

Evaluating Boar Semen for Quality

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Introduction

Evaluation of boar semen for measures of fertility is an important component to success with artificial insemination (AI). While measures of semen fertility are not highly related to fertility outcomes such as farrowing rate and litter size, use of poor quality semen with poor motility and increased abnormalities is associated with reduced fertility. As a result, the essentials for semen quality includes the basics for overall assessment of the ejaculate and the more specific measures for sperm cell concentration, motility, and percentage of normal sperm cells. It is these measures that are used to determine the number of fertile sperm cells that go into a dose of semen.

Performing an overall ejaculate assessment

Most ejaculates are expected to fall into the normal ranges for mature, healthy boars that are greater than 10 months of age and are collected 1 to 2 times a week with at least 3 days of rest between collections. This includes the following:



Normal Ejaculate Measures

Total volume of ejaculate:	200-400 mL
Sperm rich volume:	100-200 mL
Sperm cells/ejaculate:	60-130 billion
Normal sperm :	80-90%
Motile sperm :	80%
Live sperm :	90%
Doses (of 3 billion sperm):	20 doses/ejaculate

Many ejaculate collection procedures involve obtaining at least 2/3 of the whole ejaculate but not collecting the pre-sperm and post-sperm fractions. This technique limits contamination and dilution of the semen for processing. The ejaculate is typically assessed for volume by direct

measurement for volume or indirect measurement using weight.

Concentration

Concentration is an essential measure of total sperm cells in the ejaculate. This measure is important for determining the number of doses and how many sperm (motile and normal) will be in each dose. The three primary way of measuring concentration include: hemacytometer, photometer, and computer assisted semen analysis (CASA).



The hemacytometer is a glass counting chamber similar to a microscope slide. A diluted semen sample is placed on the slide and sperm cells are counted within the slide counting chamber using visual count under a microscope. This method requires an initial investment of \$1,200 for a microscope, hemacytometer, and pipets and about \$1.00 in disposables for each sample analyzed. This method takes about 10 to 15 minutes and is accurate.

The photometer method uses a machine that measures the amount of light that can pass through a semen sample. A sample is diluted at a predetermined rate and then placed into the machine to get a reading. The method requires an initial investment of \$2,000-\$3000 for the machine and pipets, and \$1.00 in disposables for each sample. The method is highly accurate if ejaculates are within the normal ranges. The method is fast and requires less than 5 minutes per sample.

The last method is the CASA system. This system uses microscopic visualization of sperm cells in a diluted semen sample and individually counts and assesses each sperm cell. The systems may cost between \$30 to \$50,000 but is

highly accurate as well as fast. Each sample may require \$3.00 in disposables for each sample measurement.

Motility

Semen sample analysis for motility is highly valued as a measure for identifying samples of poor quality. The measure of motility, while not highly related to farrowing rate and litter size, must be greater than 70% to reach acceptable fertility limits. Most doses of semen are adjusted in sperm cell numbers for non motile cells. To measure motility, the following procedures are used for semen samples:

1. dilute in extender
2. Warm at 37°C for 5-10 minutes and place on a warmed microscope slide and add a cover slip.
3. Evaluate sample at low (100 X) or high (>200 X) magnification for measures of motility on a 0 to 100% basis.
 - At low magnification, samples are often graded as good or poor, or some percentage is assigned by subjective viewing.
 - At high magnification, count 10 cells (motile and non-motile cells) in a defined field. This process is repeated in new microscope fields 5 to 10 times to get an estimate that is more precise.
 - The CASA machine also measures motility as well, and is highly accurate and fast in tracking each sperm cell. Specified procedures are followed for each machine.

Sperm cell morphology

Sperm cells are sometimes fragile, and are subject to problems in development, formation, and handling. While there are billions of sperm in an ejaculate, not all are normal. In fact, a significant percentage of the sperm in an ejaculate may be abnormal and could reduce overall fertility. For this reason, semen samples are evaluated under low to high magnification for some assessment for abnormalities.

The following procedures are followed:

1. The sperm cells are usually fixed to stop movement using a stain solution.
2. Microscope
 - If using a standard light microscope, use a stain such as Rose-Bengal, to aid in seeing the sperm cells.
 - If using a phase contrast microscope (\$3,000 or more), no stain is needed, just use the appropriate condenser settings.
3. Place a drop of stained and fixed sperm on a slide

and add coverslip.

4. Count ten sperm in a field record abnormal sperm/ 10 sperm counted and repeat in 5-10 more fields.
5. The number of abnormal cells is used to adjust for correct numbers of normal sperm cells in the final dose. Sperm cells are examined for head and tail normality. The most common problems to look for involve sperm cells with cytoplasmic droplets on the tail, detached heads, and bent or coiled tails.

Other assessments

There are few other assessments that are of great value in determining the quality of a semen sample, but measure of clumping and bacteria presence can be important. A measure of the level of agglutination, or what percent of the sperm cells in the field of view under low magnification, are stuck to each other. This can indicate some problem in the fertility of the ejaculate. In addition, under high magnification, stained or even phase contrast viewing, can often reveal live bacteria. These are usually identified by their motion in the field without sperm in the immediate vicinity. These samples are of high concern and indicate a problem in either boar health or collection methodology.

For more information, please search for the following resources in PIG:

PIG References :

- Evaluating Boar Semen

PIG Factsheets:

- Semen Collection, Evaluation