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PIC BOAR STUD MANAGEMENT MANUAL

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WELCOME TO THE 2015 EDITION OF THE PIC BOAR STUD MANAGEMENT MANUAL

We're pleased to present the 2015 edition of the PIC Boar Stud Management Manual. This updated content reflects the emerging challenges you face and the advances we are bringing to this ever-changing landscape.

Now more than ever, your business demands new technologies that not only facilitate the production of top quality, highly consistent semen, but also ensure the health and well-being of the boars in your care. At PIC, we continue to improve boar breeding in ways that help you meet challenging production targets and safeguard the food supply. Count on us as your partner to continually seek out and deliver new knowledge that keeps you globally competitive.

This manual is designed to not only educate your new employees, but also challenge more experienced personnel to reevaluate barn and lab processes. Topics include:

- Sperm Production and Collection
- Barn Management
- Laboratory Management
- Feeding & Nutrition
- Welfare & Health
- Key Performance Indicators
- Boar Life & Replacement Rate

We hope you find this manual helpful. If you have any questions, please contact your PIC representative.

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PART 1:

GENERAL ANATOMY AND PHYSIOLOGY OF THE BOAR'S REPRODUCTIVE SYSTEM

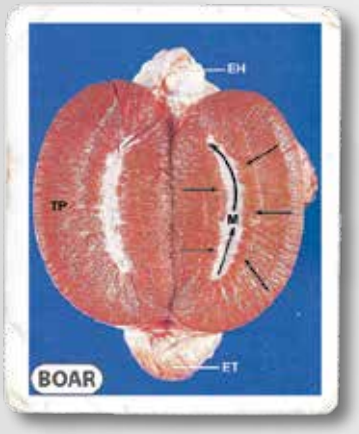
SPERM PRODUCTION

The process of boar sperm cell and testicular development starts in utero. Once born, reproductive behavior may begin as early as 1 month of age. The boar will experience increased semen production at 6 months (see Table 1).

TABLE 1. PROCESS OF BOAR MATURATION

AGE	MATURATION PROCESS
Fetus (d 20–40 gestation)	Germ cell division and differentiation
Fetus (d 60 gestation)	Testicular dissension from the abdomen into the scrotum
1–2 months	Mounting behavior displayed
3 months	2nd germ cell division and increase in testes to body weight ratio
4 months	Sperm appear in seminiferous tubules and erections can occur
5½ months	Puberty begins and sperm appear in ejaculate
6–18 months	Testes size, semen concentration and ejaculate volume increases

PICTURE 1. FLOW OF SPERM PRODUCTION

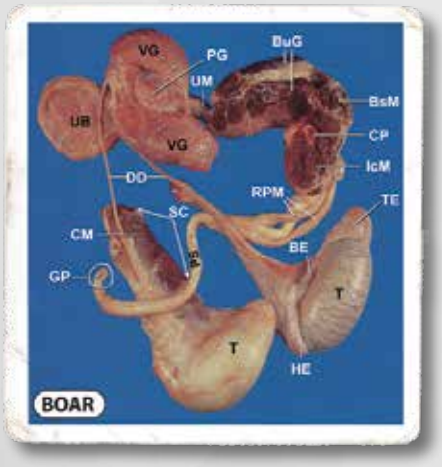


The testes are responsible for the production of sperm and testosterone.

Sperm production, or spermatogenesis, is a highly complex process that occurs within specialized compartments in the testes called seminiferous tubules. As sperm are produced, they move toward the center of the testis into the mediastinum (the white area shown as "M" in Picture 1) and then continue on to the head of the epididymis (noted as "EH" in the picture). A sexually mature boar is capable of producing 16 billion sperm cells per day from both testes (Senger, 2005). The spermatogenesis process takes 39 days on average. At any given time, there are sperm cells at different stages of development, allowing for continual sperm production.

The epididymis is comprised of three sections including the head, body and tail. Each section plays a part in sperm storage, sustenance and the completion of maturation over a period of 9-14 days. Sperm cells must travel through the epididymis to have a chance at fertilization. Without the epididymis, reproduction in the boar would not be possible.

PICTURE 2. REPRODUCTIVE ORGANS IN THE BOAR



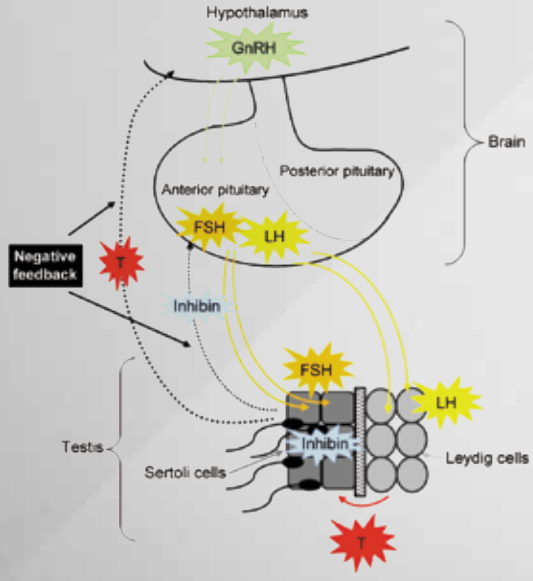
The accessory sex glands include the prostate gland, vesicular glands and bulbourethral glands. Together, these glands add seminal plasma to the sperm cells.

The prostate gland removes urine and bacteria from the reproductive tract before sperm enter the urethra.

The vesicular glands produce secretions that are viscous and milky in appearance, and comprise the majority of ejaculate volume.

Lastly, the bulbourethral glands produce the gel fraction of the ejaculate. These glands are large and dense in the boar (Knox, 2003; Senger, 2005).

FIGURE 1. SCHEMATIC OF HORMONE PRODUCTION AND REGULATION IN SPERM PRODUCTION



The penis is divided into three sections: the base, shaft and glans penis. The glans penis contains sensory nerves and sets off the process of ejaculation. The shape of the glans penis resembles a corkscrew, which is unique to the boar (Senger, 2005).

THE ROLE OF HORMONES

The brain produces the hormones GnRH (gonadotropin releasing hormone), LH (luteinizing hormone) and FSH (follicle stimulating hormone). The three work together to promote and regulate testosterone (T) production and ultimately sperm cell development and male behavior (Figure 1). These processes are essential for reproduction.

PART 2:

THE ISOLATION AND ACCLIMATION PERIOD

Typically at around 6 months of age, the boar is put into isolation for a period of 4 to 8 weeks. This allows critical time to test for diseases and establish vaccination protocols. Depending on the number of vaccinations required, these shots/injections should be spaced at reasonable intervals throughout the isolation period to minimize stress on the boar.

The isolation facility and location depend on the regional pig density. Normally, a distance of 1.5-2 miles (2.4-3.2 km) from the stud is preferable, but in a pig-dense area it is better to have the facility closer to the main stud — and if possible, attached by a covered walkway. The doorway into the stud should be locked during the isolation period until the boars are released from testing.

With the development of barn filtration, steps can be taken to reduce the possibility of spreading a disease to the main stud. If the main stud is filtered to prevent entry of disease, the isolation facility should be filtered as well. If the main stud is not filtered due to location in an area of low pig density, the isolation exhaust air can be filtered to prevent possible infection of the stud. The exhaust filters can be opened up after testing, indicating the group is negative for the diseases of concern.

Isolation Best Practices:

- Do not locate the isolation facility too close the main stud (less than 400 meters) if it does not have exhaust filters and the main stud is not filtered.
- Biosecurity of the isolation facility should be maintained by requiring all staff and service personnel to shower-in and shower-out in a separate shower facility.
- Service personnel entering the isolation facility should follow the same restrictions used at the stud and adhere to the same downtime rules. Do not forget to clean and disinfect required tools and material they take in.
- Production staff may visit and work in the isolation unit after working in the stud, but must observe one night of downtime prior to returning to the stud.
- Boars should be housed individually and not mixed during transfer to the main stud.
- All boars in isolation should be clinically monitored each day.
- Perform an initial statistical test on the boars within seven days of arrival, and then 100% testing of the population at the end of the isolation period.
- For a facility that is located away from the main barn, be sure to wash, disinfect and dry the trailer prior to moving the boars to the stud.
- Prior to release of isolation animals to the stud, communicate with your herd veterinarian or PIC Health Assurance to verify the current health status of the source herd.
- Move the boars into the stud as soon as possible after receiving negative test results.
- Managers may choose to train boars in isolation or in the stud. Either option will work as long as the proper training protocol is in place (see Part 8: Training).
- Record daily high and low ambient temperatures in the barn. High environmental temperatures have a negative impact on semen quality. With the records it could be possible to explain some drops in semen quality.

MONITORING AND TESTING

You should record data on any animals exhibiting clinical signs or undergoing treatment. Any boars that are off feed or clinically ill should have their temperature recorded and be monitored and managed on an individual basis. Increasing incidence of off-feed or feverish boars from one day to the next is indicative that disease has been introduced. It is recommended that the boar stud manager notify their veterinarian if clinical disease and/or deaths occur.

How to test whether boars can be released into the stud:

- Test a statistical sample of the boars within 7 days of arrival
- Test 100% of the isolated boars serologically for PRRS at the end of the quarantine period prior to release of animals to the stud. Use both ELISA (individual) and PCR (pooled by five) methods. Ensure the diagnostic laboratory runs the PCR prior to the ELISA to avoid contamination.
- The population must be determined to be negative by the veterinarian prior to entry into the main stud.
- See Part 10: Welfare and Health for additional details on testing in isolation.

Note: Specific disease testing requirements will vary by country and region/state. Consult your veterinarian.

Boars should be isolated upon delivery from the source farm per the PIC sales agreement (Conditions of Sale). PIC will inform the stud of any significant change in the health status of the PIC source herd. The stud or its veterinarian will be provided with the results of blood tests performed on the source farm for the boars that are destined to be placed into the quarantine facility on request. Do not move boars from the isolation into the main stud if PIC notifies you of a health concern in the source herd or if the isolation facility is experiencing a clinical outbreak of any disease.

PART 3:

DAILY CARE AND MANAGEMENT

When transferred to the barn, boars should be placed by line and then by age. Keep young boars grouped together, and avoid mixing them in with older boars.

