the spermiogram may be relatively normal.

Treatment of Vesiculitis: Antibiotic treatment (eg tilmicosin or tulathromycin) has been reported to be moderately successful in curing vesiculitis in young bulls however many cases may self cure. More radical treatments such as injection of sclerosing agents or antibiotics directly into the glands or surgical removal have been described but are not normally considered in commercial beef bulls. Treatment of chronic vesiculitis in older bulls is likely to be less effective.

SUMMARY OF SECTION 1 - PHYSICAL EXAMINATION

If the bull is shown to be free of significant physical defects that could affect fertility or the ability to mate and free from physical heritable defects that could affect progeny and has a scrotal circumference that meets minimum breed standards then it can be classified as SATISFACTORY in section 1 of the BCVA PBE Certificate (Fig 69). If abnormalities are found in any of the sub-sections then the bull is classified UNSATISFACTORY in section 1.

1. PHYSICAL EXAMINATION		
Body Condition Score (1-5)		
	*NAD	*Abnormal
Heart/Lungs	<input type="checkbox"/>	<input type="checkbox"/>
Eyes	<input type="checkbox"/>	<input type="checkbox"/>
Incisor/dental pad alignment	<input type="checkbox"/>	<input type="checkbox"/>
Musculoskeletal system	<input type="checkbox"/>	<input type="checkbox"/>
External genitalia	<input type="checkbox"/>	<input type="checkbox"/>
Internal accessory glands	<input type="checkbox"/>	<input type="checkbox"/>
Scrotal circumference	cm	
Overall result: SATISFACTORY*/UNSATISFACTORY*		
*Place X in box to indicate findings. NAD = N		

Figure 69



3. SECTION 2 – SEMEN EXAMINATION

3.1 COLLECTION METHODS

Semen can be collected from bulls in the field by various methods including, artificial vagina (AV), internal AV, electroejaculation (EEJ) and massage of the ampullae. For routine pre-breeding examination the method that is most likely to be used is EEJ as it allows a semen sample to be collected from most bulls in a crush after the physical part of the examination. Collection by external AV is more time consuming and carries health and safety risks in untrained bulls on farm. Massage of the ampullae allows a small sample of semen to be collected for examination and requires a trained assistant to aid in the collection.

Artificial Vagina Collection

Collection of semen by AV (Fig 70) can be considered the gold standard method as it allows collection of a semen sample that is most representative of an ejaculate produced during natural service. It also allows observation of service behaviour and libido if required. It is the method of choice in artificial insemination collection centres where the bulls are trained to be collected when mounting teaser animals. The technique is straightforward in AI centres where the bulls are used to handling and can be trained to mount non-oestrus teaser animals (bullocks or cows) however in the field situation collection of untrained stock bulls can be more difficult and present health and safety issues.

For collection of semen by AV in the field some forward planning is required. At least one oestrus female must be available for teasing the bull. The teaser should be restrained in a yard with good footing and in a position where there is plenty of space for the bull to jump and for the operator to get access for the AV collection. An escape route should be planned in advance if the bull is not restrained on halter/nose ring. A simple system the author has found to be effective is haltering a lightly sedated cow to a feed barrier and placing two round bales either side of the cow's shoulders to prevent excessive lateral movement (Fig 71).



Figure 70



Figure 71

The AV should be prepared when everything else is ready by filling with hot water at around 50-55°C to allow for some cooling to reach a final ideal liner temperature of around 42-45°C. The lumen temperature of the AV should be checked with a thermometer before the bull is released into the teasing area. The AV should be lightly lubricated with sterile, non spermicidal lubricant (eg. KY jelly). The collecting tube should be insulated to prevent cold shock of the ejaculate especially when collecting in winter. When the bull is released into the yard with the teaser cow the operator must be ready to step in and direct the penis into the AV by grasping the sheath. The penis should simply be directed into the AV opening as the bull is searching for the vulva and the AV should not be pushed forcibly onto the penis. Normally the bull will thrust and ejaculate into the AV as soon as the tip of the penis is stimulated by the temperature and pressure of the AV liner. When the bull thrusts the operator must be wary of positioning so as to avoid being tramped by hind feet. Once a sample has been collected the bull should be removed from the teaser cow until the sample is evaluated in case a second collection is needed.

Collecting semen by AV is without doubt the best method of semen collection for evaluation of individual suspect infertile bulls as it allows simultaneous evaluation of libido and physical ability to mate. The main disadvantages of this method are the need for teaser preparation, forward planning and the potential dangers to operators when attempting to collect. It can be time consuming and frustrating when attempting to collect from bulls unused to handling and wary of intervention and for this reason AV collection is not in widespread use internationally for routine evaluation of breeding soundness and tends to be reserved for individual examination of known suspect bulls.

Massage Of Ampullae

The technique of trans-rectal massage of the internal sex glands has been well described and is reported to be a successful method for collecting a sample of semen for evaluation in up to 97% of mature beef bulls used to handling. The same authors reported a lower success rate of 80% when using the technique in more fractious range beef bulls. The technique involves rhythmic massage of the ampullae of the ductus deferens and pelvic urethra in a cranial to caudal direction until semen is emitted from the penis and collected by another operator using a collection cone held over the opening of the sheath (Fig 72). The ampullae (Fig 73 arrow) should be “milked” by applying pressure with the thumb and forefinger for optimum success. Experienced operators should be able to collect a sample of semen within 1-2 minutes of massage if the bull is responsive to the technique. Semen collected by massage tends to have lower motility and live sperm % than that collected by EEJ probably due to the fact that many bulls do not protrude the penis and semen dribbles through the prepuce before being collected thus exposing the sperm to a more hostile environment. The % normal morphology counts were unaffected by massage indicating that the technique is a valid alternative to EEJ where a sample of semen is required for morphology assessment.



Figure 72

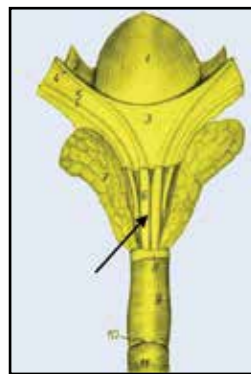


Figure 73

Electroejaculation

Electroejaculation (EEJ) is the standard method used for semen collection in the field by vets in North America and Australasia and has allowed the development of BSE as a routine procedure on farms. It has the great advantage of being easily carried out on restrained bulls without the need for teaser females. In the UK the procedure must be carried out by a veterinary surgeon and training in the procedure by an experienced operator is recommended to safeguard the welfare of the bulls. The welfare aspects and methods of reducing discomfort associated with EEJ have been reviewed recently and the conclusions of the author would suggest that the most important factor in safeguarding the welfare of bulls during EEJ is operator technique. In the author's experience the procedure can be carried out with minimal distress in the majority of bulls as the modern battery powered EEJ machines (eg Lane Pulsator IV, www.lane-mfg.com) allow complete control over the level and duration of stimulus applied.

Prior to collection by EEJ the bull should be separated from breeding females for at least 24 hours and preferably longer. For collection by EEJ the bull can be restrained in a suitable crush (Fig 74) or tethered in a race (Fig 75). It is important to have good access to the underbelly of the bull and for the bull to have non-slip footing and room to move forward and backwards to some degree during stimulation. If the neck is caught in a yolk care must be taken not to compress the cervical neck musculature too tightly as this can cause collapse or ataxia when the bull is released. Sedation should only be considered if bulls are aggressive or particularly agitated when restrained as it may increase the risk of the bull going down during EEJ or urinating during the procedure.



Figure 74



Figure 75

The author only has experience of the Lane Pulsator IV EEJ machine (Fig 76) and its use will be described in this manual. Two sizes of probe are available with this machine and recently weighted probes have become available allowing smaller diameter (60mm) probes to be used in most sizes of bulls. The 60mm weighted probe is ideal for young bulls from 15 months of age but can also be used in mature bulls (Fig 77). The smaller diameter probe is more likely to be pushed out of the rectum if a larger bull is straining so an assistant should always stand at the rear of the bull with a hand on the tailhead if required to prevent this and also to keep the probe from twisting out of position. The 75mm probe is suitable for most bull from 18 months of age however if the anal sphincter is tight the narrower 60mm probe should be used. Prior to insertion the probe should be checked to ensure the electrodes are clean and if necessary any build up of tarnish should be removed with fine wire pad/sandpaper.



Figure 76



Figure 77

Before insertion of the probe the bull is pre-stimulated for up to 60 seconds by gentle rectal massage of the ampullae, prostate and pelvic urethra. The pre-stimulation is normally carried out immediately after the examination of the internal accessory sex glands has been carried out.

During this massage the bull will often protrude the penis and start dripping pre-ejaculatory fluid. The probe is then carefully inserted ensuring a good contact with the rectal floor and avoiding pneumo-rectum. Before commencing stimulation ensure the bull is standing normally. The Lane pulsator machine allows a step-wise increase of the power range and within each step the voltage can be progressively increased by clock-wise turning of the rheostat knob. It is very important that bulls be 'acclimatised' to the electrical stimulation. This is done by setting the machine at the lowest power range setting 1 and then incrementally turning the rheostat knob on for 2 seconds and off for 2 seconds - one approach is to divide the arc the rheostat knob can be turned into 6 and the knob is then turned a sixth of the arc further each time until the knob is turned fully (Fig 78). The bull is closely observed for evidence of response to the stimulation - contraction of the preputial muscles, scrotum and shuffling of the feet. A threshold of stimulation is often recognised. Some bulls will pass through this threshold readily others will need to be 'nudged' through it - these latter bulls appear to be quite sensitive to electrical stimulation and show a more exaggerated response. It is very important that the operator ensure the bull is standing normally and stops stimulation if the bull is not - once he is standing normally again stimulation is resumed. Once this 'excitement' threshold (usually between the top of power range setting 1 and power range setting 2) is passed the bull will then start responding to the stimulation in a predictable way - as the stimulation is applied the bull will rock forward, clear pre-seminal fluid will drip from the preputial orifice and the penis will begin to extend.

Usually at the second power range setting the arc of the rheostat knob is divided into 3 or 4 and the stimulus incrementally applied as described above until the knob is turned fully - usually 2 full turns of the knob are delivered before advancing to the next power range setting. Generally when the rheostat knob is turned about one third to half-way round it has reached the maximum stimulation of the previous power range. Overall, the consecutive sequence of stimuli applied to the bull is increased exponentially. It is important that the stimuli be gradually delivered i.e. the rheostat knob is gradually turned clockwise to deliver the stimulus and then is rapidly turned anticlockwise to end the stimulus. The operator should strive to deliver the stimuli smoothly and rhythmically, watching the bull carefully and varying the stimuli according to the bull's response.



Figure 78




Figure 79



Figure 80

The manual setting allows complete control over the timing and increments of stimuli whereas the **autorun** programme takes the bull through a standard programme of stimuli which some operators prefer as it standardises the procedure. The author prefers to use the manual setting for the initial collection and if a second collection is required will often use autorun then as the bulls are less sensitive to the procedure on the second EEJ. Most bulls ejaculate at the mid-power range (power range steps 3 to 5 for the Lane Pulsator IV). Most bulls will protrude the penis partially or fully prior to emitting semen. The operator must watch carefully for a change in the appearance of the fluid from clear to opaque/milky. At this point the hand-held collection cone with attached collection tube is placed over the glans penis (Fig 79) or preputial orifice (Fig 80).



The bull is stimulated to emit one to three jets of semen. In cold conditions the collection tube can be maintained warm by placing it in a styrofoam sleeve, or an attached plastic bag containing water at a temperature of around 37°C.

Not all bulls will protrude the penis. An assistant can try pushing the sigmoid flexure cranially as the stimulus is applied and then the operator can grasp the tip of the penis with a gauze swab to keep it extruded during collection. The preputial hairs should be clipped if long, so if the bull ejaculates into the preputial cavity a minimally contaminated sample can be collected from the preputial orifice.

As soon as the sample is collected it should be kept insulated and taken to the microscope for assessment of motility. An assistant should be left with the bull ensuring the probe is not pushed out of the rectum and damaged whilst the initial motility examination is carried out. If the sample collected has poor motility then a second or third sample (occasionally required with “rusty load”) can be collected at intervals of approximately 5 minutes. Usually the bull responds in a very predictable way to subsequent collections and less hind limb muscle stimulation is observed.

Failure to Collect with EEJ

Over 95% of bulls will normally produce a suitable sample for assessment of semen by EEJ. In rare cases bulls with no obvious physical abnormalities of the reproductive system will fail to produce a sample with EEJ despite repeated attempts. These bulls cannot be classed as unsuitable for breeding without further attempts at semen collection by another method such as AV or massage of ampullae.

