



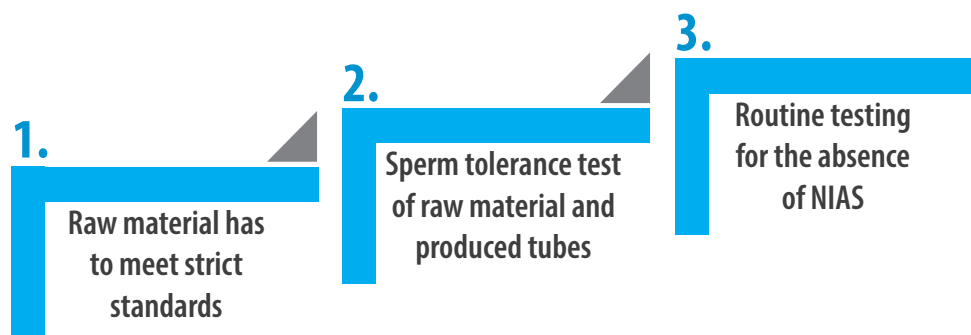
Quality standard of Minitube's boar semen tubes

Dr. Monika Esch, Minitüb GmbH

Modern pig production relies heavily on artificial insemination (AI) to ensure efficient breeding programs, fast genetic progress, and high biosecurity standards. The benefits of AI are based on the extreme improvement of availability and dissemination of superior genetics by increasing the number of inseminations per boar and the extended storage time of preserved semen, enabling shipment of semen conveniently to any place where these genetics are needed.

To ensure the above-mentioned benefits, the materials used to manufacture semen containers must be certified, tested, and proven to be biologically inactive and sperm friendly. This responsibility lies with the manufacturers of such semen containers.

Minitube as a specialized and certified producer of boar semen tubes has a rigorous quality control system in place, consisting of a three-level concept for the quality assurance and testing of raw materials and the final product.



1. Raw material standards

Minitube's boar semen tubes are entirely produced from only one raw material, i.e., low density polyethylene (LDPE). Each batch of LDPE assigned for the production of boar semen tubes at Minitube's manufacturing plant is accompanied by a batch specific certificate and meets the following standards*:



- The material has been manufactured in accordance with the European Commission's Regulation (EC) No 2023/2006 on **good manufacturing practice** for materials and articles intended to come in contact with food.
- According to the US Code of Federal Regulations issued by the Food and Drug Administration (FDA), Title 21, §177.1520(c), the material may be used in articles for packing or holding food.

Pure LDPE is by definition free from any toxic ingredients like Bisphenol A, heavy metals, phthalates or adipates.

**More country specific standards are available upon request.*

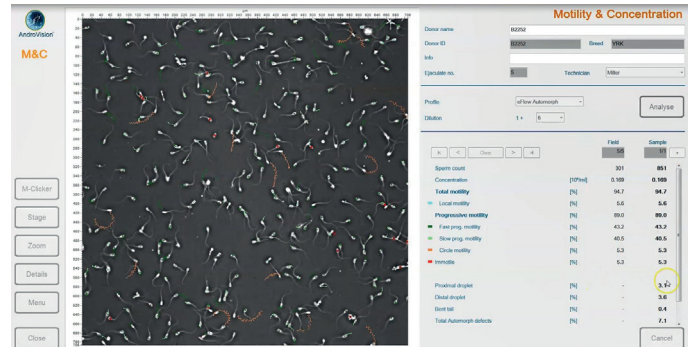
2. Sperm tolerance

Every batch of the raw material LDPE undergoes rigorous testing for sperm-friendliness before it is finally released for the production of tubes. These quality assurance tests are routinely performed at the Unit of Reproductive Medicine of The Clinics, University of Veterinary Medicine Hannover Foundation, which is a DAKkS accredited GLP laboratory in accordance with the international norm ISO/IEC 17025.



For these tests, boar semen of at least 3 different boars is extended with Androstar® Plus, divided into split samples and incubated in the boar semen tubes to be tested and in glass vials as a control. Semen is evaluated over a semen storage period of 6 days. All split samples are stored in a semen storage unit at 17°C and analyzed according to the following protocol:

- Total and progressive motility (CASA) after 24, 72 and 144 h
- Acrosome integrity and membrane integrity (flow cytometry, H33342/ PNA/PI) after 24 and 144 h
- Thermo-resistance test after 144 h storage at 17°C (incubation at +38°C for 120 min), then analysis of progressive motility (CASA)
- Mitochondria membrane potential (flow cytometry, H33342/PI/JC-1) after 24 and 144 h
- Membrane fluidity (flow cytometry, H33342/Yo Pro 1/Merocyanin (M540)) after 24 and 144 h
- Complete morphology (wet mount, phase contrast microscopy 1000x) after 24 and 144 h



3. Tests for the absence of non-intentionally added substances (NIAS)

Non-intentionally added substances (NIAS) may be accidentally added to the raw material during production or transport and therefore pose a risk to the quality of the final product. Although substances like heavy metals, endocrine disruptors and plasticizers like Bisphenol A, phthalates and adipates must not be present in the material as per its formulation and certification, those substances can cause adverse biological effects in animals, as well as in human beings in case they are added non-intentionally. Most importantly in the context of boar semen preservation, these substances can cause damage to the sperm cells during storage and hamper fertility and productivity of the inseminated sows.

Therefore, in the third level of Minitube's quality assurance program, the absence of NIAS is controlled with random checks of the raw materials for endocrine disruptors and plasticizers like Bisphenol A, phthalates and adipates as well as heavy metals.

It is a matter of principle for Minitube as a manufacturer of semen packaging materials to take full responsibility in only using substances and raw materials which are sperm-friendly and help to preserve the high quality of insemination doses.

This is only possible when the full production line is in control of the manufacturer, as is the case with Minitube's production of boar semen tubes in its own specialized production facility.



Tube production facility at Minitube