Barns should be walked daily by the manager or an assigned technician. Some key things to note:

- Look for boars that did not clean up their feed;
- Get boars up every day at feeding to observe for lameness;
- Also, watch for any coughing or signs of respiratory problems.

Consult with your veterinarian for suggested treatment protocols.

Records should be kept for any boars that are treated or off-feed.

PART 4:

BARN MANAGEMENT

Note: These recommendations pertain to both the isolation facility and the main stud.

Boars are typically quiet animals; however, it's imperative that stud staff take every precaution to prevent unnecessary risks when training, sampling, treating, walking and collecting the animals. When taking boars to and from the collection area, walk behind the boars, and use a sorting board. (See Part 10: Welfare and Health for more information.) Especially on busy collection days a system should be in place to organize the boar movement and avoid clashes.

CLIMATE CONTROL

The optimal barn temperature for sperm production is 61-64°F (16-18°C). Hot temperatures can affect semen quality negative up to 8 weeks after exposure. Ejaculate trash rates can rise up to 20% while hot periods.

Misters, atomizers, evaporative cooling and air conditioning are used to control barn temperatures — but care must be taken to avoid creating a wet environment. Each barn should feature adequate ventilation and air movement to reduce ammonia and odor levels, while maintaining acceptable ambient temperatures for both the boars and barn personnel (See Part 5: Ventilation and Air Flow for more information). Crate backing should consist of open bars, not solid panels, to allow for adequate air movement and optimum temperature around the testicles.

Boars consume 1.5-2 gallons (5.6-7.8 L) per day on average, so maintain a minimum water flow rate of about 1 quart per minute. It's imperative to measure your flow rate once per quarter — increasing to once a week during the summer months — to ensure the boars have sufficient access to water. Failure to maintain adequate water flow puts the animals at risk of tissue water depletion and dehydration. Make sure that every water nipple is working. Chemical testing of the water should also be performed to check impurity, mineral and bacteria levels twice per year. Your local municipality may require more frequent testing.

Check to ensure that there is NO stray voltage flowing through water lines and equipment.

When entering and exiting the isolation facility, each animal should be walked through a foot bath containing a copper sulfate solution. In the stud, boars should be walked through the foot bath on the wall back to the stall after collection. This will help harden the hooves and prevent lameness problems in the stud.

Mats should be placed under boars suffering leg or hoof problems to ensure comfort and promote recovery. Have enough mats on hand for 10% of stud capacity.

Each day after collections are finished, the collection area should be power washed with hot water and high pressure. Take special care to clean the warm-up area, collection pens or crates, dummy (Particularly the underside) and mats. After washing, the area should be clean of organic material (i.e. manure, semen). Once a week, after washing the collection area, disinfect it with a product made specifically for animal facilities.

Be sure to include all surfaces (i.e walls, bars on the crates).

Many different management factors in the barn can impact semen quality (Table 2).

A plan for pest control (in terms of rodents and insects) should be in place to avoid intake of pathogens. Contact a pest control professional to provide strategies for your facility. Make sure that boars have no access to rodenticides.

Ensure feed deliveries have good biosecurity practice. Transport trucks should be cleaned before supplying your facility.

Keep samples from every feed batch for 8 weeks after the end of feeding. If there is a drop in semen quality you can use them for further investigation (i.e. test on mycotoxins).

TABLE 2. MANAGEMENT FACTORS THAT IMPACT SEMEN QUALITY

SITUATION	DESCRIPTION	EFFECT ON BOARS	REFERENCE	RECOVERY
High ambient temperatures	>85°F (29°C) for 3 days or more	Sharp increase in abnormal sperm per ejaculate	McNitt and First, 1970 Wetteman et al., 1976	Two months
Moderate ambient temperatures plus high humidity	79-85°F (26-29°C) + 75% humidity or greater for 4 wks or more	Gradual increase in abnormal sperm per ejaculate	Suriyasomboon, 2005	
Fever (caused by vaccination or disease)	Body temperature >103°F (39°C) for 2 or more days	Sharp increase in abnormal sperm per ejaculate	McNitt and First, 1970	Two months
Increased and erratic collection regimens	>3 times per week	Gradual decrease in number of normal sperm per ejaculate	Kennedy and Wilkins, 1984	Rest 2 weeks
Reduced nutrient intake	>15% reduction in energy or protein intake for more than 8 weeks	Reduced libido and gradual decrease in normal sperm per ejaculate	Louis et al.,1994 a, b	Variable. It depends on severity of the restriction
Suboptimal photoperiods	>16 hours of light or <8 hours of dark	Gradual decrease in libido and no consistent changes in sperm output	Sancho, 2004	NA
Immature boars	<6 to 7 months depending on genotype	Low volume of semen; low numbers of normal spermatozoa and presence of cytoplasmic droplets	Kennedy and Wilkins, 1984	Time needed to achieve maturity

PART 5:

VENTILATION AND AIR FLOW

It's critical to maintain the right environmental conditions for boars, for several reasons:

- Optimized sperm cell and quality semen production;
- Regulation of daily maintenance feed requirements;
- Control of bacterial growth within the environment; and
- Promoting health and minimizing lameness.

The goal of a ventilation program is to achieve desired room temperature (DRT) and humidity to create comfort.

DRT refers to the optimal temperature for boar comfort within a given environment. Adjustments must be made to DRTs to account for different environments, such as flooring and building type.

- Different DRTs have an associated set point (the point at which variable stage fans increase speed) considering variable environments (flooring, building type, etc.) in order to achieve maximum boar comfort.

Table 3 shows recommended conditions for various scenarios; excerpt from PIC's Ventilation Modeling Tool available upon request).

TABLE 3. BUILDING ENVIRONMENT VARIABLES AND RECOMMENDED OPTIMIZED CONDITIONS

	EXAMPLE 1	EXAMPLE 2	EXAMPLE 3	EXAMPLE 4
Flooring type	Slats	Slats	Solid	Solid
Barn type	Solid Sided	Curtain	Solid Sided	Curtain
Desired room temperature	66	68	63	65
Winter set point	69	71	66	68
Summer set point	65	67	62	64

TECHNICAL SPECS FOR CLIMATE CONTROL

Relative humidity in a boar stud should be between 40-65%. Humidity and DRT are controlled by managing and manipulating inside and outside air exchange rates measured by cubic feet per minute (CFM).

- During normal respiration, the boar produces both heat and water vapor, which elevates barn temperature and humidity unless the vapor is properly exhausted.
- To maintain humidity and temperature, air exchange should be, at a minimum, 14 CFM.
- When humidity and temperature are outside of the optimal range, the CFM rate needs to change to properly exhaust the excess heat and replace with cooler, dryer air. (see below)
  - Cooler air holds less water vapor, reducing the barn's relative humidity.
  - When outside temperatures are above desired room temperatures, increasing ventilation rates will not improve humidity.

Air speed, measured in feet per minute (FPM), is an important measure for effectively mixing cooler air sourced from inlets to eliminate drafts and areas of condensation.

- An air speed of 800 FPM is optimal for elevated fan stages, while 400 FPM is much more practical in minimum ventilation stages.
- Routinely evaluate air speed from inlets to assure proper mixing of air within the barn.

Supplemental heaters are required to control lower temperatures, and are essential in emergency situations.

- Set the heater(s) to turn on at a minimum of 2 degrees below the set point for increasing variable fan speeds. For example, if the heater set point is 70°F (21°C), then heaters turn on at 67°F (18°C).
- If the heaters are set too close to the temperature set point, excessive amounts of liquid propane or natural gas will be used.

Fan staging is designed to keep the building as close to the DRT as possible without causing major temperature variations. Fan staging removes heat and humidity as the barn warms through increasing CFM. Fan speed does not equal CFM — i.e., 50% fan speed does not equal 50% CFM. Therefore it's important to understand the relationship between variable fan performance and fan exhaustion rate (see Figure 2).

FIGURE 2

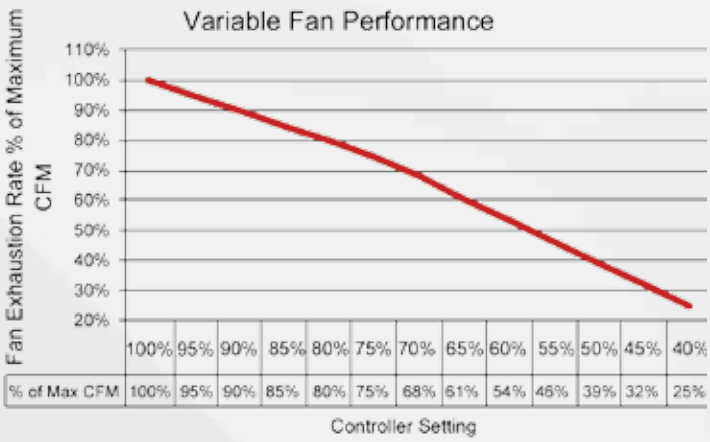


TABLE 4. ESTIMATIONS OF CFM BY FAN SIZE

FAN SIZE - INCHES	CFM OUTPUT	CFM OUTPUT WITH CONE
8	450	500
10	1100	1200
12	1500	1600
18	3500	3600
24	5700	6000
36	9700	10000
48	17000	18000
50	22000	23000
55	23000	24000

Motor curves correspond to different fan sizes and are defined as the relationship between the voltages supplied to the motor and the resulting RPM.

Incorrect matching of motor curve and fan size may either burn fans up or cause inaccurate fan speeds: i.e., a 60% fan speed setting results in 90% fan speed.

Instances in which air exchange rates increase — including rising outside temperatures, a change in season, and increased heat production due to boar activity — moderate changes in ventilation should be made. Avoid increases of 2x more CFM.

Providing an optimal environment for boars requires multiple aspects to operate together. As the total number of CFM increases the following must be considered.

- Each in^2 of ceiling inlet provides 4.5 CFM.
- Each in^2 of eave inlet provides 2.5 CFM.

If the system doesn't have the proper number of inlets open and the proper amount of attic inlet, the air will not effectively flow into the barn, no matter how many fans are running.