In some cases bulls will react as expected with EEJ, getting an erection and emitting clear pre-ejaculatory fluid yet not appearing to emit the sperm rich ejaculate when expected. If the maximal stimulus has been reached the machine should be switched off and the bull allowed to rest for a few minutes. As the penis and urethral muscles relax the ejaculate may suddenly be released so the collecting cone should be kept ready! Another technique is to allow a few minutes rest then apply 2 or 3 maximal stimuli at level 2 -3 which will often cause release of the ejaculate which has been retained in the urethra.

3.2 ASSESSMENT OF THE SEMEN SAMPLE

Once a semen sample has been obtained from the bull it should be taken immediately to the field lab area for examination of motility. The collection tube should be placed in a water bath or incubator if there is any delay in examination. The bull should be left in the crush if collection has been with EEJ and the probe left in place to allow a subsequent collection of semen if required.

Appearance / Density of Semen Sample

Studies comparing the characteristics of semen collected by EEJ and AV from the same bulls have shown no significant differences in motility, percentage normal sperm and fertilising capacity when used fresh/chilled. Semen collected by EEJ should not be compared in volume and appearance to an AV collection and should be thought of as a “biopsy” of semen to allow assessment of motility and sperm morphology.

Appearance of the sample is classified as creamy, milky or watery (Fig 81) and while this gives an indication of sperm concentration it cannot be used as a criterion for judging semen quality or output. Potential semen production capacity is best estimated by scrotal circumference in bulls. Very dense creamy semen can often have poor motility due to a high percentage dead or decapitated

sperm as seen with “rusty load”. As the density of the ejaculate can be influenced by collection method it is simply recorded but not used as a critical criterion. Occasionally certain bulls produce ejaculates with a yellow colouration (Fig 82). This is caused by riboflavin pigmentation and does not effect semen quality



Figure 81



Figure 82

Gross Motility

Gross motility is assessed by placing a 5-10mm drop of the fresh semen sample on a clean warmed slide (Fig 83) and examining under low power brightfield magnification. Whilst assessing the gross motility the tube with the semen sample should be placed in an incubator or water bath to prevent chilling. Gross motility is scored subjectively on a five point scale with the following descriptors.

Scale	Description
1	No swirl; generalised oscillation of individual sperm only
2	Very slow distinct swirl
3	Slow distinct swirl
4	Moderate fast distinct swirl; dark waves
5	Fast, distinct swirls with continuous dark waves

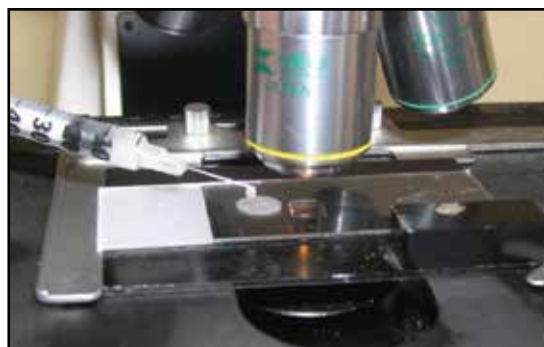


Figure 83

Gross motility depends on three factors: concentration, % progressively motile sperm and speed of progression of sperm. When any one of these factors is depressed then rapid wave motion is unlikely to be seen. Semen samples with poor concentration may have over 80% rapidly progressive motile sperm but show no wave motion. In contrast, highly concentrated sperm may show good wave motion despite having a high % dead non-motile sperm as the dead sperm are simply washed along by the live sperm.

A sample of good density that shows poor gross motility should be regarded as suspicious unless it has been handled badly and allowed to chill. As gross motility is influenced greatly by concentration of the sample then it is essential that individual progressive motility is assessed in all bulls and this is the criterion used to pass or fail motility standards.

Progressive Motility

After gross motility has been scored, a small drop (2-4mm) of semen is placed on a clean warm slide and covered with a warmed cover slip (Figs 84-85). Larger sized cover slips should be used to help spread the drop into as thin a film as possible. Insulin injection syringes are useful as an alternative to pipettes to allow the transfer of small drops of semen onto slides. Examination under x 200 - x 400 magnification will allow an estimation of % progressive forward motility (Fig 86).

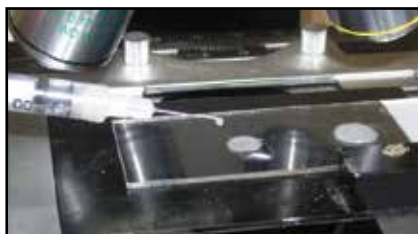


Figure 84



Figure 85

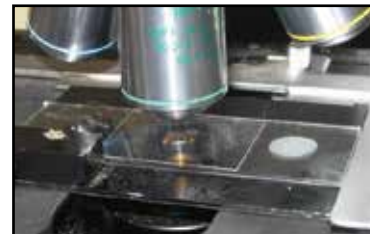


Figure 86

In the author's experience, if the semen sample is of very good density then it will normally need diluted in warmed isotonic saline before making the preparation, as even with a tiny drop of fresh semen the density of the preparation may be too great to easily estimate progressive motility. Having pre-filled tubes/vials in a warm box with 3-4ml normal or phosphate-buffered saline is sensible and a few drops of semen can be mixed in one of these tubes to give a dilution suitable for progressive motility assessment. With a good heated stage the estimation of progressive motility is easily done and even if the sample has cooled during preparation the sperm will quickly have their motility restored when warmed on the stage (37-39°C). Without a heated stage the estimation of progressive motility is likely to be highly inaccurate and liable to underestimation. Phase contrast microscopy is superior for examining wet mounts for progressive motility however if this is not available then simply lowering the condenser and adjusting the diaphragm with bright field microscopy is adequate.

The standard required to pass the semen examination section of the BCVA PBE certificate is a progressive motility of $\geq 60\%$. A bull that is producing good quality semen which has been handled properly after collection should easily pass the 60% standard. Bulls that consistently produce semen samples with poor motility will normally fail the morphology examination standards due to sperm defects eg midpiece/tail defects or detached heads. Conversely it must be remembered that bulls with $>60\%$ progressive motility may fail the morphology standards due to sperm with defects that do not impair motility eg. nuclear vacuoles or proximal drops.

With AV and EEJ collection if a first ejaculate scores poorly for gross and progressive motility and no physical abnormality of the testes has been noted a second and sometimes third collection should be attempted immediately as often a superior sample will be obtained as senescent sperm are cleared from the epididymis ("rusty load"). Ejaculates from bulls with "rusty load" are often dense and creamy but gross motility is very poor due to detached heads (see p 36) and dead sperm which clear after the epididymal stores have been cleared.

Identification of Foreign Cells in Semen

When carrying out progressive motility examination with good quality phase contrast microscopes the presence of abnormal cells (eg leucocytes, epithelial cells, spheroids, bacteria) in the semen will often be noted at this stage – see later.

3.3 SPERM MORPHOLOGY ASSESSMENT

As well as having a minimum of 60% progressive motility, the semen sample must have a minimum of 70% morphologically normal sperm to pass the semen examination section. Bulls with >20% nuclear (head) defects in a morphology counts should also be deemed unsatisfactory. The assessment of sperm morphology must be done in all cases even when motility is excellent as some sperm defects do not impair motility but have significant effect on fertilising capacity eg. nuclear vacuoles.

Preparation of Semen Smears for Morphology Assessment

Nigrosin-eosin stain is still widely used for preparing smears as the eosin is taken up by non-viable or damaged sperm allowing them to be differentiated from live sperm which appear white against the dark background of the nigrosin. When semen samples have been scored with good progressive motility (>60%) then carrying out live/dead counts is probably not necessary as the assessment of sperm viability has been done by progressive motility estimation.

Preparing good smears for morphology examination is an acquired skill which needs practice to perfect which explains the less than satisfactory results that are obtained when only doing occasional bull fertility examinations. Before making smears ensure the slides are clean and grease free by wiping with clean tissue paper. Stain should be purchased freshly at regular intervals from a reliable supplier as it has a limited shelf life. To make a smear a small 5-6 mm drop of nigrosin-eosin stain is placed at one end of a warm, frosted-end slide and using a fine dropper or insulin syringe a small drop of semen is mixed with the stain (Fig 87). After a short period the smear is made by pulling the drop of stain/semen along the slide with the edge of another slide (Fig 88-89). If the drop of stain is too big the smear will be too thick leading to a background that is too dark and will crack during drying.

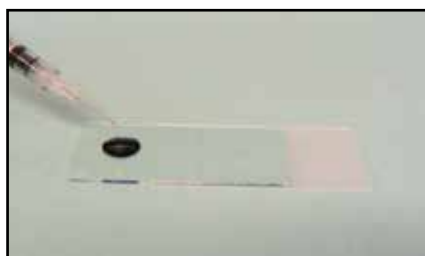


Figure 87



Figure 88



Figure 89

If too big a drop of semen is added to the stain it will create a smear that is too dense to count easily. The concentration of the semen sample will dictate the size of drop that is mixed with the stain on the slide and occasionally if the sample is very concentrated it may be better to dilute the sample in saline before attempting to make the stained smears. Care must be taken if doing this as mixing semen in cold or hypotonic solutions can create artefacts such as bent tails which must not be mis-interpreted as abnormalities in the final preparation.

After preparing at least 2 stained slides they should be air-dried as quickly as possible, labelled and stored in a slide box for detailed examination back at the practice. The main test tube containing the original semen sample should be labelled and retained until the final morphological examinations have been done as it may be necessary to make further smears for staining for other cells if this is indicated eg. methylene blue stain for leucocytes. If conditions are poor on farm for making stained smears then the semen samples (collected in screw top tubes) should be taken back to the practice and smears made there later that day. As live/dead estimates are not normally required, a short delay of several hours in making smears is not an issue and morphology will be unaffected

Morphology counts must be done under x1000 oil immersion magnification either with stained slides and brightfield microscopy (Figs 90-91) or using fresh, wet (formol saline killed) preparations and phase contrast microscopy (Fig 92). For wet mount preparations a small drop of semen (diluted if necessary) is examined under a large cover slip using x100 oil immersion phase contrast lens.

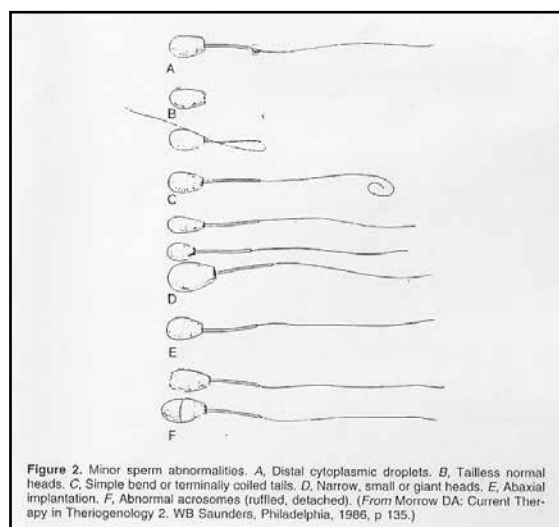
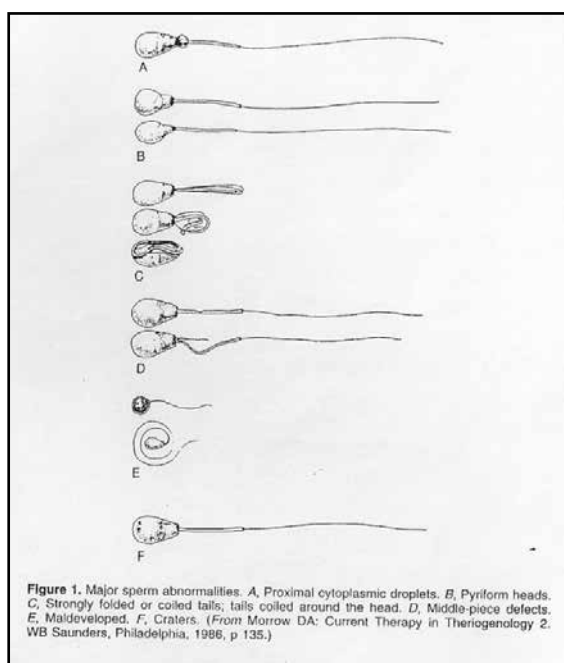


