

कृत्रिम गर्भाधान उपकरणों की सफाई और  
बंध्याकरण — रीति संहिता  
( पहला पुनरीक्षण )

**Cleaning and Sterilization of  
Artificial Insemination Equipment —  
Code of Practice**

( *First Revision* )

ICS 65.020.30

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भारतीय मानक ब्यूरो  
BUREAU OF INDIAN STANDARDS  
मानक भवन, 9 बहादुर शाह ज़फर मार्ग, नई दिल्ली - 110002  
MANAK BHAVAN, 9 BAHADUR SHAH ZAFAR MARG  
NEW DELHI - 110002  
[www.bis.gov.in](http://www.bis.gov.in) [www.standardsbis.in](http://www.standardsbis.in)

## FOREWORD

This Indian Standard (First Revision) was adopted by the Bureau of Indian Standards, after the draft finalized by the Animal Husbandry and Equipment Sectional Committee had been approved by the Food and Agriculture Division Council.

Artificial insemination (AI) is being increasingly used for improving genetic potentialities of the livestock in the country. This necessitates application of properly cleaned and sterilized AI equipment and material to help in controlling spread of diseases from one animal to the other. This standard has been formulated to provide guidelines on efficient cleaning and sterilization of equipment and material used in AI work.

This standard was first published in 1976. In this revision, after considering the suggestions of various semen stations in the country, the requirements for cleaning and sterilization of glassware, rubber ware, and other equipment have been modified to align with the latest practices. During the revision of this standard, considerable assistance has been derived from the 'Minimum standards for production of bovine frozen semen' issued by the Department of Animal Husbandry and Dairying, Ministry of Fisheries, Animal Husbandry and Dairying, Government of India, New Delhi.

The composition of the Committee responsible for the formulation of this standard is given in [Annex A](#).

For the purpose of deciding whether a particular requirement of this standard is complied with, the final value observed or calculated, expressing the result of a test or analysis, shall be rounded off in accordance with IS 2 : 2022 'Rules for rounding off numerical values (*second revision*)'. The number of significant places retained in the rounded off value should be the same as that of the specified value in this standard.

*Indian Standard***CLEANING AND STERILIZATION OF ARTIFICIAL  
INSEMINATION EQUIPMENT — CODE OF PRACTICE***( First Revision )***1 SCOPE**

This standard provides guidelines for cleaning and sterilization of equipment and material used for processing, handling and transport of semen; and for artificial insemination (AI) and allied work.

**2 CLEANING AND STERILIZATION OF  
GLASSWARE****2.1 Cleaning**

The glassware shall be washed thoroughly with running tap water and soaked in warm, non-spermicidal neutral detergent solution for about 30 min. The glassware shall be cleaned by appropriate nylon brush and rinsed with running tap water. The collection tubes shall be brushed at least 3 times and thoroughly cleaned and rinsed with distilled water. Finally, the glassware shall be rinsed three times with double distilled water and allowed to dry by keeping them inverted on a blotting paper or a drying stand made of either stainless steel or plastic.

**2.2 Sterilization**

After cleaning (*see 2.1*), the open end/s of the dried glassware shall be covered with aluminium foil and sterilized in hot air oven at 160° C for one hour or at 180 °C for 30 min. For ensuring proper sterilization, one item should be wrapped with brown paper as its mild charring indicates that proper sterilization is done.

**3 CLEANING AND STERILIZATION OF  
ARTIFICIAL VAGINA (AV)****3.1 Cleaning**

Immediately after collection, cone from the AV and water from AV jacket shall be removed. Cones and AVs shall be cleaned thoroughly with a soft sponge brush under running tap water and then submerged in warm neutral detergent for about 30 min, followed by proper rinsing in warm and clean water. Again, these shall be rinsed for three times with double distilled water.