Water — in the form of dripper systems or evaporative cooling — can be used for cooling the animals. However, the production of extra water in the air and on the floor creates risks such as elevated barn humidity, lameness and bacterial growth. So, when these cooling methods are used, minimum ventilation rates must be raised to effectively dry the floor faster than normal rates.

- The purpose of dripper systems is to cool the testes to optimize the temperature for sperm production.
- Evaporative cooling, combined with air speed, effectively cools the barn but also adds humidity to the air.
  - The addition of evaporative cooling is most effective when inside humidity is less than 70% or outside temperature is lower than the inside temperature.
- A soaking cycle should be used that allows the pads to partially dry between applications of water.

Allow pads to completely dry at least once per day.

- Use evaporative pads at only 10 degrees above DRT.
- Routinely replace the water in the reservoir, as the evaporative process causes a concentration of salts and minerals that potentially decreases the equipment's useable life.

TROUBLESHOOTING

Several factors should be considered when troubleshooting ventilation or air quality issues.

- Fan output can be influenced by the following:
  - Dirty louvers and blades may decrease fan efficiency as much as 30%.
  - Leaking pit pump covers drastically affect air exhaustion.
  - Adding fan cones improves the fan's output by 10-20% CFM.
  - Excessive static pressure (>1000 FPM air speed or 0.1 in of water) severely affect a fan's exhaustive CFM rating.
- Wet floors are a major factor in overall boar discomfort, and can make a boar feel 9 degrees cooler with the same air temperature. This can be fixed by:
  - Increasing minimum ventilation rates.
  - Assuring proper airspeed from inlets.
  - Increasing barn temperature until the floors are properly dried.
- A decrease in RPM and exhaustive output as a result of slipping fans can be detected by measuring the temperature of the pulley with an infrared thermometer. A thermometer reading of 7 degrees warmer than room temp indicates a slipping belt.
- Make sure that ventilation does not cause high air flow from the boar housing to the collection area, in order to avoid contamination of the ejaculate during collection. Additionally, high air flow makes collection personnel and boars feel uncomfortable.

PART 6:

BODY CONDITION

Body condition affects libido, semen production and the animal's ability to jump on the collection dummy. The target body condition for 90% of boars in stud is "normal." At normal body condition it is possible to feel the backbone with firm palm pressure but not to see it (especially near the tail). (See Pictures 3 through 5 to compare body types.)

PICTURE 3.
THIN BODY CONDITION

PICTURE 4.
NORMAL BODY CONDITION

PICTURE 5.
FAT BODY CONDITION

PART 7:

FEEDING AND NUTRITION

Boars arriving into the isolation unit should be on full feed to help with their transition into the new facility. This feeding rate should be maintained for 2 weeks and then dropped to 5 lbs. per day for the remaining isolation period.

Boars in normal body condition (5-6 lbs./2.3-2.7 kgs) should be fed once per day. Adjust appropriately to meet the target body condition (see Part 6: Body Condition). A fat boar should be restricted to 3.5-4 lbs./1.6-1.8 kgs. per day, while a thin boar should receive 7-8 lbs./.2-3.6 kgs. per day.

If drop feeders are used, weigh samples on a quarterly basis to ensure accuracy. Be sure to take into account changes to ingredients and bulk density.

Feeders should be adjusted every 2 weeks to maintain proper body condition and semen output. Proper maintenance of body condition will aid in libido and working ability of boars. In addition, proper feed intake levels and nutrient fortification should be provided to optimize semen production (refer to Appendices A and B).

INGREDIENTS

Mycotoxins in boar feed can have several detrimental consequences on the animal's performance, including problems in maintaining high quality semen (see Table 5). Avoid the use of by-products or co-products where mycotoxins may be concentrated. Select high quality ingredients and monitor mycotoxin levels on a regular basis. Work with your nutritionist to add a binder to the boar diet.

TABLE 5. IMPACT OF MYCOTOXINS IN FEED

MYCOTOXIN	EFFECT
Zearalenone	Delayed puberty Reduced testes size Diminished libido Poor sperm quality
Aflatoxin	Edema of the prepuce – loss of hair Poor semen quality Low sperm concentration Increased morphological abnormalities Reduced fertilization capacity
Ochratoxin	Off-feed Gastric ulcers Poor sperm quality
Trichothecenes (T2, DON, DAS)	Off-feed vomiting

(P. Matzat, summarized from various sources)

PART 8:

TRAINING PROCESSES

TRAINING FOR MANUAL COLLECTION

With respect to different lines and individuals, most boars are ready to be trained at 160 days of age or above. The process should start no sooner than that timeframe.

Identify farm staff that are willing to devote time and patience to train young boars, and begin training 3 to 5 days after arrival. You'll need to have a recording system is in place to track the progress of each boar. Training should take no more than 4 weeks for each individual animal.

Before training begins, adjust the height of the dummy to match the size of the young boars being trained. The collection area should be draft-free and have flooring with good traction.

The training protocol is as follows:

- Remove any source of distraction in the collection area.
- Ensure personnel safety. Make sure the boar is comfortable with human contact.
- Squeeze the preputial diverticulum to stimulate the boar, and make every effort to get the boar to pay attention to the dummy.
- Once the boar jumps the dummy, lock the penis and collect the ejaculate.
- Observe any possible anatomical problems with the boar (i.e., limp penis, persistent frenulum) during this process.
- While personnel are collecting from the first boar, the next boar should be placed in the warm-up area to prepare for training.
- If a boar does not show interest in jumping the dummy within 10 minutes, move him to the warm-up area and administer a natural prostaglandin. Wait 5 or 10 minutes, then return him into the collection area and retry collection.
- Once the boar is trained, repeat the process for 3 days in a row to reinforce the learning experience.
- After boars complete the training, their semen must be collected once per week.

TRAINING USING AN AUTOMATIC COLLECTION SYSTEM

An automatic collection system includes an artificial cervix (AC), slide arm, AC holder and dummy. The AC mimics a sow's cervix and provides pressure to stimulate the boar. The slide arm allows free back and forth movement during collection.

- Follow the manual collection steps (outlined above) for the first day of collection.
- On day 2, collect the first portion of the ejaculate manually for approximately 1 minute with the left hand.
- After 1 minute, attach the penis to the automatic collection system and allow the boar to finish the collection.
- Repeat the process on day 3 of training.
- Each boar will acclimate to the system in their own time. Not every boar accepts automatic collection. If he does not acclimate to the system after 4 weeks you should consider hand collecting him.
- Avoid any type of manipulation (vaccination, cutting teeth, etc.) in the collection area.
- Always collect boars until they have finished ejaculation (gel fraction of ejaculate).

See Appendix C for instructions with photos.

PART 9:

BOAR SEMEN COLLECTION

The boar should always be brought into the warm-up area first to be properly prepared for collection. Here, the boar sheath will be cleaned and the preputial diverticulum emptied of its contents. The hair around the sheath should be trimmed periodically.

Hygiene must be maintained during collection to limit bacterial contamination. Preferably, staff should double-up on gloves.

Prepare your collection vessels one day in advance, and store them in a clean, sealed, hygienic, warmed (98°F/37°C) area until use. Collection vessels should only be prepared in a clean and disinfected environment (like the lab).

Once mounted on the dummy, the boar will make attempts to unsheathe the penis. With a clean, double-gloved hand, the collector will catch and hold the glans penis (corkscrew) and follow the movement of the boar until he is locked.

Besides blood tests, it is important to make daily observations and perform post-mortem exams on any animals that die in isolation. Gross lesions and/or signs of illness such as coughing, diarrhea, and lethargy may warrant further tests.

The boar ejaculate has 4 fractions:

- Pre-sperm
- Sperm rich
- Post-sperm
- Gelatinous (boar plug)

Avoid collecting the pre-sperm fraction into the cup. This is typically a clear emission that contains urine and bacteria.

Hold the penis with the tip slightly elevated to avoid preputial fluid running down the penis into semen collection vessel.

Allow 1-2 cm of the penis to extend beyond the gloved hand. Alternatively, open the last finger to allow a free flow of semen.

The sperm rich fraction follows the pre-sperm fraction; start collection at this point, continuing until he completes the ejaculation. Typically this process takes 8-10 minutes, with some individual boars taking longer.

Collect semen into a clean disposable container, such as a polyethylene bag, Styrofoam cup, etc. All methods should be filtered to remove the gelatin material. Avoid placing the semen collection vessel on the floor, which could cause contamination. After collection, the filter should be removed from the bag in the barn and kept out of the lab. Do not squeeze the filter to collect the last drops of semen out of it.

Be sure to accurately and clearly document the boar ID, genetics and technician/collector name, and attach this information to the bag or cup that contains the ejaculate.

Transport semen to the lab as fast as possible. Try to maintain the semen temperature by isolation.

All boars, regardless of semen demand, should be collected on a regular basis. Table 6 provides a guideline for collection intervals for sire line boars. Understand that individual boars and/or lines may perform better at a different interval than suggested, assuming collections occur regularly. Generally, maternal lines should be collected once per week regardless of age.

TABLE 6. COLLECTION INTERVAL BY BOAR AGE FOR SIRE LINES

AGE	INTERVAL
<12 months	1 x per week
≥12 months	3 x every 2 weeks

PART 10:

BOAR WELFARE AND HEALTH

BODY TEMPERATURE AND APPETITE

Rectal temperatures should to be taken for boars that are off-feed or show clinical signs of illness. If the boar's temperature remains >104°F, skip collection for that day and notify the herd veterinarian immediately. Consider a diagnostic work-up including PRRSv PCR testing on serum or blood swabs. If the number of off-feed and/or feverish boars increases from one day to the next, the stud should be closed and a full diagnostic work-up initiated. (See more on Stud Closure, below.)

DIAGNOSTIC TESTING

Weekly PRRSv PCR testing of blood or serum should be conducted at a frequency and number to achieve a minimum 95% confidence level at 5% prevalence, based on sample type and the sensitivity/specificity of the PCR test.

More rigorous sampling is at the discretion of the stud. Pooling samples up to 5 per pool is permitted.

The blood swab technique has been used for the last several years and is an effective way to collect blood for PRRS PCR testing on a weekly basis. Another method using the tarsal vein on the back leg of the boar is a good technique for collecting blood for PRRS PCR and ELISA. Contact you herd veterinarian or PIC for instructions on blood collection. Samples for PCR should be collected and submitted according to diagnostic laboratory protocol.