Figure 93

3.4 IDENTIFICATION AND SIGNIFICANCE OF ABNORMAL SPERM

An abnormal spermiogram, when interpreted along with a bull's history and physical examination findings, can indicate the severity of abnormal testicular function and be used to aid in giving a prognosis for recovery to normal sperm production. The morphology examination can be thought of as a biopsy of testicular function however care must be taken in over-interpretation of single ejaculates as the spermiogram can vary considerably between ejaculates taken over short time periods. *Where an abnormal spermiogram is found without any physical abnormality in the reproductive tract or history of environmental/toxic/stress insult then re-examination of bulls should be recommended after a period of at least 60 days to allow a full spermatogenic cycle to take place and assess recovery of what may be a transient problem.*

Although various classification systems have been used for defining sperm defects such as primary and secondary, major and minor (see figs below) it is not an exact science and the relationship between various defects and fertility is not clear cut. In general, defects of major significance are caused by disruption of spermatogenesis and will result in primary defects such as proximal droplets, deformed heads and dag-defect of tail. These are the type of defects you would expect to find in a bull with testicular degeneration. Disturbances of sperm maturation during epididymal transport or storage will lead to secondary defects such as detached heads, distal droplets and distal midpiece reflex of sperm tails. Some sperm defects may be "compensable" by increasing sperm dose and density and therefore their effect on fertility may be hard to predict. An example of a compensable defect would be midpiece reflex defect which simply prevents sperm from being progressively motile thus unlikely to reach the ovum. Semen with 30% distal midpiece reflex defect may have little effect on fertility if the remaining sperm are live and normal and present in sufficient numbers. Conversely "uncompensable" defects such as nuclear vacuoles may have a significant effect on fertility at a level of 30% as these sperm will reach the ovum and penetrate the zona thus preventing fertilisation by normal sperm capable of establishing a healthy embryo. Density of ejaculate will not alter the probability of a successful outcome in fertilisation thus this defect will have a much more significant effect on fertility than midpiece reflex. When doing a morphology count if more than one defect is present in a sperm cell it should be counted in the category of the most significant defect present. Eg a sperm with a deformed head and a midpiece reflex present should be recorded as a head defect.



3.5 COMMON SPERM ABNORMALITIES

Normal sperm

Normal sperm should have regular shaped heads with a centrally attached midpiece and straight tail (Figs 94-96). Some variation in head shape is acceptable with some bulls of normal fertility producing sperm with narrow based heads. Sperm with tapered or narrow heads present in smaller numbers along with other abnormal sperm are likely to be abnormal and present due to a disturbance in spermatogenesis.

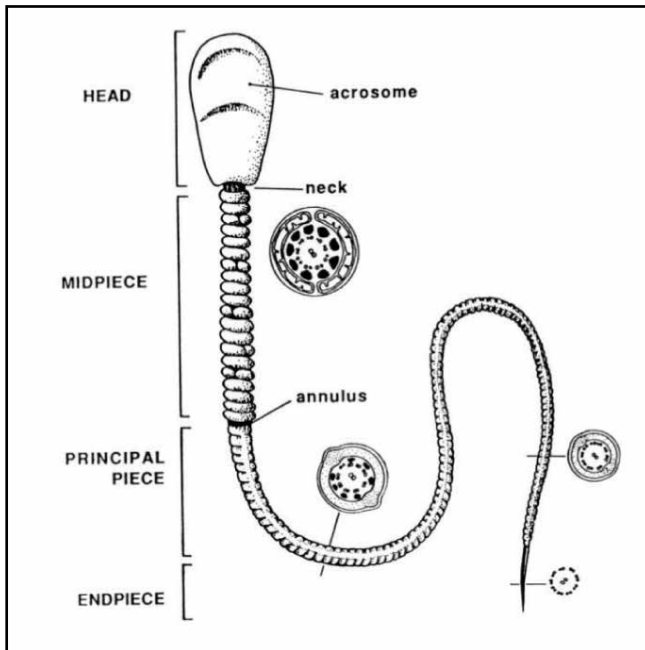


Figure 94



Figure 95



Figure 96

Defects Of The Sperm Head

Pyriform / Tapered heads

A moderate degree of pyriformity in the absence of other sperm defects may be normal in some bulls. More severely pyriform (pear shaped) (Figs 97-98) or tapered heads (Fig 99) have been associated with reduced fertility and can be classified as uncompensable defects. These defects are usually found along with other sperm defects indicating degeneration.



Figure 97

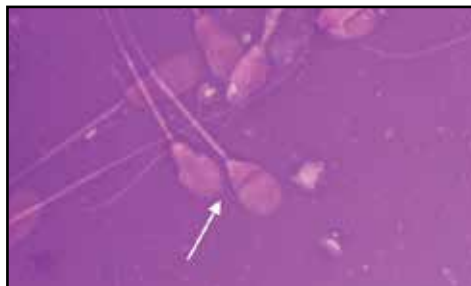


Figure 98



Figure 99

Microcephalic (Figs 100-101) and Macrocephalic Sperm

These defects are normally only seen in small numbers and may result from the uneven meiotic distribution of chromosomes in spermatocytes. Most of these abnormal sperm die before reaching the spermatid stage and are phagocytosed by Sertoli cells.



Figure 100



Figure 101

Nuclear Vacuoles

Nuclear vacuoles (craters) can be seen in several forms. Small vacuoles arranged in a row along the equatorial region of the nucleus are known as “diadem defect”. These can be hard to visualise with nigrosin-eosin smears and are more easily identified using phase contrast preparations (Fig 102). Larger confluent vacuoles are much easier to identify (Figs 103-105). These should be considered an uncompensable defect and likely to have a significant effect on fertility if present at > 20% in spermiogram. Semen samples from bulls with this defect may have good motility as sperm are live and progressively motile.



Figure 102



Figure 103

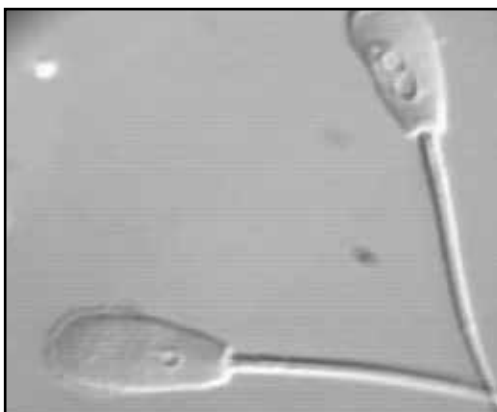


Figure 104



Figure 105

Knobbed Acrosome Defect

This defect is identified as a flattening or indentation at the apex of the acrosome (Figs 106-107). This defect can be of a genetic or environmental origin. If present in high numbers on repeated examinations then this is likely to be of genetic origin and bulls should be culled as they will be subfertile/infertile. Small numbers of sperm with this defect will be seen in bulls with disturbed spermatogenesis due to environmental causes thus the prognosis will depend on the underlying cause.

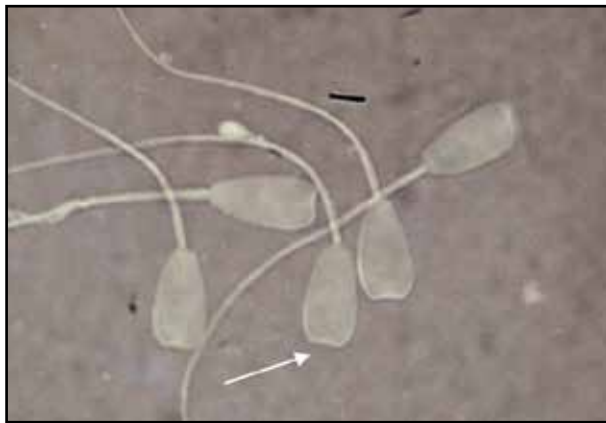


Figure 106



Figure 107

Detached Heads

Small numbers of detached heads are seen in most semen samples however this defect can be present in high numbers in cases of sperm accumulation/senescence (rusty load). If sperm accumulate abnormally in the cauda epididymis then the ejaculate may appear dense (Fig 108) but often contains a high % of dead sperm and detached heads leading to poor motility (Fig 109). These can often be cleared after several ejaculates but if they persist then they may be present due to stress, abnormal testicular thermoregulation or if present along with other sperm defects, due to testicular or accessory gland pathology.



Figure 108



Figure 109

Defects of the Sperm Tail

Distal Midpiece Reflex / Bent Tails

This is the commonest abnormality of the sperm tail seen in bull semen and originates during sperm maturation in the epididymis. The defect produces a bent tail of between 90-180° that occurs at the junction of the distal midpiece and principal piece. A cytoplasmic drop is normally trapped at the bend (Figs 110-113). The effect on fertility will be related to the % of the defect present as these sperm are not progressively motile. This defect must be differentiated from simple bent tails which may be caused by mishandling of semen samples or exposing semen to hypotonic shock (see p38).

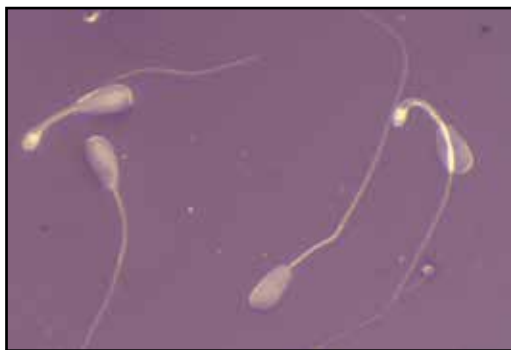


Figure 110



Figure 111



Figure 112



Figure 113

When 180° bends are present at the midpiece/principal piece junction without cytoplasmic drops (Figs 114-115) then these sperm should be classed as abnormal unless there is evidence that mishandling of the semen could have caused artefactual bending. Proper progressive motility examination will also allow visualisation of the bent tails con-firming they are genuine defects



Figure 114



Figure 115

Severely Coiled Tails / Dag Defect

Sperm with the dag defect have a severely coiled tail due to midpiece developmental abnormalities (Figs 116-118). Normally this defect is only seen in small numbers along with other sperm defects associated with disturbances of spermatogenesis. If present in a high percentage as a sole defect it is likely to be of genetic origin.



Figure 116



Figure 117



Figure 118

Coiled Principal Piece

Normally this defect it only seen in a small percentage of sperm and may be associated with stress or abnormal tes-ticular thermoregulation (Figs 119-121). Fresh wet mount preparations are useful to distinguish if the bent tails are artefactual.



Figure 119



Figure 120



Figure 121

Bowed Midpiece

This is normally an artefact of staining and drying however if large numbers are present, wet-mount preparations should be examined to confirm if a structural abnormality is present in the ejaculated sperm that may impair forward motility (Fig 122).

Bent Tails caused by Hypotonic Shock

Prolonged exposure to hypotonic solutions eg stains, water or urine can cause contortion or bending of the distal portion of the principal piece (Fig123-125). This must be differentiated from genuine tail abnormalities such as distal midpiece reflex.



Figure 122



Figure 123



Figure 124



Figure 125

Proximal Cytoplasmic Droplets

Most sperm have a cytoplasmic droplet present when entering the caput epididymis but normally the droplets are shed during epididymal transport. Proximal droplets present in ejaculated sperm are therefore not normal and a sign of disturbed epididymal or testicular function. If present in high numbers they are likely to lead to reduced fertility though the mechanism of this is not clear. Immature/pubertal bulls may produce ejaculates with a higher than normal % of sperm with proximal droplets (Figs 126-127).



Figure 126



Figure 127

In some ejaculates, large numbers of detached cytoplasmic droplets are seen in stained smears or wet mounts and must not be confused with foreign cells such as leucocytes which are larger. Cytoplasmic drops do not normally take up eosin stain (Fig 128) and can be identified clearly in phase contrast preparations (Fig 129).



Figure 128



Figure 129

Distal Cytoplasmic Droplets

Cytoplasmic droplets are normally shed from the distal midpiece position at the time of ejaculation when the sperm mix with seminal fluids therefore the presence of large numbers of sperm with distal droplets may indicate some abnormality of seminal fluids or ejaculation. Sperm with distal droplets should probably be counted as **normal** sperm during morphology counts as there is no evidence that they are associated with reduction in fertilising capacity (Figs 130-131).



Figure 130



Figure 131

Miscellaneous Sperm Defects

Tail Stump Defect

An uncommon defect that is probably of genetic origin. Sperm have a short tail stump that may be obscured by cyto-plasmic droplet material in many cases.

Mitochondrial Sheath Defects (segmental aplasia)

Sperm with this defect appear to have gaps in the midpiece structure (Fig 132) due to missing mitochondrial material. The sperm may appear fractured at this point but if fracture does not occur this sperm defect may have no effect on fertility.