**3.2 Sterilization**

After cleaning (*see 3.1*), fully assembled AVs shall be autoclaved at 34.4 kPa (5 psi) pressure for 20 min. During sterilization, the valve of AV shall be kept open. Alternatively, AV sterilizer (use double distilled water in the sterilizer) may be used for proper sterilization of AVs. The AV and cones shall be sterilized for 30 min, after water starts boiling in the AV sterilizer. Thereafter, AVs filled with water and covered with aluminum foil, at both ends shall be stored overnight in an incubator at 45 °C to stabilize the AV temperature.

**4 CLEANING AND STERILIZATION OF  
RUBBERWARE****4.1 Cleaning**

The washing and cleaning procedure of rubber wares is similar to that of glassware (*see 2.1*). Care shall be taken to clean the rubber wares with sponge brush instead of nylon brush. Plastic tips shall be cleaned by water jet with force using a suitable syringe.

**4.2 Sterilization**

Sterilization technique, however, differs owing to the thermo-sensitivity of the rubber items. Thermo-resistant rubber ware shall be sterilized by autoclaving at 20.7 kPa to 57.6 kPa (3 psi to 4 psi) pressure for 10 min. Plastic tips shall be autoclaved at 34.4 kPa (5 psi) pressure for 20 min. The rubber tubing for semen filling shall not be reused.

**5 STERILIZATION OF BUFFERS**

**5.1** Fresh double distilled water or ultra-pure water shall be autoclaved at 103.4 kPa (15 psi) pressure for 15 min and used for preparation of the buffer/dilutor.

**5.2** Buffer shall be sterilized by microfiltration where 0.2 µm membrane filter may be preferred. If the buffer is prepared on the previous day, it should be stored in the refrigerator and antibiotics should be added next day in the morning after warming it to 34 degree celcius.

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## **IS 8103 : 2024**

**5.3** If, buffer is prepared on previous day, the antibiotics along with egg yolk shall be added on the day of semen collection.

### **6 STERILIZATION OF FILTER PAPERS AND BACTERIOLOGICAL MEDIA**

**6.1** A bunch of clean filter papers (No. 1) of standard brand (thrashed to remove dirt, if any) shall be wrapped in thick cotton cloth for sterilization in an autoclave at 34.4 kPa (5 psi) pressure for 20 min. Alternately, these can be sterilized dry in suitably sized petri dishes in hot air oven at 180 °C for 30 min.

**6.2** Bacteriological Media shall be autoclaved at 103.4 kPa (15 psi) pressure for 15 min.

### **7 DISINFECTION OF LIQUID NITROGEN (LN) CONTAINERS USED FOR TRANSPORT OF SEMEN**

The LN containers returned/received from outside shall be disinfected thoroughly with 4 percent sodium carbonate solution.

### **8 CLEANING AND STERILIZATION OF EQUIPMENT USED FOR ARTIFICIAL INSEMINATION (AI) WORK**

To control spread of diseases from one animal to the

other, proper cleaning and sterilization of AI gun and other accessories is essential and shall be done as given below:

**8.1** Artificial insemination gun, scissors and other accessories shall be cleaned whenever they get soiled or at least once a week with hot water and then air dried.

**8.2** Artificial insemination gun and the scissor shall be sterilized with isopropyl alcohol after drying. The AI gun piston (if removable) and the scissors should be wiped clean with water after each insemination. Soaps are lethal to semen, and therefore, should not be used to clean equipment.

**8.3** Bull apron shall be autoclaved at 34.4 kPa (5 psi) pressure for 20 min.

**8.4** Surgical equipment shall be autoclaved at 103.4 kPa (15 psi) pressure for 30 min.

**8.5** To ensure better hygiene, use of individually packed sterilized sheaths instead of sheaths available in bulk packing is recommended.