Monthly PRRSv ELISA screening (30 individual samples) or weekly sampling of a similar number is recommended.

Consider immediate PRRSv PCR testing from blood samples of any boars with a fever, off-feed or showing other clinical signs.

These samples should be PCR tested individually, rather than as a member of a pool.

CRITERIA FOR STUD CLOSURE

The decision to suspend shipment of semen from a boar stud relies heavily on the professional judgment of the manager and herd veterinarian. Semen must not be collected for shipment from individual boars if there is any question of health status on collection day. Temperatures should be recorded on any boars suspected of having a health problem. Suspension of semen sales must be considered when clinical disease (i.e., cough, scours, off-feed) is evident or elevated temperatures (>104°F or 40°C) are present in more than 5% of the boars in the stud. If the number of off-feed and/or feverish boars is less than 5% but is increasing daily, the stud should be closed for a diagnostic work-up. Additional grounds for potential closure can be raised if the manager or herd veterinarian has other disease risk concerns (i.e., a biosecurity breach). Suspicion of clinical or sub-clinical disease must be reported to the herd veterinarian to determine whether distribution of semen can continue.

Confirm positive diagnostic results for diseases transmitted in semen such as PRRS.

HANDLING AND EUTHANASIA

Mature boars are large and powerful and may cause injury to caretakers during normal handling. Special care should be taken when moving, treating or taking samples from boars. If detailed examination or treatment is required, the boar should be safely, effectively and humanely restrained. For safety reasons, the boar's teeth should be cut on a regular basis. Use a wire saw and cut the fang to approximately ½ inch.

In the event the boar needs to be euthanized, refer to the document On-farm Euthanasia for Swine: Recommendations for the Producer (AASV and NPB, 2009) for the proper protocol.

TRANSPORT OF BOARS

Drivers employed to transport boars should be TQA® certified.

Stocking density should be based on weight, temperature and distance traveled (see Table 7).

TABLE 7. STOCKING DENSITIES FOR BOARS BASED ON WEIGHT, TEMP, AND DISTANCE

WEIGHT		FT² REQUIREMENTS PER BOAR			
LB	KG	<80°F (27°C)	80-90°F (27-32°C)	>90°F (32°C)	>90°F (32°C) AND >250 MILES
241-258	109-117	3.6	3.9	4.3	5.1
259-305	118-138	4.3	4.7	5.1	6.1
306-364	139-165	4.9	5.4	5.9	7.1
365-399	166-181	5.6	6.1	6.7	8
400-449	181-203	6.4	7	7.7	9.2
450-499	204-226	6.9	7.6	8.3	10
500	227	7.6	8.4	9.1	10.9

PART 11:

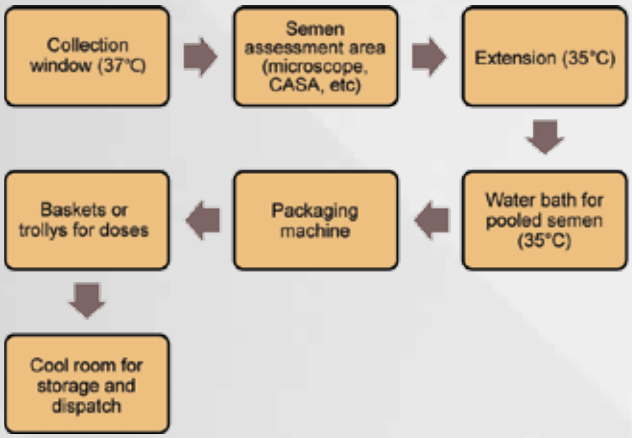
LABORATORY MANAGEMENT

Barn personnel SHOULD NOT be allowed in the laboratory at any time unless they have showered and changed into clean clothing. Lab personnel should wear different attire than what they wore in the barn. These items should be washed separately from barn clothes.

Additional best practices to follow:

- Lab personnel should wear lab coats and hairnets.
- Hand cleaning and disinfection prior to lab access should be obligatory.
- No eating or smoking in the lab.
- Clean countertops with a bleach solution as soon as production is finished for the day.
- The filling machines should be cleaned after every production day following the manufacturer instructions.
- Rinse hoses with deionized (DEI) water, soaked them in alcohol, and then re-rinse and hang to dry before the next production day.
- Avoid touching any surface that gets in contact with semen during processing (pipette tips, inner side of bags, etc.).
- The lab should be designed to promote efficiency in processing semen (Figure 3).

FIGURE 3. FLOW OF THE LABORATORY



WATER QUALITY

Purified water is the largest component of a dose of semen — therefore water quality and monitoring are paramount.

You may purchase purified water (the best option for fewer than 100 boars) or install a water purification system in the stud. The cost can be variable, depending upon the quality and the origin of the water source.

Daily monitoring of the water is required to ensure consistent quality. Many studs in the United States use a Myron 'L' 250 II device to monitor Megaohms (MΩ) and have this mounted in the lab for an immediate visual indicator.

TABLE 8. PARAMETERS FOR WATER GRADES AND SPECIFICATIONS

PARAMETER	TYPE I	TYPE II	TYPE III	TYPE IV
Electrical conductivity, max, µS/cm at 298 K (25°C)	0.056	1.0	0.25	5.0
Electrical resistivity, min, Ω-cm at 298 K (25°C)	18.0	1.0	4.0	0.2
pH at 298 K (25°C)	A	A	A	5.0 to 8.0
Total organic carbon (TOC), max, µg/L	50	50	200	No Limit
Sodium, max, µg/L	1	5	10	50
Chlorides, max, µg/L	1	5	10	50
Total silica, max, µg/L	3	3	500	No Limit

A = The measurement of pH in Type I, II, and III reagent waters has been eliminated from this specification because these grades of water do not contain constituents in sufficient quantity to significantly alter the pH.

Reagent water grades and specifications, microbiological contamination levels and water quality specifications should be on hand for in-lab use (Tables 8–10; ASTM, 1991). Pure water systems installed in boar studs are designed to produce water between Type I and Type III grades. If the water samples do not meet these specifications, an extensive analysis should be performed to correct the problem.

TABLE 9. TYPES OF MICROBIOLOGICAL CONTAMINATION

PARAMETER	TYPE A	TYPE B	TYPE C
Max heterotrophic bacteria count	10/1000 mL	10/100 mL	100/10 mL
Cfu[a]/mL	0.01	0.1	10
Endotoxin, EU[b]/ml	0.03	0.25	NA

[a]CFU = Colony forming units; [b]EU = Endotoxin units

TABLE 10. SPECIFICATIONS FOR IN-LAB WATER QUALITY

WATER QUALITY SPECIFICATIONS	
Bacteriology	<1 cfu[a]/mL
Purity inorganics	=18 MΩ[c] @77°F (25°C)
Organics	>0.001 AU@254 nm
TOC[d]	>50 ppb[b]
pH	6.8 to 7.2

[a]CFU = Colony forming units;
[b]PPB = Parts per billion;
[c]MΩ = Megaohm – cm (electrical resistance);
[d]TOC = Total organic carbon

SEMEN ARRIVAL

A stud that features a pass-through window from the barn into the lab should have a warming cabinet, set to 98°F (37°C), to pre-warm the collection cups prior to collection. The pass-through window should be designed as a sluice system. Only one of the two windows should be opened at once. To avoid the barn's contaminated air from flowing into the lab, it should be run on positive pressure. The pass-through chamber has to be cleaned and disinfected after every production day.

Likewise, if the stud uses a pneumatic tube delivery system, it too should have a warming cabinet close to the collection area. Avoid placing extender vats or semen processing close to the semen arrival.

Other best practices:

- The ejaculate of the boars must be clearly identified by boar ID and genetics.
- Never bring dirty containers into the pass-through window.
- The lab technicians should be immediately advised when an ejaculate enters the pass-through window.

The ejaculate should be properly evaluated and extended within 10 minutes of arrival.

SEMEN ASSESSMENT

When the semen arrives at the lab, first examine its color and odor to determine if blood and/or urine are present. In addition, an assessment of semen motility and morphology should be immediately performed to ensure it meets predetermined quality standards (see Table 11).

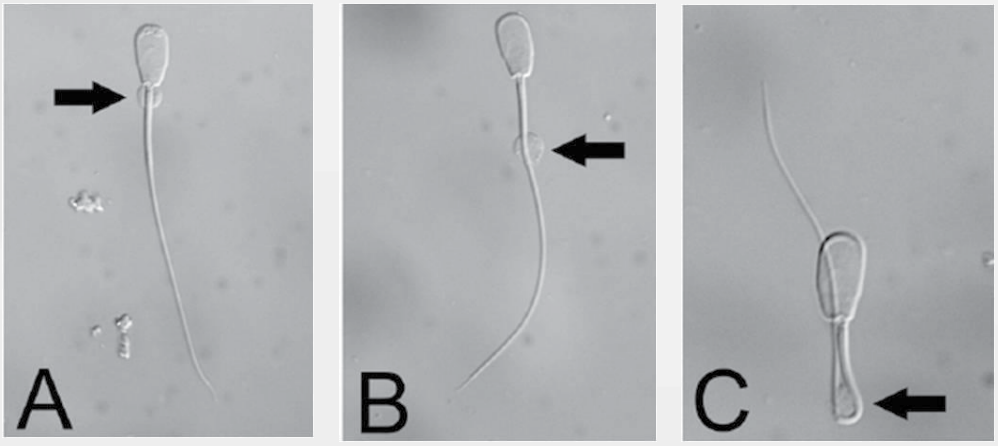
1) Measure the total weight of the ejaculate in grams by using a calibrated scale.

TABLE 11. CRITERIA FOR ACCEPTABLE SEMEN QUALITY

STANDARDS FOR SEMEN QUALITY	
Gross motility	≥80%
Normal sperm	≥70%
Cytoplasmic droplets, proximal and distal	<15%
Agglutination	<30%

2) Prepare a sample for evaluation by diluting the raw semen with extender or a sodium citrate solution in a 1:20 dilution. (If the ejaculate appears watery, use a dilution of 1:10; if it appears very creamy or concentrated, use a dilution of 1:40. This detail is important, as it can impact the accuracy of semen concentration.)