Figure 132

Abaxial Tails

This defect appears to have no effect on fertility as long as the tail attachment allows normal motility.

Accessory Tails / Double Tails

Small accessory tails may be present alongside a normal tail (Figs 133-134) and this condition may be inherited. The effect on fertility is unknown. Double tails are occasionally observed (Fig 135).



Figure 133

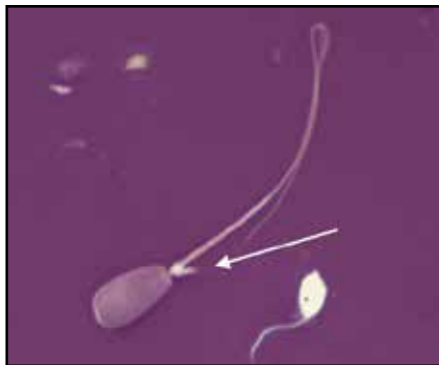


Figure 134



Figure 135

Foreign Cells in Semen

A variety of foreign cells may be found in semen and may indicate pathological inflammation somewhere in the re-productive tract or may simply be contaminants. The types of cells that may be found include red and white blood cells, epithelial cells, bacteria and spheroids (spermatogenic epithelial cells). EEJ collected samples from bulls which fail to get a normal erection may be contaminated with white cells and bacteria as the ejaculate runs out of the sheath and over contaminated preputial hairs and this must be considered when examining samples.

Epithelial cells are normal when present in small numbers but large numbers may indicate inflammation of the ac-cessory sex glands or urethra. Fig 136 shows an epithelial cell and a single white blood cell which do not take up eosin stain. Fig 137 shows the appearance of epithelial cells in a phase contrast wet preparation.



Figure 136



Figure 137

White Blood cells

Leucocytes may be seen in numbers in cases of vesiculitis or infection of other accessory glands and if they are not-ed on the wet preparation motility examination (Figs 138-139) then an air-dried smear of semen can be stained with methylene blue/giemsa to confirm their identity (Fig 140). Smears for leucocyte identification can be prepared exactly the same way as an anthrax blood smear preparation. Intact leucocytes do not take up eosin stain therefore appear as round white objects with a diameter 1-2 x the length of a sperm head on morphology smears (Fig 141). Normal semen should not contain more than occasional leucocytes however if the sample is not collected directly from the tip of the erect penis but from the opening of the sheath then they may be contaminants and simply indicate a bala-noposthitis. In cases of chronic severe vesiculitis the white cells may be clumped together in large numbers and noted grossly as pus in the sample at the time of collection (see p 22) .



Figure 138



Figure 139

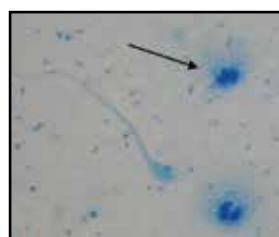


Figure 140



Figure 141

Bacteria may be present in significant numbers in semen samples from bulls with infection of accessory sex glands or may simply be contaminants. Like leucocytes, bacteria normally show up unstained on nigrosin/eosin smears (Fig 142) but can be identified easily with methylene blue staining (Fig 143).



Figure 142

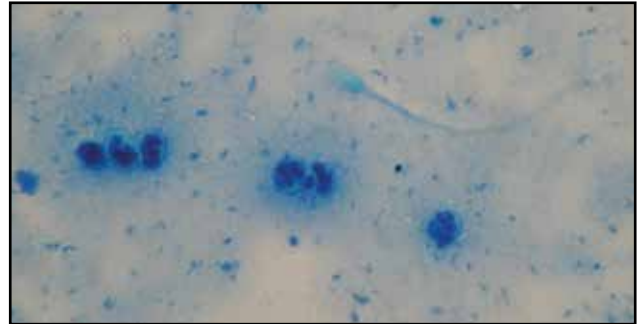


Figure 143

Spheroids are spermatogenic epithelial cells that have been shed from the seminiferous tubules and their presence may indicate testicular pathology such as degeneration or hypoplasia. Spheroids may appear similar to leucocytes but tend to be more irregular in size and often larger (Fig144). They often have a multinucleate or granular appearance visible with phase contrast microscopy wet preparations.



Figure 144

Staining Artefacts

A commonly noted artefact seen in nigrosin-eosin smears is a bright white halo surrounding the acrosome (Figs 145-146). This is caused by terminal movement of live sperm as the stained preparation dries and is more likely to be seen when smears are made immediately after semen collection and the mixing time of semen and stain prior to making the smear is short. Dead sperm will not show this stain drying artefact.



Figure 145



Figure 146

SUMMARY OF SECTION 2 - SEMEN EXAMINATION

If a representative ejaculate is collected which has a **progressive motility of $\geq 60\%$** and **normal sperm morphology count of $\geq 70\%$** with the absence of significant numbers of foreign cells such as leucocytes then the bull is deemed SATISFACTORY in section 2 of the PBE certificate.

2.SEMEN EXAMINATION			
Collection method	EEJ <input type="checkbox"/>	AV <input type="checkbox"/>	MASSAGE <input type="checkbox"/>
Appearance/density	Creamy <input type="checkbox"/>	Milky <input type="checkbox"/>	Watery <input type="checkbox"/>
Gross motility	/5		
Progressive motility	%		
Morphologically normal sperm	%		
Overall result: SATISFACTORY*/UNSATISFACTORY*			

4. SECTION 3 - ASSESSMENT OF MATING ABILITY

On the BCVA Bull PBE Certificate it must be indicated in this section whether or not the bull has been assessed for normal service behaviour and mating ability. In most cases this will not be done unless requested by potential purchaser or if required for an infertility investigation. In North America and Australia, libido and serving capacity are often assessed using serving capacity tests which involve counting the number of successful services that bulls achieve with restrained teaser females in a fixed time period. Serving capacity tests are not used in the UK therefore to assess mating ability a suitable oestrus female must be available on the day of the PBE. The bull should be observed achieving intromission and ejaculatory thrust with at least one service within 10 minutes of being introduced to an oestrus female expected.



The assessment of normal libido is extremely difficult with a single observation and may be influenced by many factors such as age, experience, inhibition by another dominant bull and concurrent disease therefore it is critical that farmers regularly observe bulls during the mating period to assess mating behaviour.

Similarly, bulls may be observed serving normally during a PBE or at the start of mating but subsequently acquire a problem such as spiral deviation or hind limb/back pain that will prevent normal intromission. It is important that purchasers/owners realise the limitations of the PBE with respect to this section and appreciate that just because a bull has passed physical and semen quality standards this is not a guarantee that it will perform as required when mixed with breeding females.

3.ASSESSMENT OF MATING ABILITY
This Bull has/has not* been observed exhibiting normal service behaviour and mating ability
normality detected
*Delete as Required

5. SECTION 4 - CLASSIFICATION

There are 3 possible classifications of bulls in the BCVA Bull PBE Certificate. Bulls that are certified **SUITABLE FOR BREEDING** must have passed the physical examination standards and have semen with individual progressive motility $\geq 60\%$ and $\geq 70\%$ morphologically normal sperm. If the mating ability section 3 has not been carried out this must be made clear by deleting the appropriate classification. An unsatisfactory finding in one or more of the sections means the bull must be classified as **UNSUITABLE FOR BREEDING**. The comments box on the certificate can be used to give more information on the reason/s for the bull being classified as **UNSUITABLE**. For conditions such as interdigital fibroma the bull is classified as **UNSUITABLE** however by pointing out that this is the only abnormality detected a potential purchaser may still choose to purchase and use the bull. Similarly with bulls that fail due to marginal sperm morphology but no physical defects it may be suggested in the comments box that the bull is reassessed after 60 days at which time sperm parameters may have returned to normal.

4. CLASSIFICATION	
In my opinion, in terms of the standards detailed on the reverse of this certificate, the examination findings would indicate that on this date the bull identified above is:	
SUITABLE for BREEDING* (based on meeting the requirements of sections 1 and 2 only) – mating ability has NOT been assessed	
SUITABLE for BREEDING* (based on meeting the requirements of sections 1, 2 and 3)	
UNSUITABLE FOR BREEDING*	
<small>NB: This certificate does not include any testing for infectious/contagious diseases, the results of which should be reported separately.</small>	
COMMENTS:	
Name of Veterinary Surgeon: MRCVS Date:	
Signed:	
Practice stamp/address: Certificate No.	





Summary

The Pre-Breeding Examination described can be used to screen bulls prior to sale or use to reduce the risk of unsuit-able bulls being used for breeding. It must be accepted that the correlation between physical traits, semen motility and morphology standards and fertility is not clear cut however as a risk reduction strategy it must be seen as good practice to examine all bulls prior to breeding. In the author's experience it is not unusual to find one bull in five (20%) with a physical problem of the reproductive tract and /or sub-standard semen quality that will render them sub-fertile or sterile. Identifying these bulls must be considered an important part of herd fertility management. Farmers must be encouraged to observe bulls closely during the mating period and record service dates and returns to heat if possible to allow early detection of problems that may occur despite pre-breeding examination of bulls. Veterinary surgeons must invest in the correct equipment and carry out PBE to a high standard using repeatable methods to ensure the process has credibility.

Further Reading

For those interested in this subject the following texts provide excellent further reading material and have been re-ferred to extensively by the author during the preparation of this manual.

Bull Breeding Soundness Evaluation. 3rd ed. Barth A D. Published by Western Canadian Association of Bovine Practitioners, Saskatoon, Canada, 2013 info@wcabp.com

Evaluation of Bulls for Breeding Soundness 1st Ed - Guidelines for the Societies of Sheep, Beef and Dairy Cattle Veterinarians of the NZVA. Parkinson T and Bruere N. Pub Vet Learn, Massey University, Palmerston North, New Zealand, 2007

Evaluating and Reporting Bull Fertility. Entwistle K and Fordyce G. Published by The Australian Association of Cattle Veterinarians, Indooroopilly, Queensland, 2003

Abnormal Morphology of Bovine Spermatozoa - Barth A D and Oko R J . Iowa State University Press, Ames 1989



APPENDICES

Appendix 1

BCVA Bull Pre-Breeding Examination Certificate

Date of Examination: _____



Owner:	
Address:	

BULL	Name:
	Ear number :
	Breed :
	Date of Birth/Age:

Reason for examination:

Pre-sale Check ☐

Pre-breeding Check ☐

Examination for Insurance Purpose ☐

1. PHYSICAL EXAMINATION

Body Condition Score (1-5)		
	*NAD	*Abnormal
Heart/Lungs	<input type="checkbox"/>	<input type="checkbox"/>
Eyes	<input type="checkbox"/>	<input type="checkbox"/>
Incisor/dental pad alignment	<input type="checkbox"/>	<input type="checkbox"/>
Musculoskeletal system	<input type="checkbox"/>	<input type="checkbox"/>
External genitalia	<input type="checkbox"/>	<input type="checkbox"/>
Internal accessory glands	<input type="checkbox"/>	<input type="checkbox"/>
Scrotal circumference		cm
Overall result:	SATISFACTORY*/UNSATISFACTORY*	

2. SEMEN EXAMINATION

Collection method	EEJ <input type="checkbox"/>	AV <input type="checkbox"/>	MASSAGE <input type="checkbox"/>
Appearance/density	Creamy <input type="checkbox"/>	Milky <input type="checkbox"/>	Watery <input type="checkbox"/>
Gross motility	/5		
Progressive motility	%		
Morphologically normal sperm	%		
Overall result:	SATISFACTORY*/UNSATISFACTORY*		

3. ASSESSMENT OF MATING ABILITY

This Bull has/has not* been observed exhibiting normal service behaviour and mating ability

*Place X in box to indicate findings.

NAD = No Abnormality detected

*Delete as Required

4. CLASSIFICATION

In my opinion, in terms of the standards detailed on the reverse of this certificate, the examination findings would indicate that on this date the bull identified above is:

SUITABLE for BREEDING* (based on meeting the requirements of sections 1 and 2 only) – mating ability has NOT been assessed

SUITABLE for BREEDING* (based on meeting the requirements of sections 1, 2 and 3)

UNSUITABLE FOR BREEDING*

NB: This certificate does not include any testing for infectious/contagious diseases, the results of which should be reported separately.

COMMENTS:

Name of Veterinary Surgeon: MRCVS

Date:

Signed:

Practice stamp/address:

Certificate No.

