SCHEDULE 1

Regulation 5

Conditions for the licensing of bovine embryo collection teams for all purposes including for intra-Area trade

The collection, processing and storage of embryos must be carried out either by the collection team veterinarian or under his authority by one or more technicians who are competent and trained by the collection team veterinarian in methods and techniques of hygiene.

SCHEDULE 2

Regulation 5

Part I

Permanently sited laboratories for all purposes including for intra-Area trade

1. The permanently sited laboratory, must have—
   (a) a work surface, a microscope and cryogenic equipment;
   (b) a room where embryos can be manipulated which is adjacent to but physically separate from the area used to handle the donor animals during collection;
   (c) a room or area equipped for cleansing and sterilising instruments and equipment used in embryo collection and manipulation; and
   (d) where micro-manipulation of the embryo which involves penetration of the *zona pellucida* is to be carried out, suitable laminar-flow facilities.

Part II

Mobile laboratories for all purposes including for intra-Area trade

1. The mobile laboratory must have—
   (a) a work surface, a microscope and cryogenic equipment; and
   (b) two separate sections, one for the examination and manipulation of embryos which shall be a clean section, and the other for accommodating equipment and materials used in contact with the donor animals.

2. The mobile laboratory shall always have contact with a permanently sited laboratory to ensure the sterilisation of its equipment and the provision of fluids and other products necessary for the collection and manipulation of embryos.

Part III

Mobile laboratories for the purposes of trade within Northern Ireland

1. The mobile laboratory shall—
   (a) have separate parts so that there is no contact between used and unused equipment and materials;
   (b) carry sufficient equipment to enable the examination and manipulation of embryos to be carried out without contaminating them; and

(c) have contact with a permanently sited laboratory to ensure the sterilisation of its equipment and the provision of fluids and other products necessary for the collection and manipulation of embryos.

SCHEDULE 3

Regulation 6(1) and 6(3)

Conditions relating to the collection, processing and storage of embryos *not intended* for intra-Area trade

1. Embryos shall be processed in either a permanent laboratory facility or a mobile laboratory facility licensed under regulation 5(1) or (2), which is not situated in a zone subject to prohibition or quarantine measures.

2. All implements which come into contact with the embryos or the donor animal during collection and processing shall be properly disinfected or sterilised before use.

3. Products of animal origin used during collection of the embryos and in the transport medium shall be obtained from sources which present no animal health risk or are treated before use in such a way that such risk is prevented.

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

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

6. Each embryo container and the containers in which they are stored and transported shall be clearly marked with—

(a) the appropriate distinguishing number of the collection team;

(b) the date of the collection of the embryo; and

(c) either—

(i) the breed and identification of the donor sire and donor cow, or

(ii) a code from which this information can be readily established.

7. Each embryo shall be washed at least 10 times in a special fluid for embryos which shall be changed each time. Each wash shall be a 100-fold dilution of the previous wash and a sterile micropipette shall be used to transfer the embryo on each occasion.

8. After the last wash each embryo shall be subjected to microscopic examination over its entire surface to determine that the *zona pellucida* is intact and is free from any adherent material.

9. Each consignment of embryos that has successfully undergone the examination provided for in paragraph 8 shall be placed in sterile containers marked in accordance with paragraph 6 and which shall be sealed immediately; and in this paragraph "consignment of embryos" means a quantity of embryos removed in one operation from a single donor.

SCHEDULE 4

Regulation 6(1) to (3)

Conditions relating to the collection, processing, storage and transport of embryos for the purposes of intra-Area trade

1. Embryos shall be collected, processed and packed without coming into contact with any other consignment of embryos not meeting the requirements of these Regulations relating to embryos intended for intra-Area trade.

2. 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. Embryos shall be processed in either a permanent laboratory facility or a mobile laboratory facility licensed under regulation 5(1), which is not situated in a zone subject to veterinary prohibition or quarantine measures.

4. All implements which come into contact with the 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 embryos and in the transport medium shall be obtained from sources which present no animal health risk or are to be so treated prior to use so 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(**1**). 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 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 embryos and the breed and identification of the donor sire and donor cow, as well as the distinguishing number of the team can be readily established.

9. Each embryo shall be washed at least 10 times in a special fluid for embryos which shall be changed each time and which shall contain tryspin, 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 embryo on each occasion.

10. After the last wash each embryo shall be subjected to microscopic examination at a magnification of at least 50x over its entire surface to determine that the *zona pellucida* is intact and is free from any adherent material. Any micro-manipulation 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 micro-manipulation may only be carried out on an embryo having an intact *zona pellucida.*

11. Each consignment of 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 and sealed immediately.

12. Each 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 a veterinary officer of the Department.

(**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, USA

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

SCHEDULE 5

Regulation 6(2) and 8(*d*)

Conditions relating to donor animals for the purposes of Regulation 6 or 8

Part I

Collection from live animals

1. The donor cow must have spent at least the previous six months in an EEA State or, in the case of any other country, in the country of collection.

2. The donor cow must have been present in the herd of origin for at least 30 days prior to collection.

3. The donor cow must come from a herd which is—

(a) officially tuberculosis free;

(b) officially brucellosis free or brucellosis free; and

(c) either enzootic bovine leucosis free or for which certification has been obtained that there has not been any clinical case of enzootic bovine leucosis during the past three years.

4. During the year before collection of the embryo, the donor cow must not have been present in a herd (or herds) which have shown any clinical sign of infectious bovine rhinotracheitis or infectious pustular vulvovaginitis.