3) A microscope can be used to assess gross motility using a 98.6°F (37°C) warmed slide and cover or a computer-assisted semen analysis (CASA) system. Some CASA systems can also assess the morphology in the same sample used for motility. If CASA isn't used, prepare a killed sample and count 100 cells to get the % normal cells in an ejaculate.



A: Proximal cytoplasmatic droplet
B: Distal cytoplasmatic droplet
C: Distal midpiece reflex

Record any presence of cellular debris and sperm cell clumping or agglutination.

Ideally, less than 10% of ejaculates should be discarded due to semen quality. If this number is higher, a detailed analysis is in order to get to the root cause.

CONCENTRATION ASSESSMENT

There are various methods for measuring ejaculate concentration, including hemacytometers, photometers, spectrophotometers and CASA systems. These approaches rely on the proper mixing of raw semen and pipetting techniques to ensure that a representative diluted sample of the ejaculate is used for the analysis.

Depending on the equipment available, the measurement will be expressed as total sperm cells x millions per mL of raw semen (see Part 13 for training).

SEMEN EXTENSION

Semen extenders are available from different suppliers. Dependent on their ingredients, they provide nutrition, pH-stabilization and temperature endurance to the sperm cells and help to maintain viability for days.

Lab personnel need to know how many doses they are producing ahead of time so they can prepare sufficient extender for the regular collection day. A general rule of thumb is to multiply the target number of doses by the total volume per dose, and add 5-10% more to have enough extender for semen-dose production and ad-hoc uses like raw semen dilution, spills, pre-extension or last-minute orders. Prepared extender is only for same-day use. Do not store it over time.

Follow the extender manufacturer's instructions to the letter. It's critical to accurately weigh purified water and extender. Deviations from this can alter the osmolarity of the mix. The extender needs to be continuously mixed for one hour to allow the components to stabilize prior to adding it to semen. Make sure that there is no extender left in the bottom of the vat. To check if the water – extender ratio is correct, a refractometer (see Appendix H) can be used.

The temperature of the extender should be maintained at 95°F (35°C). At collection, semen has a temperature of 98-100°F (37-38°C) and there is a 2-3 degree temperature drop of the ejaculate during the evaluation process. Consequently, the extender needs to be kept at 95°F (35°C). If multiple step dilution is used, extender temperature can vary from this recommendation.

After the ejaculate and new extender are mixed, view a sample in the microscope prior to filling the doses. Make sure that the sperm cells are not negatively affected (motility, morphology).

Extenders may contain one or more antibiotics that can be modified and tailored to your situation.

It's important that you maintain open communication with your supplier.

Make sure that the extender powder is stored in a cool and dark place. Follow manufacturer guidelines.

DISPENSING SEMEN DOSES

After extension, semen should either be put into a water bath of 95°F/35°C for pooling, or immediately dispensed for doses.

Prior to dispensing, semen should be gently mixed since sperm may have settled. If high volume of pooled semen has to be processed, mixing while dispensing is recommended. The entire process from the time the semen arrives in the window to dispensing doses should take 20 minutes.

SEMEN COOLING AND PACKAGING OF COOLED DOSES

Cool rooms are used for storing and cooling the semen prior to dispatch. Temperatures here should be maintained at 59-63°F (15-17°C), with a stir fan used to ensure air circulation. Record daily high and low temperatures in the cool room.

Wire shelves or bakers trolleys are used to move, store and cool the semen. The design of these units provides optimal flow of cool air and a more uniform cooling of the semen doses. The doses need to drop from the extension temperature (95°F or 35°C) to the preservation temperature (59-63°F or 15-17°C). With the use of modern extenders, doses can be immediately moved to the cool room.

Semen should be cooled for four hours prior to dispatch. This is especially important for semen being shipped in a double-boxed Styrofoam™ cooler combination, as these coolers maintain a constant temperature in shipping. Semen for external shipping should be packaged and sealed inside a controlled (63°F or 17°C) environment. Temperature loggers can be used to monitor temperatures in transit, but the key to successful shipping is to fully cool the semen prior to packaging.

SEMEN SHIPPING AND TRANSPORT

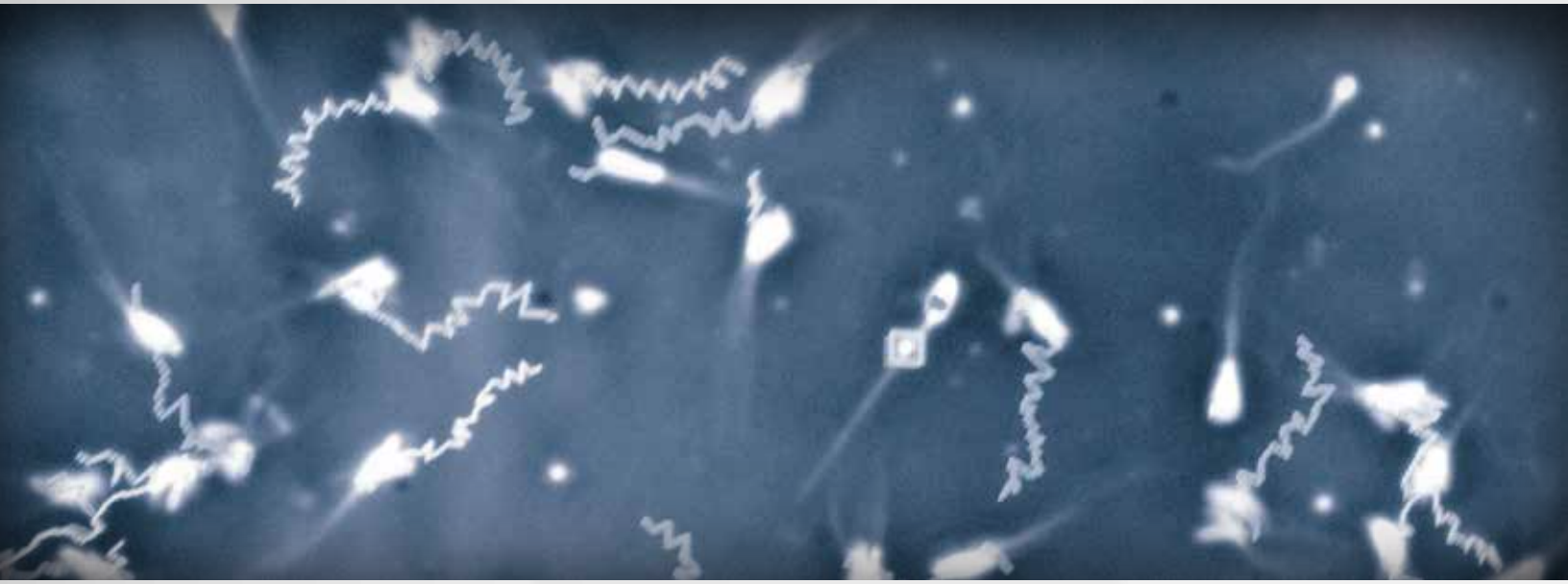
Semen shipped via an external courier, such as UPS or FedEx, should be packaged into a double-boxed Styrofoam™ cooler and be delivered Next Day Air™. Again, semen for external shipping should be packaged and sealed inside a controlled (63°F or 17°C) environment.

In the winter (<40°F or 4°C) use 2 warmed gel packs and in the summer (>80°F or 26°C) use one frozen (or two refrigerated) gel packs in between the coolers. For all shipments, one or two room-temperature gel packs should be placed inside the inner cooler or the single cooler. See Appendix D for further packaging instructions.

For semen that is sent via an internal courier, the temperature should be noted at the drop-off location.

PART 12:

LABORATORY QUALITY CONTROL



POST-PRODUCTION MOTILITY EVALUATIONS

Ensuring the quality of the dose of semen produced is of the utmost importance. The best indicator the stud has to assess the viability of the extended dose is to perform a post-production motility check on all batches and single sire collections.

The recommended process:

- Save a sample of each batch or single sire collection in a 5 ml glass tube as well as a sample in the tube or bag used for packaging. Samples should be prepared for evaluation according to the directions provided by the manufacturer of the extender (see Table 12).
- Perform post-production motility checks on Days 1, 3 and 5 at a minimum, where the day of collection is 0.
- If doses are <70%, perform the post-production motility check a second time to confirm results. If results are confirmed, a call should be made to customers who received the semen instructing them to discard it.

EXTENDER PREPARATION AND TRACEABILITY

- Post a printed reference guide in the extender preparation and extension area so the technician can quickly reference the pure water-to-extender ratios recommended by the manufacturer. A guide can be prepared for every type of extender available in the particular stud (see Appendix E).
- Create a recording system to keep track of the amount of extender used. This record should include extender type, name and manufacturer lot number and the result of first ejaculates motility check (refer to Table 13).

TABLE 12. MANUFACTURER GUIDELINES FOR SAMPLE PREPARATION FOR MOTILITY RECHECK

MANUFACTURER[a]	EXTENDER	SAMPLE	TEMP	TIME	EVALUATION
IMV	Gedil	1-5 ml cooled, extended	37˚C	10 minutes	Motility
Magapor	Vitasem	3 ml cooled, extended	37˚C	5 minutes	Motility
Minitube	Androhep Enduraguard	5 ml cooled, extended	38˚C	20 minutes	Motility

[a]For example purposes only. PIC does not endorse specific extender manufacturers.

TABLE 13. EXAMPLE EXTENDER RECORDING SYSTEM

EXTENDER: XXX		EXTENDER RATE: XX G/KG				
DATE	WATER, KG	EXTENDER, G	LOT #	INITIALS	MOT.	NOTES
11/1/2012	1 kg	50 g	1234			

DISPOSABLE MATERIALS RECORDS

- Document when suppliers change, and record any changes to the products that come into direct contact with semen. Include dates and lot numbers with each entry.
- Perform an in-lab trial to monitor for potentially detrimental effects, such as decreased motility.
- For example, when a new lot of tubes is received, dispense a semen sample from the new lot number into a tube and another sample with the current lot number using the same boar (or pool of boars) into a tube.
- Evaluate both tube samples on Days 1, 3 and 5 for motility and morphology.
- To measure the effects of consumables such as gloves, cut a piece from a new glove and a piece from a glove from the current lot number, immerse them into semen samples from the same pools, and evaluate the semen as stated above.
- Note: Different trials have shown that not all reprotoxic effects can be detected by such in house tests.