BCVA Bull Pre-Breeding Examination - Certification Guidelines



The aim of this certificate is not to guarantee bull fertility but to reduce the risk of potentially unsuitable bulls being used for breeding. A bull that meets the requirements for section 1 and 2 should have no obvious physical abnormality that would render it unsuitable for natural service, and have the potential to be fully fertile based on semen quality.

NB. This certificate does not include any assessment of health status. Any tests carried out for infectious diseases (e.g. BVDV) should be reported separately.

Section 1: PHYSICAL EXAMINATION

To meet the requirements of this section the bull will have to demonstrate freedom from significant physical defects that could affect fertility or the ability to mate and freedom from heritable defects that could affect progeny.

Body condition/clinical examination

Should be assessed on a 1-5 scale where 1 is very thin and 5 is obese. Bulls in very thin body condition score, 2 or less, should be classified as unsatisfactory. The heart and lungs should be auscultated.

Jaws/eyes

Bulls should be inspected for severe over/undershot jaw and gross ocular defects such as cataracts, carcinomas etc which may interfere with vision and ability to seek out females.

Musculoskeletal defects

Bull should be inspected for evidence of lameness whilst walking on a smooth level surface. Lamé bulls will fail section 1 and be classified as unsatisfactory. Bulls with severe conformational defects of the limbs e.g. post-hock, sickle-hock, valgus deformity or serious foot defects such as corkscrew claw or interdigital fibroma should be classified as unsatisfactory in section 1.

External genitalia

The scrotum and contents should be carefully palpated and scrotal circumference measured (see below). Bulls with gross physical abnormalities such as epididymitis or orchitis would be classed unsatisfactory. Slight variation in size and position of testicles is acceptable though breed standards may vary, and bulls may be rejected at pre-sale society inspections if any variation in size/shape is present.

The sheath/penis should be palpated for swellings, adhesions, discharges, papillomata etc. The tip of the penis should be inspected for normality and if it has not been visualised during semen collection this should be noted in "comments".

Scrotal Circumference (SC)

Breed society standards, where published, should be used as minimum SC standards to meet the requirements of this part of the examination. SC standards given below (Society for Theriogenology) should be used when no alternative breed standards are available.

Age months	12-15	>15≤18	>18≤21	>21≤24	>24
Min. SC	30cm	31cm	32cm	33cm	34 cm

Internal accessory sex glands

Seminal vesicles, prostate and ampullae should be palpated per rectum to check for any abnormalities.

Section 2: SEMEN EVALUATION

The semen samples must meet a minimum set of standards as detailed below.

Gross Motility

Scored on a 1-5 scale. A good semen sample would normally score at least 3. However, as gross motility is influenced by the concentration of the sample, the assessment of progressive motility is required for all bulls.

Scale	Description
1	No swirl; generalised oscillation of individual sperm only
2	Very slow distinct swirl
3	Slow distinct swirl
4	Moderate fast distinct swirl; dark waves
5	Fast, distinct swirls with continuous dark waves

Individual Progressive Motility

The minimum requirement is a progressive motility of at least 60%. Bulls with gross motility scores of 3-5 would normally be judged satisfactory for progressive motility if semen is handled well and examined on a heated stage. Bulls that score <60% for progressive motility should have a second ejaculate collected immediately to rule out sperm accumulation and senescence as a potential cause. Continued failure to achieve ≥60% progressive motility will normally be caused by a high percentage of morphologically abnormal or dead sperm which will be confirmed at the next stage of the examination.

Semen Morphology

100 sperm cells should be counted using X1000 oil immersion microscopy with nigrosin/eosin smears or wet preparation phase contrast. To meet the requirements of this section 70% or more of sperm should be morphologically normal, with no more than 20% showing nuclear defects. In marginal cases (65-70%) at least 2 counts of 100 sperm cells should be carried out.

Bulls with no apparent physical abnormality of genitalia but with marginally poor morphology should be classed as unsatisfactory. However a note can be added to the comments section suggesting re-examination after 60 days when recovery may be evident if temporary degeneration has been the cause. Young bulls aged 12-15 months may have a poor morphology count due to immaturity and can be re-assessed after 2-3 months.

Section 3: ASSESSMENT OF MATING ABILITY

As libido is difficult to assess and define, this part of the examination simply confirms whether the vet has observed normal service behaviour and intromission when the bull was presented with a female in oestrus. At least one successful service within 10 minutes of being presented to an in-oestrus female should be expected. If this part of the examination is not carried out, then bulls can still be classified as SUITABLE FOR BREEDING based on meeting the requirements of parts 1 and 2 only. The onus is on the owner/purchaser to observe the bull closely at the start of breeding period to monitor libido and mating ability.

Section 4: CLASSIFICATION

Any bull classified as SUITABLE FOR BREEDING must meet the requirements of the physical examination and achieve minimum standards for scrotal circumference, progressive motility (60%) and sperm morphology (70%).

Appendix 2

1

PASS / FAIL

BULL PRE- BREEDING EXAMINATION RECORD

Owner :

Date:

Reason for test:

Identity (Name & Eartag)			
Breed			
Age/DoB			
Condition Score (1-5)			
Physical inspection		NAD or COMMENT	
Heart /lungs			
Eyes			
Jaws			
Musculosekletal system			
External genitalia – penis/sheath			
Scrotal circumference (cm)			
Internal accessory sex glands			
Palpation of testicles & epididymides	Right:		
	Left:		
		1 st Sample	2 nd Sample
Penis Extruded (Y/N) OK?			
Volume (ml)			
Appearance/density		Creamy Milky Watery	Creamy Milky Watery
Gross Motility (1 – 5)			
Motility – Progressive Linear (%)			

Bull Semen Morphology Examination Record

Sperm Morphology	%	
Normal		
Head defects		
Coiled tails/dag defect		
Distal midpiece reflex /bent tails		
Coiled principal piece		
Proximal droplets		
Detached heads		
Other		
White cells etc		
Total abnormal %		

SUMMARY OF EXAMINATION

Overview of Pedigree Bull Production

- Is it conducive to good fertility?

Doreen Corridan, Munster Cattle Breeding Group. Mallow, Co. Cork

In Ireland the latest statistics available for a complete year are for 2016. In 2016 there were 1.36 million calves born in the dairy herd, 40-50% were sired by dairy AI bulls, 5-10% by beef AI bulls, 10-20% by dairy stock bulls and 20-30% by beef stock bulls. Therefore, stock bulls sire 30-50% of calves born to the dairy herd. In 2016 there were 924,701 calves born to the suckler beef herd, 20-25% were sired by beef AI sires and the 75-80% were sired by beef stock bulls. In total 40% of the dairy herd and 80% of the beef herd are bred to stock bulls. The number of beef calves born in dairy and suckler herds in Ireland in 2016 by the sire breed of the calf is summarised in Table 1.

Table 1. Number of beef calves born in dairy and suckler herds in Ireland in 2016 by the sire breed of the calf.

Sire Breed	AA	BA	BB	CH	HE	LM	SA	SH	SI	Total
Suckler herd	94,316	8,814	31,620	327,902	37,783	349,257	12,641	11,470	50,898	924,701
Dairy Herd	249,399	709	26,021	13,543	166,453	62,973	3,183	3,707	16,607	542,595

Pedigree bulls are used exclusively except in exceptional cases. The breed profile of sires used in the dairy herd are Angus and Hereford predominantly, in the suckler herd Limousin and Charolais dominate. Of the 924,701 beef calves born 37,533 were pedigree. Ireland is unique in that apart from the pedigree herds which produce the bulls for service the majority of the suckler beef cows are crossbred. The number of pedigree registered calves born in 2016 and the number of breeders in Ireland is summarised in Table 2.

Table 2. Number of pedigree registered calves born in 2016 and the number of breeders in Ireland.

Breed	Number of Breeders	Number of Pedigree Calves Born
Hereford - HE	576	4504
Shorthorn - SH	218	1206
Angus - AA	1200	9311
Charolais - CH	1582	8675
Simmental - SI	500	2524
Limousin - LM	1540	9227
Blonde d'Aquitaine - BA	39	371
Belgian Blue - BB	19	377
Piedmontese - PD	14	218
Aubrac - AU	102	600
Salers - SA	114	920
Parthenaise - PT	32	335

The pedigree herds that produce stock bulls have on average six pedigree calves born per year. In 2015 a Beef Data & Genomics Programme (BDGP scheme) was introduced in Ireland; this has led to a substantial increase in recording and genomic testing in these herds. A total 25,000 herds have enrolled, and 1,250,000 genomic samples have been collected to-date. For the purposes of this paper, I restricted the dataset to bulls that were sold to herds in the BDGP scheme to ensure valid data.

Of the 16,000 pedigree bulls born in 2013, 8600 were sold to herds for breeding and of these 2750 were sold into BDGP herds. The reason why I restricted to BDGP herds was; (i) we know the herds are recording all their progeny ancestry and performance, (ii) the bulls are genotyped (to confirm ancestry) and (iii) for stock bulls the herd-owners record their reasons for culling or selling. Overview of Pedigree bulls used in BDGP herds born since 2013, including number of progeny born to May 2018 is summarised in Table 3.

Table 3. Overview of Pedigree bulls used in BDGP herds born since 2013, including number of progeny born to May 2018.

Bull Details	Number Progeny Born								
Breed	Number	%	Total	Average	Zero	1 - 20	21-50	51-100	>100
AA	270	9.8%	12,322	45.6	28	40	86	100	16
AU	15	0.5%	768	51.2	2	1	4	7	1
BA	34	1.2%	1,692	49.8		2	18	12	2
BB	54	2.0%	1,786	33.1	6	16	21	9	2
CH	1026	37.2%	49,942	48.7	26	117	442	391	50
HE	144	5.2%	6,758	46.9	9	29	44	48	14
LM	958	34.7%	48,585	50.7	23	89	405	388	53
PI	2	0.1%	86	43.0		1		1	
PT	27	1.0%	1,411	52.3	4	3	3	16	1
SA	44	1.6%	2,628	59.7			17	23	4
SH	25	0.9%	911	36.4	1	5	12	7	
SI	159	5.8%	8,073	50.8	3	21	59	65	11
Total	2758	100.0%	134,962	48.9	102	324	1111	1067	154

A total of 102 bulls out of 2758 failed to leave any progeny. An overview of Pedigree bulls used in BDGP herds born since 2013, including alive/dead status to May 2018 is summarised in Table 4.

Table 4. Overview of Pedigree bulls used in BDGP herds born since 2013, including alive/dead status to May 2018..

Breed	Grand Total	Dead/culled	%	Alive
AA	270	90	33%	180
AU	15	2	13%	13
BA	34	8	24%	26
BB	54	8	15%	46
CH	1026	215	21%	811
HE	144	47	33%	97
LM	958	185	19%	773
PI	2	2	100%	
PT	27	7	26%	20
SA	44	11	25%	33
SH	25	9	36%	16
SI	159	56	35%	103
Total	2758	640	23%	2118

An overview of reasons for culling of Pedigree bulls used in BDGP herds born since 2013 to May 2018 is summarised in Table 5.

Table 5. Overview of Pedigree bulls used in BDGP herds born since 2013, Summary of reasons for culling from BDGP herds to May 2018.		
Reasons for culling	Bull Count	% total
Docility	26	8.5%
Low Euro-Star values	33	10.8%
Feet/legs & lameness	67	21.9%
Risk close mating's	51	16.7%
Injury	71	23.2%
Infertility	22	7.2%
Poor progeny	11	3.6%
Surplus to requirements	25	8.2%
Total	306	100.0%

A total of 23% of the bulls born in 2013 have been culled to date. The main reasons for culling were injury 23.2%, followed by feet/legs & lameness at 21.9%. Infertility accounted for 7.2% of bulls being culled. This figure may have been higher in the past as in the past five years the number of bulls being fertility tested each year has increased substantially and most breed society sales now have a requirement for bulls to have passed a presale fertility test. However, there are 66 bulls alive born in 2013 without any recorded progeny.


Pedigree beef bull production in Ireland

Pedigree bulls are purchased in Ireland from 12 to 18 months of age, the majority are purchased privately on farm from the breeder, 10-15% are sold at sales. In general prices range from €2,000 to €5,000 with a small number of exceptional bulls selling for more than €10,000. Usually it is the first prize winners or the champions at the presale show that command the higher prices. Often these bulls have been shown throughout the summer at shows as calves from six months of age onwards. As pedigree breeding is not a very profitable enterprise (margins are similar to other beef systems in Ireland, without the single farm payment margins of - €150 to + €250 per hectare not including a labour charge), breeders focus on achieving success at the presale shows to maximise their sale price. These shows are judged purely on visual looks and the prize winners will have gained in excess of 2kg/day from birth and need to exhibit extreme muscling and a 4-5 fat score for success, many purchasers apply the same criteria when purchasing on farm. Many of these prize-winning bulls are more fit for slaughter than fit for service.