## ANNEX A

*(Foreword)*

## COMMITTEE COMPOSITION

Animal Husbandry and Equipment Sectional Committee, FAD 32

<i>Organization</i>	<i>Representative(s)</i>
Sher-e-Kashmir University of Agricultural Sciences & Technology of Jammu, Jammu	DR BHUPENDRA NATH TRIPATHI ( <b>Chairperson</b> )
All India Poultry Breeders Association, New Delhi	DR A. K. RAJPUT DR R. K. JAISWAL ( <i>Alternate</i> )
Animal Welfare Board of India, Faridabad	MS PRACHI JAIN DR DEBALINA MITRA ( <i>Alternate</i> )
Bihar Animal Sciences University, Patna	DR DEEP NARAYAN SINGH DR RANJANA SINHA ( <i>Alternate</i> )
Dau Shri Vasudev Chandrakar Kamdhenu Vishwavidyalaya, Anjora	DR DHIRENDRA BHOSLE DR O. P. DINANI ( <i>Alternate</i> )
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Federation of Indian Animal Protection Organizations, New Delhi	DR SIRJANA NIJJAR DR DINESH MOHITE ( <i>Alternate</i> )
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National Dairy Development Board, Anand	DR R. O. GUPTA DR A.V. HARIKUMAR ( <i>Alternate</i> )
National Dairy Research Institute, Karnal	DR ARUN KUMAR MISRA DR SURENDER SINGH LATHWAL ( <i>Alternate</i> )
National Egg Coordination Committee, New Delhi	SHRI AJIT SINGH SHRI BHAGWATI SINGH ( <i>Alternate</i> )
National Institute of Animal Nutrition and Physiology, Bengaluru	DR RAVI KIRAN G. DR RAMACHANDRAN ( <i>Alternate</i> )

**IS 8103 : 2024**

<i>Organization</i>	<i>Representative(s)</i>
PETA India, Mumbai	DR KIRAN AHUJA MS FARHAT U. I. AIN ( <i>Alternate</i> )
People for Animals, New Delhi	MS GAURI MAULEKHI MS SHREYA PAROPKARI ( <i>Alternate</i> )
Poultry Federation of India, Sonipat	SHRI RANPAL DHANDA SHRI RAHUL KHATRI ( <i>Alternate</i> )
Tamil Nadu Veterinary and Animal Sciences University, Chennai	DR S. MEENAKSHI SUNDARAM DR M. R. SRINIVASAN ( <i>Alternate</i> )
Uttar Pradesh Pandit Deen Dayal Upadhyaya Pashu Chikitsa Vigyan Vishwavidyalaya Evam Go-Anusandhan Sansthan University (DUVASU), Mathura	DR YAJUVENDRA SINGH DR MUNEENDRA KUMAR ( <i>Alternate</i> )
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SHRI PRADEEP SHARMA  
SCIENTIST 'B'/ASSISTANT DIRECTOR  
(FOOD AND AGRICULTURE), BIS

Panel on Expert Panel for Review of Standards on Animal Husbandry Equipment Panel, FAD 32 : P2

<i>Organization</i>	<i>Representative(s)</i>
ICAR - Indian Veterinary Research Institute, Bareilly	DR SUBRATA KUMAR GHOSH ( <i>Convener</i> )
Guru Angad Dev Veterinary and Animal Sciences University, Ludhiana	DR NAVDEEP SINGH
National Dairy Development Board, Anand	DR R. O. GUPTA
Tamil Nadu Veterinary and Animal Sciences University, Chennai	DR S. MEENAKSHI SUNDARAM



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## BUREAU OF INDIAN STANDARDS

### Headquarters:

Manak Bhavan, 9 Bahadur Shah Zafar Marg, New Delhi 110002

Telephones: 2323 0131, 2323 3375, 2323 9402

Website: [www.bis.gov.in](http://www.bis.gov.in)

### Regional Offices:

	Telephones
Central : 601/A, Konnectus Tower -1, 6 <sup>th</sup> Floor, DMRC Building, Bhavbhuti Marg, New Delhi 110002	{ 2323 7617
Eastern : 8 <sup>th</sup> Floor, Plot No 7/7 & 7/8, CP Block, Sector V, Salt Lake, Kolkata, West Bengal 700091	{ 2367 0012 2320 9474
Northern : Plot No. 4-A, Sector 27-B, Madhya Marg, Chandigarh 160019	{ 265 9930
Southern : C.I.T. Campus, IV Cross Road, Taramani, Chennai 600113	{ 2254 1442 2254 1216
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