SUPPLIER QUALITY CONTROL SPECIFICATIONS

Request that your supplier detail all quality control regulations they have in place for consumable production. For example, the extender supplier should outline the protocol for the biological testing of plastic materials. Also ask suppliers about ISO 9000 certification.

EQUIPMENT CALIBRATION

Perform calibration on scales weekly; pipetting techniques and heatstage temperatures monthly; and infrared laser thermometers annually. You'll need the following for your lab:

- A set of master weights;
- A sensitive scale (readability to 0.001 g) for pipette volumes (for single channel air displacement pipetters); and
- An infrared laser thermometer calibration kit.

PICTURE 6.
WATER SOFTENER EXAMPLE

PICTURE 7. REVERSE OSMOSIS WATER METER

PICTURE 8. DEIONIZING TANKS FOR WATER SYSTEM

PURE WATER ANALYSIS

If your stud has a pure water system (Pictures 6-10), you must establish a verification process to ensure all components are operating properly. The frequency of water analysis is dependent on the starting quality of the water and the source (i.e., well, WEB water).

Carbon and sand filters should be used to capture gross particles. This equipment is functional for >500 K gallons of water. Check these filters every quarter.

Monitor the salt levels in the water softener (Picture 6) to ensure the proper ratio of water used per gallon of soft water produced.

The reverse osmosis (RO) machine needs to be serviced once or twice a year, replacing cartridges and filters. The use of RO meters and test strips can be used to locally monitor water produced by the pure water system (example in picture 7 is manufactured by Myron L Company).

DEI tanks must work in pairs and need to be replaced twice a year (Picture 8). If the system has indicator lights that change from green to red, this means you have a one-week window of time to replace the depleted tank. The pair of tanks operate in the "working and polishing" positions. The tank that is depleted is the one in the "working" position; when the replacement tank arrives, the existing good tank goes from the polishing to the working position and the new tank goes to the polishing position.

The 0.25 micron system and UV lamp should be changed every year (Pictures 9-10).

PICTURE 9. MICRON SYSTEM

PICTURE 10.
EXAMPLE OF A UV LAMP

Water lines from the UV lamp to the water outlets in the lab should be sanitized every month to control parafilm bacteria. Sanitize the water lines and faucet outlets in the lab using a laundry bleach solution according to the manufacturers guidelines. Soak the hoses overnight and then thoroughly rinse them out.

Review the whole system weekly for possible water leaks.

There are many quality control measures in the lab. It's helpful to post a checklist of measurements (see Table 14) prominently in the lab, with the name of the person responsible for each task.

TABLE 14. THE TABLE PROVIDES A CONCISE SUMMARY OF THE QUALITY CONTROL MEASURES AND THE FREQUENCY

QC MEASURE	FREQUENCY
Motility rechecks	Days 1, 3 and 5
Conductivity	Each extender batch
Disposable materials	New lot numbers or products
Scale calibration	Weekly
Pipette calibration	Monthly
Infrared thermometer calibration	Yearly
RO machine	Replace filter 2x per year
DEI tanks	Replace 2x per year
Water lines and faucets	Sanitize monthly
.25 micron system	Replace 1x per year
UV lamp	Replace 1x per year

THIRD PARTY ANALYSIS ASSESSMENT

To ensure the extended dose of semen meets minimum quality standards, a rigid monthly assessment of the diluted extender, water and extended semen doses needs to be performed.

Establish a program to periodically monitor the overall quality of the semen doses produced in the stud. This consists of sending a random set of extended semen doses for quality control checks including sperm cell concentration, gross motility, morphology and semen dose volume. At the same time, send pure water samples, diluted extender samples and extended semen samples for bacteriology. The protocol for water and extender sample preparation is provided in Appendix F of this manual. After the results of the third-party evaluation are delivered, compare them with your target concentrations.

- The number of semen samples sent should equal 1% of a day's production or a minimum of 10 doses randomly selected among batches.
- The frequency of submission is routinely scheduled monthly and samples randomly selected from all batches.
- Be sure to establish targets for every parameter measured, and the accepted variation ranges.
- When consumable source or lot numbers change, samples from the same batch should be sent using both sources/lots and the third party made aware of the change.
- The third party doing the semen evaluation service must be a truly independent entity. Consult with a PIC representative for recommendations.

PART 13:

PERSONNEL MANAGEMENT AND TRAINING

Each boar stud should maintain an employee to boar ratio of 1:60 — with 60% of the employees working in the barn and 40% in the lab.

- All employees should be PQA Plus® certified.
- Stud personnel that work directly with the boars should be trained on animal movement and handling as well as safety.
- Lab personnel should be trained by an employee with multiple years of experience or through a third-party training program. A induction plan covering all jobs (semen evaluation, extender mixing, machines, etc.) should be in place.
- Employee standardization should be conducted once per quarter, with intense cross referencing of lab employees in terms of slide prep, semen assessment, and other lab functions.

If manual semen evaluation is done, exact morphology (evaluation of 100 single cells in one ejaculate to define the percentage of normal cells in the ejaculate) from several (borderlined) ejaculates after production can help to improve the lab employee's skills.

PART 14:

KEY PERFORMANCE INDICATORS

As in sow farms, boar studs should track parameters that are indicative of boar performance and semen quality. Table 15 reflects key performance indicators studs should record and review on a weekly basis.

TABLE 15.

KPI	TARGET
Total sperm per ejaculate	>30 billion
Collections per boar per week	1.2
Untrainable boars	<3.0%
Prostaglandin use	<1.5%
Trashed collections	<6%
Unused doses	<5%
Boar morality[a]	<5%
Boars not in production (lame, ill, etc.)	<5%

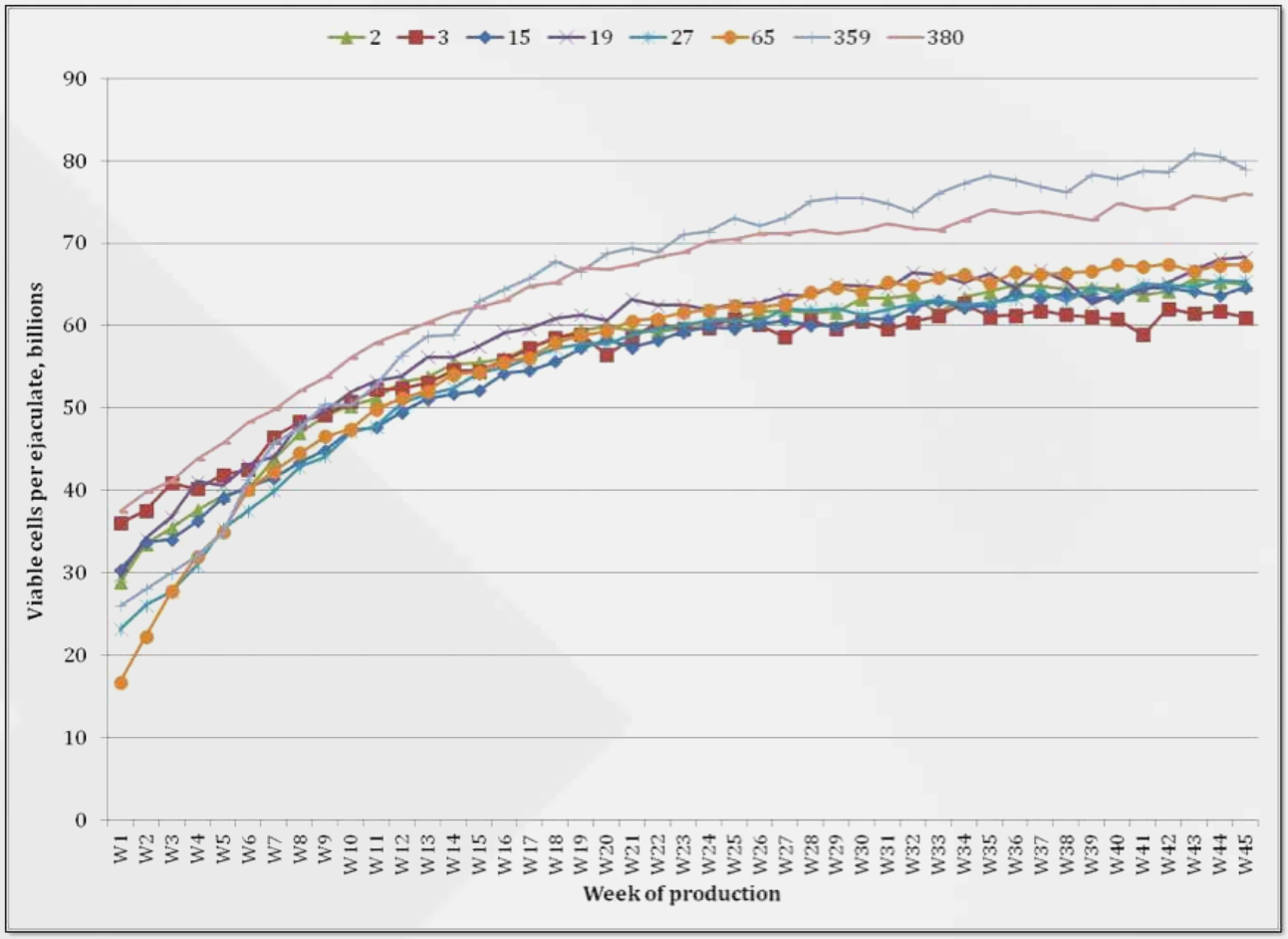
[a]Dependent on season and breed

PART 15:

PRODUCTION BENCHMARKS

Few studies have investigated the timeline for when the semen quality of an AI boar begins to deteriorate. Research (Wolf and Smital, 2009) suggests that semen volume, total sperm number and functional sperm reach their maximum by the time a boar is 2 years old. Sperm concentration increases until 11 months of age, followed by a decrease in concentration until boars are 3 years old. The percentage of abnormal sperm increases with time from 8 to 48 months of age. Motility, however, steadily decreases with time but only by a 1% decrease.