To maximise fertility, bulls, need to grow naturally at 1-1.5kg /day to ensure optimum development. The level of feeding required from an early age to compete at the presale shows at 12 – 15 months has a negative impact on the musculo-skeletal system, hoof quality, liver and testicular function.

This is compounded by the fact that purchasers buy their bulls literally the week they need them to breed cows. Often these bulls go from receiving more than 10 kgs of concentrates per day in a confined pen without any exercise to being let run at pasture with breeding females and without supplementary concentrates. These bulls fatigue quickly and lose weight rapidly often 50-100 Kgs in 30-60 days, this has a direct adverse effect on fertility.

Purchasers are reluctant to purchase their bulls 2-3 months in advance to allow time for acclimatization to their new home, time to reduce the level of concentrates to a predominantly forage diet, time for vaccination pre-service and time for the bull to be trained to serve. In addition,



they rarely have adequate bull power available with the result of having too high a ratio of females to bulls.

In Ireland especially in seasonal calving dairy herds herdowners prefer to purchase a young bull each year often at 12-15 months and sell them at the end of the season as opposed to keeping a mature bull. Often the reasons given are the hassle of having bulls on a dairy farm, the lack of appropriate handling facilities for the winter and the fear of aggression from mature bulls. There is a distinct lack of confidence in handling mature bulls in Ireland, consequently there is an over reliance on young bulls. Stock bulls are rarely monitored for quality of their progeny and sub fertile bulls often go unnoticed. The value of a mature bull that is delivering superior progeny and a high 6-week incalf rate are not highlighted. Poor progeny only accounted for 3.6% of culling in BDGP herds.

The main reason for culling from BDGP herds was injury 23.2% and feet/legs & lameness at 21.9%. The majority of injuries can be avoided by not allowing bulls run with females until they are confident at mating, having the appropriate ratios of females to the bull and taking cognisance of the bulls age, care in avoiding fat bulls that fatigue easily and not over working young bulls. Issues with feet and legs can be greatly reduced by growing bulls naturally and avoiding over feeding at an early age, managing nutritional transitions, avoiding excessive weight loss and over working young bulls. Docility was at 8.5% as a reason for culling, docility can be increased easily by genetic programmes. However, due to the small herd size and the level of handling in pedigree herds it can be difficult to get an accurate assessment of the genetic merit of a bull for docility. Lack of confidence in handling of mature bulls also allows bulls to get rewarded for poor behaviour, continued poor behaviour leads to issues in handling them, resulting in herdowners culling bulls for perceived docility issues when it was poor stockmanship that was the main issue.

Summary

In Ireland due to our integrated database, progeny test programmes the usage of genomics and with fertility contributing 30%+ of the index, female fertility has increased dramatically. Male fertility has literally been ignored. However, there is an opportunity here to gather data, from herds on stock bull performance, from veterinarians on bull soundness examinations and feed it into the database to develop a male fertility index.

We need a cultural change to encourage herdowners to purchase bulls in advance, increase the age at first service, rely more on the index rather than relying on a high level of feeding to estimate the genetic potential, allowing young bull's time to develop mating ability and finally have enough bull power on farms to allow for the correct ratio of males to females. Stock bulls need to be monitored more for the quality of their progeny and their fertility; this will lead to an appreciation of the economic value of a mature bull that is delivering superior progeny and a high 6-week incalf rate.

Problems with bulls in Artificial Insemination Centres

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Introduction

Objective of the AI centre is to get as much high quality germplasm from the highest genetic merit (demand) bulls as possible. This differs slightly from the requirements for a bull for use in natural service as the bull in an AI centre must meet stringent health standards and the semen must be capable of been frozen and thawed and still retain high fertility. The following is a summary of problems that affect bulls in the National Cattle Breeding Centre (NCBC).

Current practices

At the National Cattle Breeding Centre (NCBC), each year we create a pipeline of young bulls of different breeds for the different breeding programmes. Bulls are selected and brought to rearing units where they are isolated in small groups. At approximately 10 months of age they are moved to the Quarantine Barns at one of the collection centres and from there, subject to meeting the required health standards, they move into the semen collection centre. Bulls are eliminated from the process at various stages and for different reasons. The largest group to be eliminated is the group of bulls that are not selected based on their genomic evaluations or health status. From an initial screen of 4,000 bulls for which genomic evaluations are completed, only 50 were selected to remain in the programme. In addition to genomic and health screening, bulls are also screened for lethal genetic defects such as Bovine Leucocyte Adhesion Deficiency (BLAD), Congenital Vertebral Malformation (CVM) and Brachyspinia and all carriers are all removed from the programme.

Early life management of bulls


Bulls in the rearing unit are managed to exploit “the importance of early life nutrition in regulating the timing of puberty in both bulls and heifers” (see Kenny et al., 2017). There is significant evidence that an improved metabolic status in early calfhood, advances maturation of the hypothalamic–pituitary–gonadal axis, therefore, facilitating earlier sexual development, onset of puberty and production of semen. Some bulls don't enter the system until they are older than 6 months and at that stage it is too late to advance puberty by exploiting the benefits of early life nutrition.

Health requirements

There are specific health requirements that must be met in order for a bull to enter an approved EU semen collection centre and failing these can result in a small number being eliminated. The current legislation is about to be updated which may lead to further changes and impact on bulls entering a semen production centre. A particular problem when purchasing young bulls for the AI programme is managing possible test issues for maternally derived antibodies for the regulated diseases.

Management of young bulls

At NCBC, once young bulls reach 8-10 months of age they are assembled into small groups, screened for disease and moved to a quarantine facility where they are further screened for diseases including Campylobacteriosis and Trichomonosis. On completion of a minimum of 28 days in quarantine the bulls are moved into the barns. In the barns the bulls are housed in individual pens (on occasion with young bulls they may be housed 2 per pen for a short period this can be helpful for mounting



observations). We do not have any females in the bull barns and we do not use mechanical teasers or electro ejaculation, rather, bulls are trained to mount other bulls. Training to mount consists of releasing bulls into an open area in groups of 3 to allow interaction. Once the bulls start to mount they are encouraged to “jump” into the artificial vagina (AV) as quickly as possible to provide reward and avoid bulls becoming bored with mounting.

In order to minimise stress on the bulls and to create a good relationship with the stockmen, very young bulls do not have a nose ring inserted and do not undergo halter training until they have produced enough semen for their AI breeding programme (usually 800 doses). The training process is ideally carried out in the collection area to establish a positive perception of the area for the young bull. We allow the bull to mount no more than 3 times and if the bull fails to ejaculate after 2 attempts with the AV we do not persist and return the bull to his pen to avoid creating bad habits. Over-use of the AV in young bulls can also cause irritation to the penis.


Semen collection in young bulls

In preparing the AV for the young bulls we tend to use a lower temperature 35-37°C compared to the mature bulls (37-40°C). The AV liner is smeared with Vaseline jelly. We do not use different size AV for young and mature bulls. However, for young bulls we may add air to the water chamber to increase the sensation. Young bull collection frequency typically starts at once per week, the frequency will increase as the semen density increases and the numbers of immature sperms reduce in the ejaculate. A small percentage of young bulls fail to give semen or give semen of insufficient quality in their first season. Usually these bulls are removed because they will be superseded by the next intake of genetically superior bulls.

We select teasers for young bulls based on temperament, stature and “attractiveness” to the young bulls. Typically, bulls from the Jersey breed are popular for young dairy bulls. Beef bulls are different because they are typically older at time of intake (past puberty) and have had a nose ring inserted and are halter trained. Most beef breed sires are overweight at time of selection and require a period of “slimming down”. Initially they are collected up to 3 times per week to “clean them out” and during this period the typical ejaculate is high density, large volume and poor quality. Once good quality sperms are observed, typically the frequency of collection for a young bull is reduced to twice per week but this is usually assessed on a case by case basis. Older bulls and bulls on very frequent collection can have reduced libido, though it may still be important to maintain this frequency of collection because of demand. We employ a number of tactics including, holiday (give the bull a break from collection), change the teaser (change bull, breed, colour), change the collector, change location (new barn or collect in a different location in the same barn). Some bulls respond to these tactics others may only exhibit a temporary improvement.

Aggression H&S statement

One of the key concerns for any bull stud is the safety of the staff and animals. We categorise the risk from bulls depending on our assessment based on observation. Aggressive bulls are clearly identified through their territorial aggression, pawing the ground, bellowing etc. However, once haltered and exposed to high levels of interaction with the staff they usually submit. Another type and potentially more dangerous group of bulls are “nervous bulls”. These bulls do not usually exhibit the normal signs of aggression but can react aggressively when startled. Managing this group involves frequent handling to avoid the bull over-reacting in the presence of a handler. However, a small number of bulls are unmanageable and pose a significant risk to staff these bulls are removed. All bulls are monitored for aggressive behaviour on entry and during their entire stay as their categorisation may change with time. Our observations, based on the housing systems across all 5 barns at NCBC, has been that in the modern barns, where we use mechanical cleaning,



bulls tend to be more nervous compared to bulls in the barns where cleaning-out requires a higher level of bull people interaction.

Main veterinary issues at the stud

Hoof Care: Hoof trimming is carried out on a case by case basis; corrective trimming may need to be carried out more frequently on beef bulls that were overweight before entering the stud. Similarly, bulls that have been housed on deep litter bedding for prolonged periods need frequent hoof trimming.

Injury: The most common injury encountered is a self-inflicted injury. This typically occurs when bulls are playing (buck leaping) in their pens. On very rare occasions, bulls can injure themselves or other bulls in the collection area.

Lameness: As the age profile of the of the bulls has changed to younger due to the use of genomic selection the frequency of lameness and injury has reduced significantly.

Sexual health issues

Penile warts: We receive young bulls who have or develop penile warts at an approximately rate of 1.5% / year. To-date all cases have been in the Holstein Frisian population. Large warts are removed (ligation) and most cases resolve and once the warts are gone the bull is allowed to proceed to enter a collection barn and a semen collection routine. We have had 12 cases in the last 10 years and only one case failed to clear up.

Vesiculitis: This presents as pus and blood in the ejaculate and a sample is submitted to the pathology laboratory to identify the pathogen and its sensitivity to antibiotics and a treatment programme is developed. Our experience has been that bulls tend to have recurrent infection or fail to respond to treatment. The incidence of vasculitis in our stud has decreased over recent years.

Diet

We feed a high dry matter diet such as haylage at 70% DM and grass nuts (not silage at 40% DM) to reduce the liquid waste. This also keeps the bulls cleaner for semen collection and reduces the odours in the barn.

Semen Laboratory Issues

While considering the reasons we reject individual ejaculates and straws from bulls or even reject bulls from the programme it is probably worth noting that the process of applying “criteria” for developing a semen quality standard has evolved over time and been built on the grounds of feedback from huge datasets from *in vivo* field fertility. The fertility model used is based on a predictive model of assumption of pregnancy and is constantly adjusted as more information on the bulls enter the database. If we look at a cohort of bulls in current production with over 100 insemination records, average phenotypic pregnancy rate for 2017 is 62%±0.001% and average overall pregnancy rate (including calving data) was 57%±0.01% for 2017. There are differences between breeds as shown in Table 1 (P<0.05).

In order to consistently maintain high fertility we assess all parameters with possible implications on semen quality. Semen from the bulls is transported to the laboratory for processing and there are a number of reasons that semen ejaculates can be rejected at this stage including:

Semen Assessment: The aim of semen assessment and quality control checks is to ensure only the very best quality semen is released to maintain a consistent level of fertility in the field. Semen

Table 1. Mean Phenotypic Pregnancy Rate across Different Breeds used for Conventionally Frozen Inseminations between 2014 and 2017.					
Breed	No. of Animals	Count of all conventional frozen straws analysed (2014-2017)	Average phenotypic pregnancy rate for all frozen Straws (2014-2017)		
Simmental	6	26948	53.7 ±	3.81	^a
Charolais	8	82286	53.1 ±	2.44	^b
Limousin	15	126174	54.1 ±	1.56	^a
Hereford	11	128413	57.3 ±	2.09	^{bc}
Aberdeen Angus	10	203907	57.4 ±	1.93	^{abc}
Holstein	95	858614	58.0 ±	0.70	^{bc}
Friesian	9	43447	60.8 ±	2.01	^{ac}
Total	154	1469789	56.3	2.08	

^{abc} Values with different superscripts differ (P <0.01; values are mean ± sem). Reference: Sire_31JAN2018_FOR_NCBC data file published by the Irish Cattle Breeding Federation (ICBF).

is monitored for the percentage of live sperms and motility. Motility is also given a “type” score. This is a five point motility scale with 5 being the most forward progressive and zero being the least progressive.

