

## SCHEDULE 4

Regulations 10 and 11

### CONDITIONS RELATING TO THE COLLECTION, PROCESSING STORAGE AND TRANSPORT OF BOVINE EMBRYOS FOR THE PURPOSES OF INTRA-AREA TRADE

1. Bovine embryos shall be collected and processed by an approved collection team, without coming into contact with any other consignment of bovine embryos not meeting the requirements of these Regulations relating to bovine embryos intended for intra-Area trade.

2. Bovine embryos shall be collected in a place which is isolated from other parts of the premises or holding and which must be in good repair and easy to cleanse and disinfect.

3. Bovine embryos shall be processed (that is, examined, washed, treated and placed in identifiable and sterile containers) in either a permanent laboratory facility or a mobile laboratory facility, which is not situated in a zone subject to prohibition or quarantine measures.

4. All implements which come into contact with the bovine embryos or the donor animal during collection and processing shall be disposable or shall be properly disinfected or sterilised prior to use.

5. Products of animal origin used during collection of the bovine embryos and in the transport medium shall be obtained from sources which present no animal health risk or are treated prior to use in such a way that such risk is prevented. All media and solutions shall be sterilised by approved methods according to the recommendations of the manual of the International Embryo Transfer Society<sup>(1)</sup>. Antibiotics may be added to the media in accordance with that manual.

6. Storage flasks and transport flasks shall be properly disinfected or sterilised before the commencement of each filling operation.

7. The cryogenic agent used shall not have been previously used for other products of animal origin.

8. Each bovine embryo container and the containers in which they are stored and transported shall be clearly code-marked in such a way that the date of collection of the bovine embryos and the breed and identification of the donor sire and donor dam, as well as the registration number of the team can be readily established.

9. Each bovine embryo shall be washed at least 10 times in a special fluid for bovine embryos which shall be changed each time and which shall contain trypsin, in accordance with internationally recognised procedures. Each wash shall be a 100-fold dilution of the previous wash and a sterile micropipette shall be used to transfer the bovine embryo on each occasion.

10. After the last wash each bovine embryo shall be subjected to microscopic examination at a magnification of at least 50× over its entire surface to determine that the *zona pellucida* is intact and is free from any adherent material. Any micromanipulation which involves penetration of the *zona pellucida* must be carried out after the last wash and examination in the facilities approved for the purpose. Such micromanipulation may only be carried out on a bovine embryo having an intact *zona pellucida*.

11. Each consignment of bovine embryos that has successfully undergone the examination provided for in the preceding paragraph shall be placed in a sterile container marked in accordance with paragraph 8 of this Schedule and sealed immediately.

12. Each bovine embryo shall, where appropriate, be frozen as soon as possible and stored in a place which is under the control of the team veterinarian and which is subject to regular inspection by an official veterinarian.

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(1) Edited by D A Stringfellow and S M Siedel and published in November 1990 by the International Embryo Transfer Society. It is obtainable from their headquarters at 309 West Clark Street, Champaign, Illinois, U.S.A.

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**13.** Each collection team must submit routine samples of flushing fluids, washing fluids, disintegrated bovine embryos, non-fertilised ova etc., resulting from its activities to the appropriate Minister for official examination for bacterial and viral contamination.