FIGURE 4. VIABLE CELLS PER EJACULATE PER LINE



Estimates for sperm cell and dose production for PIC lines can be made based on data from owned, affiliate, user group and customer boar studs (Figures 4-6; Tables 16-17).

TABLE 16. VIABLE CELLS PER EJACULATE BY GENETIC LINE AND PRODUCTION WEEKS

LINE	W1-5	W6-10	W11-15	W16-20	W21-25	W26-30	W31-35	W36-40	W41-45
2	35.0	46.1	53.8	58.2	60.0	62.0	63.2	64.6	64.8
3	39.3	47.5	53.3	57.4	59.8	59.9	61.0	61.2	61.0
15	34.7	43.5	50.4	56.0	58.8	60.3	62.1	63.6	64.3
19	36.5	47.4	55.4	60.3	62.5	64.0	65.7	64.6	66.5
27	28.6	42.3	51.3	56.7	60.0	61.6	62.6	63.7	65.1
65	26.8	44.2	52.3	57.5	61.4	63.5	65.5	66.6	67.2
359	30.2	47.1	57.9	66.6	70.8	74.3	76.0	77.3	79.5
380	41.6	52.0	60.2	65.4	69.1	71.3	72.5	73.7	75.1

FIGURE 5. DOSES PER EJACULATE PER LINE

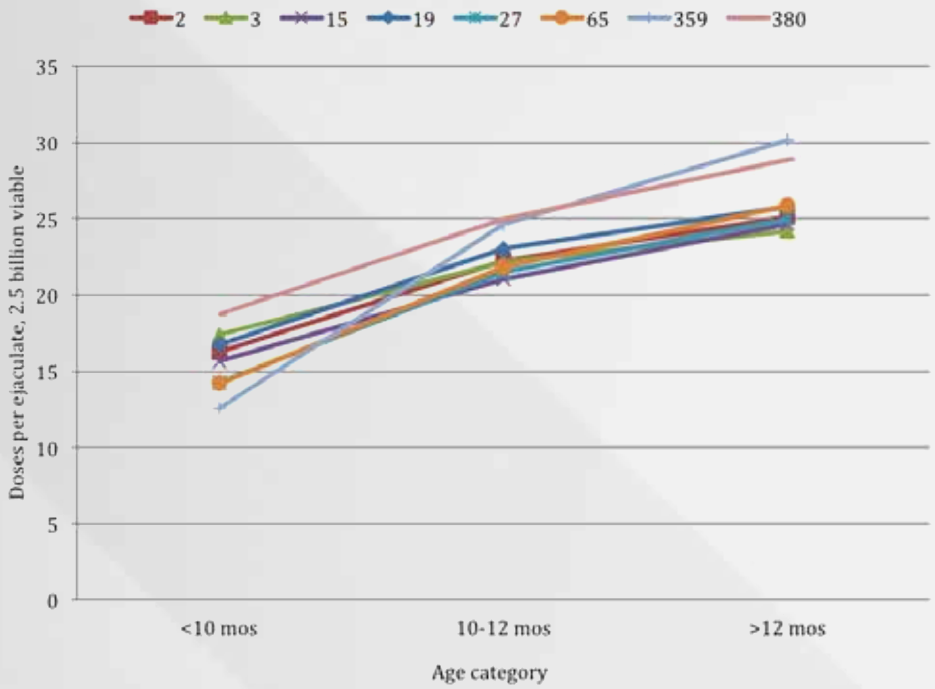
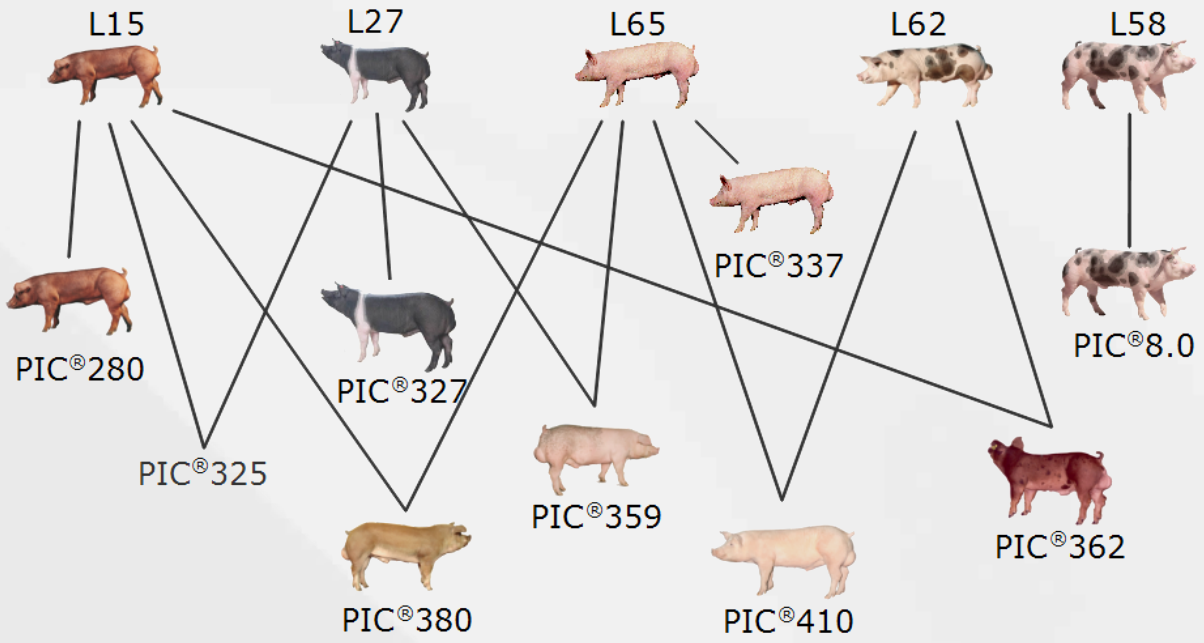


TABLE 17. AVERAGE NUMBER OF DOSES PER COLLECTION BY AGE AND GENETIC LINE ASSUMING DOSES OF 2.5 BILLION VIABLE SPERM CELLS AND AN AVERAGE OF 1.2 COLLECTIONS PER WEEK OVER THE BOAR'S LIFETIME.

LINE	<10 MOS	10-12 MOS	>12 MOS
2	16.2	22.2	25.1
3	17.4	22.1	24.2
15	15.6	21.1	24.7
19	16.8	23.0	25.8
27	14.2	21.4	25.0
65	14.2	21.8	25.8
359	18.0	24.6	30.1
380	18.7	24.9	28.8

FIGURE 6. PIC SIRELINE MAKEUP



PART 16:

BOAR LIFE AND REPLACEMENT RATE

Over the past few years, PIC, in association with university economists, has developed an economic model to determine the optimum time to cull a boar in a boar stud. Optimum Boar Life (OBL) uses customized cost inputs from studs (housing, feeding, purchase price, isolation costs, royalties, etc.) and projected revenues (the value of the doses of semen produced by the boar and the genetic value of the boar compared to its potential replacement) to objectively determine the optimum time a boar should remain in stud. There are two models of OBL that accommodate integrated customers that own both a boar stud(s) and breeding sows, and also a gene transfer center model that is specifically for customers who own a boar stud and sell semen.

This negates the need for "target" replacement rates, and provides objective metrics based on accurate, real-time information. Please contact Genetic Services at PIC for more information on OBL and its use in your system.

REFERENCES

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Appendix A:

NUTRIENT REQUIREMENTS FOR SWINE

NUTRIENT	UNITS	NRC[a] 2012	NSNG[b] 2010	PIC 2011
Vitamin A	IU/lb	1818	4,000	5,000
Vitamin D	IU/lb	91	300	800
Vitamin E	IU/lb	20	30	50
Vitamin K	mg/lb	0.5	2	2
Choline	mg/lb	568	0	300
Niacin	mg/lb	4.5	22	20
Riboflavin	mg/lb	1.7	4	4.5
d-Panthothenate	mg/lb	5.5	12	15
Vitamin B-12	mcg/lb	15	20	17
Folic Acid	mcg/lb	591	0	750
d-Biotin	mcg/lb	91	0	250
Thiamine	mg/lb	0.5	0	1
Pyridoxide	mg/lb	0.5	0	1.5
Zinc	ppm	100	165	125
Iron	ppm	100	165	100
Manganese	ppm	20	30	50
Copper	ppm	5	16	15
Iodine	ppm	0.5	0.3	0.65
Selenium	ppm	0.3	0.3	0.3

[a]NRC = Nutrient Requirements of Swine
[b]NSNG = National Swine Nutrition Guide

Appendix B:

PIC DIET SPECIFICATIONS AND EXAMPLE BOAR DIET

MINIMUM DIET SPECIFICATIONS[a]

NUTRIENT	
NRC ME, Kcal/lb	1400
Protein, %	16
Fiber, %	4.5 to 6.0
SID lysine[b], %	0.62
Calcium, %	0.80
aPhosphorus[c], %	0.40
Added salt, %	0.45
Linoleic acid, %	1.90

[a]Amount/lb of complete diet
[b]SID = Standardized ileal digestible
[c]a = available

EXAMPLE BOAR STUD DIET

INGREDIENT	PERCENT
Corn	69.32
Soybean Meal (2.62% SID Lysine)	13.75
Soybean Oil	1.00
Monocalcium Phosphate, 21% P	1.10
Limestone	1.20
Salt	0.45
Lysine HCl	0.11
DL-Methionine	0.02
L-Theronine	0.05
Soy Hulls	12.50
PIC Boar Stud VTM + Phystase	0.50
	100

Appendix C:

STEP BY STEP INSTRUCTIONS FOR USING AN AUTOMATED COLLECTION SYSTEM

1. Prepare the artificial cervix (AC) by placing it through the ring and wrapping the outer lining around it.

2. Squeeze the preputial diverticulum to empty the contents.

3. Once extended, clean the penis with a single-use disposable paper towel.

4. Attach the AC to the glove in the palm of your hand by exposing the tape. When the boar starts to thrust, grab and extend the penis.