Morphology: This is also part of this assessment, we observe impacts on motility associated with gross morphology problems such as bent tails, coiled tails, head shape, droplets etc.

Quality Control: There are two quality control checks, the first is on initial assessment (raw semen) which determines whether the ejaculate will be processed. The second is after the freezing and thawing process.

Over the past 6 years we have carried out and number of research trials, internal assessments and data analytics to identify how we can reduce rejection rates of ejaculates and straws while maintaining field fertility. In order to do this we had to first identify where potential variation could arise and so we have carried out experiments to address the following potential sources of variation in our system. Some findings from our research are summarised below.

Temperature Control in transit: Temperatures between 15 and 20°C are optimal transit temperatures for fresh semen prior to processing and although ranges between 5 and 15°C yielded acceptable results, temperatures below 5°C had a significant negative impact on semen quality and subsequent 60 day non-return rate (Murphy et al., Submitted to Journal of Dairy Science).

Addition of extender at the barns (Transport Media): It was observed that raw (undiluted) semen transported to the laboratory underwent cell shock as evidenced by dramatic coiled tail morphology. Once samples were diluted in a 1:1 dilution of extender/semen (vol/vol) immediately post-collection at the barns, we observed normal acceptance rates of ejaculates and straws during the Quality Control process following between 1 and 3 hrs of transit to the laboratory.

Rejection rates: Overall rejection rates were assessed in order to determine the main reasons for rejection. Individual ejaculates were categorised into rejected at the barn, rejected at initial Quality Check (QC), rejected at post-thaw QC, rejected other or accepted and these accounted for 0.6%, 2.7%, 20.5%, 0.03% and 76.2%, respectively (n=15,278 ejaculates). Of the ejaculates rejected in the barns there was no significant difference between barns ($P>0.05$), however there was variation between technicians collecting ejaculates ($P<0.05$).

Initial QC Rejections: The majority of ejaculates rejected on initial QC (n=412) was due to reduced progressive motility scores or gross morphological abnormalities with beef breeds presenting with a significantly higher rejection rate than dairy breeds (46.4% and 40.5% versus 5.6% and 7.3% for both motility and morphology for beef versus dairy bulls, respectively; $P<0.05$).

Final QC Rejections: The majority of rejected ejaculates following the freeze-thaw process and insult to sperm cells (n=3134). Total progressive motility was the main factor leading to ejaculates not passing QC and accounted for 71% of rejections. The remainder was based on tail defects, head defects, gross morphology, droplets, processing errors and non-conforming products (17.4, 3.8, 4.4, 1.8, 0.9 and 0.7%, respectively). Interestingly, there were significant differences between breeds and between bulls for incidence of morphological abnormalities leading to rejection of those ejaculates ($P<0.05$).

Technician Effect: There was no effect of laboratory technician on rejection rate at post-thaw QC, although there was a difference between laboratory technicians in rejection rate on initial QC.

Freeze time: The quality of the semen and subsequent rejection rate was not affected by time between processing straws and freezing straws (3 hrs versus 21 hrs; $P>0.05$).

Extender batch control (egg yolk): The choice of diluent was also scrutinised and it was found that egg yolk batch is a source of variability on weekly rejection rates ($P<0.05$). As a result several alternative extenders were tested for both *in-vitro* and *in-vivo* trials. No superior product to that in current use was identified within these trials (Murphy et al., In Press Animal Reproduction Science).

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Identifying Problem Bulls at Farm Level.

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Introduction

Grass-based dairy and beef production demands highly seasonal calving patterns immediately in advance of or at the start of the grazing season. This requires a rapid resumption of oestrous cyclicity in cows following calving, high oestrous detection and submission rates in herds where AI is used and high bull fertility irrespective of using AI or natural service. Irish Cattle Breeding Federation (ICBF) data would suggest that almost 50% of the calves from dairy cows and 77% of the calves from beef cows are sired by natural service bulls. This obviously reflects the convenience of using natural service bulls and also the importance of the natural service bulls in both the dairy and beef industries. The fertility of bulls is of paramount importance. However, in herds dependent on natural service bulls to get cows pregnant, it's simply not just about turning in a bull with a herd of cows and expecting calves to arrive 9 months later. International data would suggest that up to 4-5% of bulls are infertile at any one time with a further 20-30% being classified as sub-fertile. Both conditions can have major effects on herd reproductive, productive and economic performance particularly where these conditions go undetected for a number of weeks. This paper describes practical aspects of conducting a Bull Breeding Soundness Evaluation (BBSE) at farm level and summarises the results from more than 500 bulls tested over the past 10 years..

Why a Bull Breeding Soundness Evaluation?

Bull breeding soundness evaluation (BBSE) is a valuable veterinary tool for identifying problem bulls. This may be done as a routine procedure pre-breeding or pre-sale, or it may be done where there is reason to suspect that the bull is not performing as well as expected, where an unacceptable number of repeats are observed in the cows.

Infertility - an inability to achieve pregnancy - is very rare in bulls, but sub-fertility where the bull can achieve pregnancies, but not at optimal levels is quite common. Most studies would show that 20-25% of bulls are sub-fertile. There is a huge potential benefit for the dairy and beef industries if these sub-fertile bulls can be identified and removed from breeding programmes.

What does a Bull Breeding Soundness Evaluation involve?

It is vital that the BBSE is performed to a high standard, and that there is a high level of consistency among vets in the way the BBSE is carried out. This should include examination of the heart and lungs, eyes, teeth, and the locomotion system. Both the external and internal sex organs should be evaluated including the penis, testicles, epididymides, prostate gland, seminal vesicles and ampullae. Special care should be taken to record the scrotal circumference accurately. Semen is collected and gross motility, progressive motility and sperm morphology is assessed.

A report or certificate is issued on completion of the BBSE, and the bull is deemed to be acceptable or unacceptable, or having passed or failed. There are minimum acceptable scrotal circumference measurements based on the age of the bull, from 30cm at 12 months of age to 34cm from 24 months and older. There are also internationally accepted parameters for measuring gross motility, progressive motility and sperm morphology. Failure to reach these targets, results in failure for the bull.

The age of the bull when the BBSE is performed is very important. Many bulls may not reach puberty until 15-18 months of age, which means if they are tested at a year old they will fail. In this situation retesting at a later date is recommended. This will not suit an owner who is trying to sell a bull, and in our situation in Ireland this is a common scenario every year when yearling bulls are being tested prior to the sales. Pressure may be applied by some owners to try to “pass” the bull, but as vets, we must use our knowledge of bull reproductive physiology and pathology to reach the correct conclusion. And if the bull has to be retested at a later date, then so be it.

Carrying out a BBSE

The BBSE may be performed on the farm or at the veterinary premises. On the farm a “lab” must be set up as close as possible to the cattle crush, so that the semen can be examined as quickly as possible after collection. Essential items including a microscope with heated stage, a water bath or heated block for keeping semen vials warm, electro-ejaculator with ancillary items such as slides, cover slips, stains, scrotal measuring tapes etc, must be brought on-farm and set up as well as possible. Unfortunately, there are very few ideal farms for bull testing and a degree of improvisation is usually required. In contrast, at the veterinary premises a higher quality microscope may be in use and generally practitioners are working in a much more controlled environment. However, if a farmer has a number of bulls to test it is unreasonable to expect him to bring them all to the surgery, so quite a number of BBSE’s are performed on farm every year. However, it is vital to remove or control as many variables as possible on the farm, and particularly to control the temperature of the semen from collection to evaluation. Otherwise erroneous conclusions can be made on the quality of the semen.

The quality of the microscope used is a limiting factor in the ability to assess the sperm. A basic microscope will suffice for evaluating gross motility of the semen; a microscope with phase-contrast is hugely beneficial for progressive motility, but the real benefit of the higher quality microscope is for assessing sperm morphology. Simply put, it is very difficult to identify some important and significant morphological defects with a poorer quality microscope.

Some results of BBSE examinations of bulls in Ireland

Over the past 10 years, this veterinary practice has carried out more than 512 BBSE on bulls. The bulls examined can be categorised as “pre-sale”, “pre-breeding” and “post-breeding”. In the latter category bulls would be examined on the basis of low recorded conception rates in herds that season. Pass, fail and inconclusive rates for bulls by category are summarised in Table 1.

Table 1. Pass, fail and inconclusive rates for bulls by category (499 bulls tested)				
No bulls Tested	Pre-Sale	Pre-Breeding	Post-Breeding	Overall
	225	154	133	512
No Pass	186	116	60	362
(%)	(83%)	(75%)	(45%)	(71%)
No Fail	37	35	71	143
(%)	(16%)	(23%)	(53%)	(28%)
No inconclusive	2	3	2	7
(%)	(1%)	(2%)	(2%)	(1.40%)

This is the first report of BBSE on Irish bulls. The highest fail rate was recorded in the post-breeding category, reflecting that these were a population of “problem” bulls. Almost 25% of bulls failed the pre-breeding test reflecting that some of these bulls may have low fertility in a previous season compared with 16% fail rate for pre-sale bulls. The pass, fail and inconclusive rates by breed of bull are summarised in Table 2.

Table 2. The pass, fail and inconclusive rates by breed of bull							
	AA	CH	He	LM	HOL-FR	Other	Overall
No Tested	109	66	97	48	140	50	510
No. Pass	89	31	65	27	114	38	361
(%)	(82%)	(47%)	(67%)	(50%)	(81%)	(76%)	(71%)
No. Fail	18	35	28	23	26	12	142
(%)	(17%)	(53%)	(29%)	(48%)	(19%)	(24%)	(28%)
No. inconclusive	2	0	4	1	0	0	7
(%)	(2%)	(0%)	(4%)	(2%)	(0%)	(0%)	7(1%)

In this practice the fail rate for Limousin and Charolais breed bulls tested was much higher than for any other breed examined and warrants further study to establish if this is a real issue with these breeds or is it specific to the sample of bulls examined in this practice area. Compulsory BVD testing started in Ireland in 2013. BBSE failure rates in all bulls by year are summarised in Fig1.

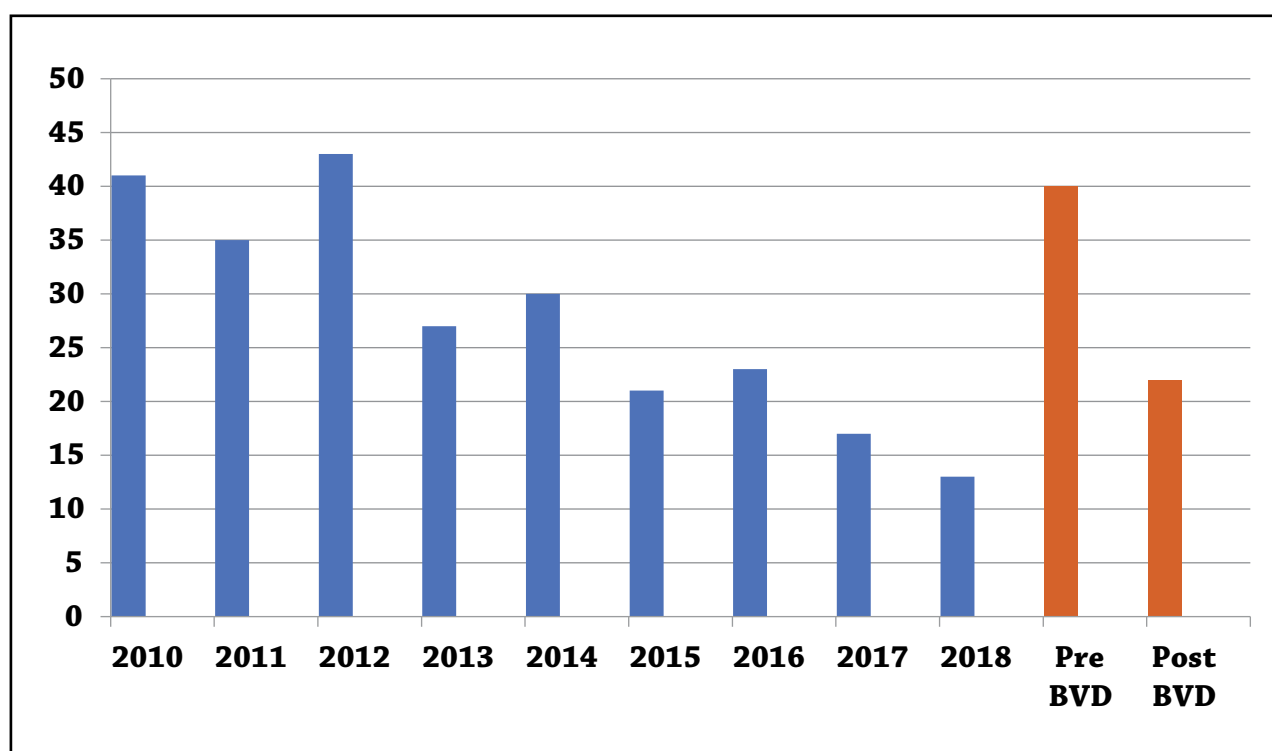



Fig1. BBSE failure rate in all bulls by year

It would appear that the fail rate for all bulls fell by almost 50% following the introduction of compulsory BVD testing. While this decline was evident in all the 3 categories of bulls tested the drop in fail rate was most evident in the pre-sale category which would be the youngest bulls examined. While this is an interesting observation, it is not possible to ascribe the decline in fail rate solely to BVD eradication.