5. On the day of embryo collection the donor cow—

(a) shall be kept in a holding which is not subject to veterinary prohibition or quarantine measures;

(b) shall show no clinical sign of disease.

Part II

Collection after slaughter

1. The donor cow shall not have been designated for slaughter as part of a national disease eradication programme, nor shall it have come from a holding subject to veterinary prohibition or restriction.

2. The abattoir where the ovaries and other tissues are collected must not be situated in a zone subject to veterinary prohibition or quarantine measures.

SCHEDULE 6

Regulation 7

Conditions for the licensing of bovine embryo production teams

1. The production, processing and storage of embryos must be carried out either by the team veterinarian or under his authority by one or more technicians who are competent and trained by the team veterinarian in methods and techniques of hygiene.

2. The production team personnel must be trained in appropriate disease control and laboratory techniques, particularly in procedures for working in sterile conditions.

3. The team must have at its disposal a permanently-sited processing laboratory which must—

(a) have adequate equipment and facilities, including a separate room for recovering oocytes from ovaries, and separate rooms or areas for processing oocytes and embryos, and storing embryos;

(b) have laminar-flow facilities.

4. Where oocytes and other tissues are to be collected in an abattoir, the production team must have at its disposal suitable equipment for the collection and transport of the ovaries and other tissues to the processing laboratory in a hygienic and safe manner.

SCHEDULE 7

Regulation 8(*c*)

Conditions relating to the production of embryos

1. Embryos shall be produced by a licensed bovine embryo production team, and, in the case of embryos intended for intra-Area trade, without coming into contact with any other consignment of embryos not meeting the requirements of these Regulations relating to embryos intended for intra-Area trade.

2. Ovaries, oocytes and other tissues intended to be used in embryo production 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. Each bovine embryo production team must submit routine samples of flushing fluids, washing fluids, disintegrated embryos, non-fertilised ova etc, resulting from its activities to the Department for official examination for bacterial and viral contamination.

4. When ovaries, oocytes, and other tissues are to be collected at an abattoir, the abattoir should be officially approved and under the control of a veterinary officer of the Department who shall carry out ante- and post-morten inspection of donors.

5. Materials and equipment coming into direct contact with ovaries, oocytes and other tissues shall be sterilised before use, and after sterilisation, used exclusively for those purposes. Separate equipment shall be used to handle oocytes and embryos from different batches of donor animals. All laminar-flow facilities shall be properly cleansed and disinfected between batches.

6. Embryos shall be produced, processed and placed in identifiable and sterile containers in a permanent laboratory facility which is not situated in a zone subject to prohibition or quarantine measures.

7. Products of animal origin used during production of the embryos and in the transport medium shall be obtained from sources which present no animal health risk or are to be so treated prior to use so 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. Antibiotics may be added to the media in accordance with that manual.

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

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

10. Oocytes, semen and embryos shall be processed using the laminar-flow facility. However, the centrifugation of semen may be carried out outside the laminar-flow facility, as long as full hygienic precautions are taken.

11. Once the embryo has been produced, each 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 embryos and the breed and identification of the donor sire and donor dam or batch, as well as the registration number of the team can be readily established.

12. After the culture procedure has been completed, each embryo shall be washed at least 10 times in a special fluid for embryos which shall be changed each time (and which shall, for embryos intended for intra-Area trade, contain tryspin, 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 embryo on each occasion.

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

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

15. Each 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 a veterinary officer of the Department.

16. Ovaries, oocytes and other tissues shall not be allowed to enter the processing laboratory until completion of the post-mortem inspection of the batch of donors. If any disease that might be transmitted in the material and make it unsuitable for producing embryos is found in the batch of donors, or in any animals slaughtered in that abattoir on that day, all tissues from the batch must be traced and discarded.

17. Only embryos from the same batch of donors should be stored in the same ampoule or straw.

SCHEDULE 8

Regulation 15(4)

Bovine embryo transfer

FORM OF CERTIFICATE

Serial No.....

1. Registration number of the nominated embryo transplantation team .....

2. Name and address of the owner of the animals identified in the attached Annex

.....

.....

.....

3. I hereby certify that the animal(s) identified in the attached annex was/were examined by me on (date) ..... at (address of premises)

.....

.....

.....

and

(*a*) showed no clinical sign of disease;

(*b*) showed no significant abnormalities of the reproductive tract(s) or birth canal(s); and

(*c*) were in appropriate bodily condition and of a suitable size and conformation to receive the intended embryo(s) as specified in the attached Annex.

4. On the basis of the above examination, I am of the opinion that the animal(s) is/are suitable to receive the embryo(s). I know of no reason existing at the time of my examination which would cause me to believe that the animal(s) would not be able to carry to term a normal calf of the breed and type specified and to calve naturally.

Signed ..... RCVS

Name (Block Capitals).....

Date.....

Name of Practice .....

Address of Practice .....

.....

.....

ANNEX

	Recipient Identification (Ear Tag No)	*Recipient Breed and Type*	*Breed and Type of Intended Embryo(s)*
1			
2			
3			
4			
5			
6			
7			
8			
9			
10			
11			
12			

This Annex is provided in respect of the certificate, Serial No ..... as laid down in Schedule 8 to the Bovine Embryo Collection, Production and Transplantation Regulations (Northern Ireland) 1996.

Signed..... RCVS

Name..... (Block Capitals)

Date .....

NOTE: The examining Veterinary Surgeon is required to sign immediately beneath the last entry on the above Annex