5. Place the ends of AC into the holder and press down on the trigger. The tip of the penis should extend slightly beyond the end of the AC.

6. After collecting the pre-sperm fraction remove and discard the inner bag from the AC.

7. Place the outer bag inside the collection cup and use the ring and mouth of the cup to create a seal.

8. Attach the collection cup to the dummy, but don't use excessive force or it will bend. Release the sliding arm lock to allow free movement during the collection process. During collection, semen goes through the outer bag of the AC toward the collection bag filter located within the collection cup. Following ejaculation, the boar will withdraw his penis from the AC and dismount. Release the tension on the trigger to remove the AC and collection cup.

9. Remove the AC from the ring and discard.

10. Remove the top part from the collection bag that contains the filter, and discard. The ejaculate is now in the collection bag and can be delivered to the lab for processing.

Appendix D:

PACKAGING SEMEN DOSES FOR SHIPMENT USING DOUBLE COOLERS

1. Prepare liners and coolers.

2. Layer doses inside Thermalast bag within inner cooler.

3. Add a room temperature gel pack.

4. Put on the lid and seal with tape.

5. Wrap inner cooler with Thermalast bag.

6. Put inner cooler inside outer cooler and add gel packs (warm or cool depending on season).

7. Put on the lid and seal with tape.

8. Place in box for shipping.

Appendix E:

EXTENDER PREPARATION GUIDE

EXTENDER: XXX			MANUFACTURER'S RATIO:			50 G/KG OF WATER			TUB: 5KG		
Extender Volume (L or kg)	Water to add (kg)	Extender to add (g)	Extender Volume (L or kg)	Water to add (kg)	Extender to add (g)	Extender Volume (L or kg)	Water to add (kg)	Extender to add (g)	Extender Volume (L or kg)	Water to add (kg)	Extender to add (g)
1	1	50	26	26	1300	51	51	2550	76	76	3800
2	2	100	27	27	1350	52	52	2600	77	77	3850
3	3	150	28	28	1400	53	53	2650	78	78	3900
4	4	200	29	29	1450	54	54	2700	79	79	3950
5	5	250	30	30	1500	55	55	2750	80	80	4000
6	6	300	31	31	1550	56	56	2800	81	81	4050
7	7	350	32	32	1600	57	57	2850	82	82	4100
8	8	400	33	33	1650	58	58	2900	83	83	4150
9	9	450	34	34	1700	59	59	2950	84	84	4200
10	10	500	35	35	1750	60	60	3000	85	85	4250
11	11	550	36	36	1800	61	61	3050	86	86	4300
12	12	600	37	37	1850	62	62	3100	87	87	4350
13	13	650	38	38	1900	63	63	3150	88	88	4400
14	14	700	39	39	1950	64	64	3200	89	89	4450
15	15	750	40	40	2000	65	65	3250	90	90	4500
16	16	800	41	41	2050	66	66	3300	91	91	4550
17	17	850	42	42	2100	67	67	3350	92	92	4600
18	18	900	43	43	2150	68	68	3400	93	93	4650
19	19	950	44	44	2200	69	69	3450	94	94	4700
20	20	1000	45	45	2250	70	70	3500	95	95	4750
21	21	1050	46	46	2300	71	71	3550	96	96	4800
22	22	1100	47	47	2350	72	72	3600	97	97	4850
23	23	1150	48	48	2400	73	73	3650	98	98	4900
24	24	1200	49	49	2450	74	74	3700	99	99	4950
25	25	1250	50	50	2500	75	75	3750	100	100	5000

Appendix F:

WATER SAMPLE PREPARATION FOR THIRD PARTY ANALYSIS

(developed by G. Althouse, J. Morales and B. Thompson)

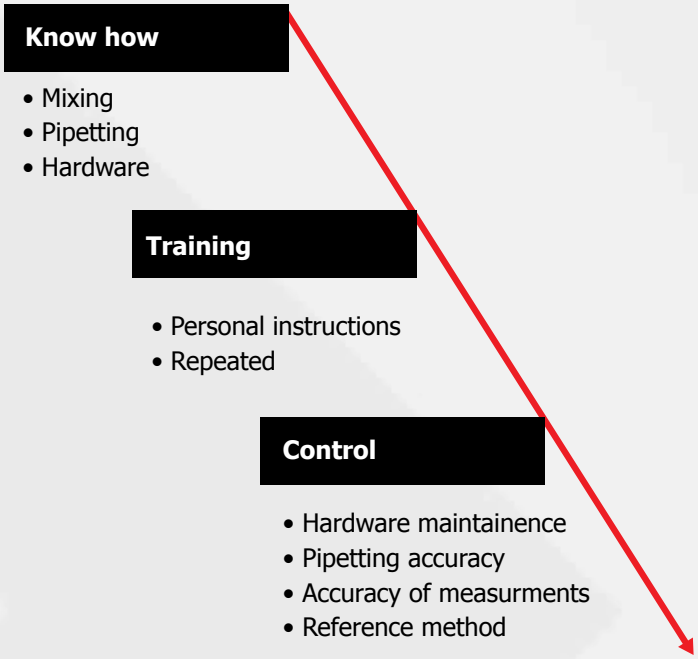
1. Water samples should be collected into a sterile Whirl-Pak® bag and sealed immediately after collection.
   a. Put on disposable gloves before sampling.
   b. Wipe the outer surface of the faucet with lint-free tissue lightly sprayed with 70% alcohol; be sure it is not saturated. Lightly insert the tissue into the end of the faucet. Wait 30 seconds for the alcohol to evaporate.
   c. Allow the RO water to run for 3 minutes to completely flush the lines.
   d. Open the Whirl-Pak® and collect the sample mid-stream from the flow.
   e. Sample both faucets with the same technique.

2. Diluted extender sampling:
   a. The individual who weighs and adds the extender to the water should wear a N95 mask and use disposable gloves when working with the extender powder.
   b. Use a similar technique with an alcohol wipe on the tubing from the peristaltic pump. Wipe the outer surface of the tubing connecting to the Auto-diluter and swab the inside of the tubing. Allow time for the alcohol to dry.
   c. Collect approximately 1 liter of diluted extender into a container. Decrease the flow and free-catch a sample into a Whirl-Pak®; seal immediately.

Appendix G:

ACCURACY OF PIPETTING AND SEMEN CONCENTRATION EVALUATION

INFLUENCES/MEASURES



TESTING PIPETTING ACCURACY

MEASURE	10 µl
1	9.7
2	9.7
3	9.3
4	9.5
5	9.8
6	9.8
7	9.9
8	9.6
9	9.9
10	9.4
MW	9.7
SD	0.2
CV	2.1
eS	-3.40

← Desired value (DV)

$$CV = \text{Coefficient of Variation} = \frac{SD}{MW} \times 100$$

$$eS = \text{Systematic error} = \frac{MW\text{-}DV}{DV} \times 100$$

In the shown example the standard deviation is 0,2 (or 2%). The systematic error is -3,4 %, so the measurements are too low. Reasons for the systematic error could be a poor calibrated pipette, wrong handling also.

Appendix H:

USE OF REFRACTOMETERS FOR SEMEN EXTENDER CONTROLS

Preparing the liquid semen-extender is one of the most vulnerable points at semen processing. Wrong water:extender powder ratios can have a negative effect on the viability of preserved semen-cells. Dependent on the scale of wrong mixing, this could lead to minimal negative effects up to 100% semen-mortality in the dose. Refractometry can be used as suitable, cheap and easy to use tool to control if the extender is prepared probably.

Refractometer:
Best choice is a Brix 18 refractometer. It has a scale from 1 to 18% Brix, divided in 1% main steps.

Calibration:
Calibrate the Refractometer every week according to the user manual. In most cases purified water is used to set the 0% Brix.

Set your benchmark:
The Brix-value varies between different extenders, dependent on their ingredients. Water-quality also influences the value. To set your benchmark it is required to know about your specific value which should be between 4 and 5% Brix in most cases. Measure your Brix value on five production days in a row to define your acceptable range. Note: This procedure has to be repeated every time you change extenders or extender-ingredients.

How to use the refractometer:
Make sure that your device is calibrated. Use a pipette to put a drop extender on the blue measurement area. It is important that the whole area is covered with fluid. Look trough the ocular and read the brix value. If the value is out of range (more than ± 0,2) make sure that your refractometer is calibrated and used in the right way, then repeat the measurement. In case the extender stands out of range do not use it for semen preservation and prepare a new vat.

Critical points:
- Refractometer should be calibrated regular
- Brix value can change if extender has changed or other ingredients are added
- Brix value can change if water-quality changes
- Measurement should be done at similar extender-temperatures every time
- The whole blue measurement-area has to be covered with fluid

Appendix I:

CHECKLISTS

Critical control points semen dose concentration

- Measurement
  - Scale calibration
  - Pipette calibration
  - Handling pipette (eS, CV)
  - Mixing prior sampling
  - Calibration photometer (settings CASA system)
  - Correct sample dilution
  - Clean cuvette/measurement chamber
  - Cuvette/measurement chamber without air bubbles
  - Impurities ejaculate (blood, germs, dust particles)

- Processing
  - Accuracy of dilution
  - Mixing prior/during filling process
  - Dose Volume

Critical control points bacterial contamination

- Barn
  - No preputial infections
  - Boars preputial hair trimmed
  - Boars are cleaned prior collection (dirt brushed down)
  - Preputium squeezed/emptied prior collection
  - Double gloved method (right technique)
  - First, clear ejaculate fraction discarded
  - Filter for collection vessel
  - Collection vessel no contact to floor
  - Clean/dry storage of collection bags, -filters, -vessels, -gloves
  - No contamination of heat cabinet
  - No squeezing of filter after collection
  - Fast delivery to lab for processing
  - Collection area cleaned daily

- Lab
  - No environmental bacterial contamination (surface samples)
  - Hand-washing and disinfection prior lab access
  - No hand touching of material for direct semen contact
  - Material with direct semen contact free from contamination
  - Quick semen extension/addition of antibiotics after collection
  - No water contamination
  - Pipes and hoses clean
  - Right antibiotic concentration used
  - Critical bacteria a sensitive for used antibiotics

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