Libido testing

In Ireland and the UK, unlike some other parts of the world, the libido and service capacity of the bull is not an integral part of the BBSE. A bull may have perfect semen but if his libido is low or if



his service capacity is poor then he will not fulfil his potential. To assess the libido simply put the bull in a pen with a cow in heat and observe. If the bull manages one or two successful services in a 10 minute period then libido is unlikely to be an issue. The service capacity is a measure of whether or not the bull jumps on the cow in the correct manner, or whether his ability to jump is impaired by any injury or arthritic conditions. It is not uncommon to see a bull with very good semen, and good libido, but who jumps on the front of the cow. These issues may not be completely black and white and repeated observations may be required to reach the proper conclusion.

Testing for vendor or purchaser

There may be a certain mental conflict in the mind of the bull tester depending on whether the BBSE is being done at the behest of the vendor or the purchaser. Testing for a vendor usually involves a pass or a fail, and the vendor may choose to retest the bull at a later date, or he may decide that the bull in question will never be up to the required standard and he can make alternative arrangements and feed the bull for slaughter. It is often more complicated when testing for a purchasing farmer. Frequently, this occurs when a purchased bull is not delivering the required results on farm. These bulls may fail a BBSE, and the new owner will usually contact the seller demanding compensation. If the vendor doesn't have a BBSE cert pre-sale, he often ends up replacing the bull and compensating the buyer, and sometimes this isn't necessarily fair!

We assume that the sperm evaluation at the time of the BBSE is effectively like a biopsy of the previous 10 weeks in the life of the bull's testes. A bull which has been reared in perfect bio-secure conditions on the farm of origin is then moved on to a farm that may have endemic BVD or IBR or some other infectious agents. If the bull becomes infected, the ensuing viremia or bacteraemia can adversely affect sperm production with the result that high levels of cows repeating are observed. If a BBSE is done at this juncture, the bull will almost certainly fail. However, this bull will recover over a period of weeks to months, but in a seasonal system as we predominantly have in Ireland and Britain, this is not good enough for the buyer.

Until we reach a stage where all bulls are tested pre-sale, and where there is consistency in terms of bio-security, vaccination programmes and nutrition between origin farms and destination farms, the above scenarios will continue to ensue. Regrettably this day is a long way off.

Safety

Working on bull fertility is rewarding work but it is also potentially dangerous work, because bulls are potentially dangerous animals. It should not be attempted where doubts exist about the quality of the facilities, or where there are doubts about the handler's ability to control the bulls. The safety of both the operators and the bull is always of paramount importance.

Veterinary Bull Fertility Examination By kind permission of Veterinary Ireland

Date of Examination:

Certificate No:

Owner:	
Address:	
Herd No:	

Bull:	Tag Number:
	Name:
	Breed:
	Date of Birth/Age:
	Other:

REASON FOR EXAMINATION: Pre -Sale Check ☐ Pre -Breeding Check ☐ Breeding Check ☐
 Examination for Insurance Purposes ☐ Other ☐ (Please Specify:)

KEY: * Place X in box to indicate Findings NAD = No Abnormality Detected ** Delete as Required

SECTION 1. PHYSICAL EXAMINATION

Body Condition Score: (1 - 5)		
Weighed: Y: <input type="checkbox"/> N: <input type="checkbox"/>	Weight: Kg.	
	*NAD	*Abnormal
Heart / Lungs:	<input type="checkbox"/>	<input type="checkbox"/>
Eyes:	<input type="checkbox"/>	<input type="checkbox"/>
Incisor / Dental Pad Alignment:	<input type="checkbox"/>	<input type="checkbox"/>
Musculoskeletal System:	<input type="checkbox"/>	<input type="checkbox"/>
External Genitalia:	<input type="checkbox"/>	<input type="checkbox"/>
Internal Accessory Glands:	<input type="checkbox"/>	<input type="checkbox"/>
Scrotal Circumference (SC):	cm	
Overall Result:	Satisfactory** / Unsatisfactory**	

SECTION 2. SEMEN EXAMINATION

Collection method:	EEJ <input type="checkbox"/>	AV <input type="checkbox"/>	MASSAGE <input type="checkbox"/>
Appearance / Density:	Creamy <input type="checkbox"/>	Milky <input type="checkbox"/>	Watery <input type="checkbox"/>
Gross Motility:	/ 5		
Progressive Motility	%		
Morphologically Normal Sperm:	%		
Overall Result:	Satisfactory** / Unsatisfactory**		

SECTION 3. ASSESSMENT OF MATING ABILITY

This Bull has **/ has not** been observed exhibiting normal service behaviour and mating ability.

SECTION 4. CLASSIFICATION

In my opinion, in terms of the standards detailed on the reverse of this Certificate, the examination findings would indicate that on this specific date the bull identified above is:

SUITABLE for BREEDING** (based on meeting the requirements of Sections 1 AND 2 ONLY) - mating ability has NOT been assessed

SUITABLE for BREEDING** (based on meeting the requirements of Sections 1, 2 AND 3)

NOT SUITABLE FOR BREEDING**

NB: This Certificate does not include any testing for infectious/contagious diseases, the results of which should be reported separately.

COMMENTS:

Veterinary Practitioner:

Name: Reg No: /

Signed: Date:

Practice Stamp / Address

Guidelines

The aim of this Certificate is not to guarantee bull fertility, but to reduce the risk of potentially unsuitable bulls being used for breeding. A bull meeting the requirements of Section 1 AND Section 2 should have no obvious physical abnormality that would render it unsuitable for natural service, and should have the potential to be fully fertile, based upon its semen quality.

NB: This Certificate does NOT include any assessment of health status. Any tests carried out for infectious diseases (e.g. BVDV) should be reported separately.

Certificate Number

Each Certificate shall have a unique number (e.g. vet-code, date, examination number on that date = 6100/20150216/03).

Section 1: PHYSICAL EXAMINATION

To meet the requirements of this Section the bull will have to demonstrate freedom from significant physical defects that could affect its fertility, or its ability to mate, and freedom from heritable defects that could affect its progeny.

Body Condition / Clinical Examination

Body Condition Scoring of bulls should be recorded using the Edmonson (1-5) scale.

Weighed / Weight

Where the owner has scales available, and where the bull has been weighed, this should be noted and weight recorded in Kg.

Heart / Lungs

The bull should be examined for any signs of heart or lung defects, which may interfere with fertility or ability to stay with cows or achieve intromission.

Jaws / Eyes

The bull should be inspected for severe over/undershot jaw and gross ocular defects such as cataracts, carcinomas, etc., which may interfere with vision and the ability to seek out females.

Musculoskeletal Defects

The bull should be inspected for evidence of lameness whilst walking on a smooth, level surface. Lamé bulls will fail Section 1, and will be classified as "Unsatisfactory". Bulls with severe conformational defects of the limbs, e.g. post-hock, sickle-hock, valgus deformity or serious foot defects such as corkscrew claw or inter-digital fibroma, should be classified as "Unsatisfactory" in Section 1.

External Genitalia

The scrotum and contents should be carefully palpated and scrotal circumference measured (see below). Bulls with gross physical abnormalities, such as epididymitis or orchitis should be classified as "Unsatisfactory". Slight variation in size and position of testicles is acceptable, though breed standards may vary, and bulls may be rejected at pre-sale society inspections, if any variation in size or shape is present.

The sheath / penis should be palpated for swellings, adhesions, discharges, papillomata, etc. The tip of the penis should be inspected for normality and if it has not been visualised during semen collection, this should be noted in the "Comments".

Scrotal Circumference (SC)

Breed Society standards, where published, should be used as minimum SC standards in order to meet the requirements of this part of the examination. SC standards given below (Society of Theriogenology) should be utilised when no alternative Breed Standards are available.

Age In Months	12-15	>15 ≤18	>18 ≤21	>21 ≤24	>24
Min. SC	30cm	31cm	31cm	33cm	34cm

Internal Accessory Sex Glands

Seminal vesicles, prostate and ampulla should be palpated per rectum to check for any abnormalities.

Section 2: SEMEN EVALUATION

Semen samples must meet a minimum set of standards, as detailed below.

Gross Motility

Scored on a 1 to 5 scale. A good semen sample would normally score at least 3. However, as gross motility is influenced by the concentration of the sample, the assessment of progressive motility is required for all bulls.

Scale	Description
1	No swirl; generalised oscillation of individual sperm only.
2	Very slow distinct swirl.
3	Slow distinct swirl.
4	Moderate fast distinct swirl; dark waves.
5	Fast, distinct swirl with continuous dark waves.

Individual Progressive Motility

The minimum requirement is a progressive motility of at least 60%. Bulls with gross motility scores of 3 to 5 would normally be judged "Satisfactory" for progressive motility if semen is handled well and examined on a heated stage. Bulls that score <60% for progressive motility should have a second ejaculate collected immediately to rule out sperm accumulation and senescence as a potential cause. Continued failure to achieve ≥60% progressive motility will normally be caused by a high percentage of morphologically abnormal or dead sperm which will be confirmed at the next stage of examination.

Semen Morphology

100 sperm cells should be counted using X1000 oil immersion microscopy with nigrosine / eosin smears or wet preparation phase contrast. To meet the requirements of this section, 70% or more of sperm should be morphologically normal, with no more than 20% of sperm showing nuclear defects. In marginal cases (65%-70%) at least 2 counts of 100 sperm cells should be carried out.

Bulls with no apparent physical abnormality of genitalia, but with marginally poor morphology should be classed as "Unsatisfactory". However, a note can be added to the comments section suggesting re-examination after 60 days when recovery may be evident if temporary degeneration has been the cause. Young bulls aged 12-15 months may have a poor morphology count due to immaturity and can be reassessed after 2-3 months.

Section 3: ASSESSMENT OF MATING ABILITY

As libido is difficult to assess and define, this part of the examination simply confirms whether or not the Veterinary Practitioner has observed normal service behaviour and intromission when the bull was presented with a female in oestrus. At least one successful service within 10 minutes of being presented to an in-oestrus female should be expected. If this part of the examination is not carried out, then bulls can still be classified as "SUITABLE FOR BREEDING" based on meeting the requirements of Section 1 AND Section 2 only. The onus is on the owner / purchaser to observe the bull closely at the start of the breeding period to monitor libido and mating ability.

Section 4: CLASSIFICATION

Any bull classified as "SUITABLE FOR BREEDING" must meet the requirements of the physical examination and achieve minimum standards for Scrotal Circumference (SC), Progressive Motility (60%) and Sperm Morphology (70%).

[illegible]

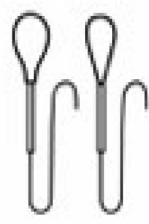
Notes

[illegible]

Bovine Sperm Morphology



normal sperm



pyriform heads



tapered heads



microcephalic & macrocephalic sperm



vacuoles (craters)



clumped chromatin



detached heads



decapitated defect



rolled-crested-giant head syndrome



teratoids



knobbed acrosome



ruffled membranes & detached acrosomes



distal midpiece reflex and coiled tail



mitochondrial sheath defects and fractures



abaxial, accessory & multiple tails



dag defect



stump tail defect



corkscrew defect



pseudodroplet defect



coiled principal piece



bowed midpiece



hypotonic or cold shock



proximal & distal droplets



white blood cells, medusa cells, spheroids

