OVERSEAS MARKET ACCESS REQUIREMENTS NOTIFICATION
ANIMAL PRODUCTS ACT 1999
STANDARDS
MINISTRY OF AGRICULTURE AND FORESTRY NEW ZEALAND

Ref**:** AE-EU-00
Date**:** 7 July 2011

Statutory Authority

Pursuant to section 60 of the Animal Products Act 1999:

(i) I notify the following overseas market access requirements and specifications, entitled European Union Germplasm Export Requirements Part 3 Bovine Embryos

(ii) Revoke European Union Germplasm Export Requirements Part 3 Bovine Embryos: 1 June 2011.

This notice takes effect from date of signing.

Dated at Wellington this 12th day of July 2011.

Signed: Matthew Stone BVSc MACVSc MVS (Epidemiology)
Group Manager
Animal Imports and Exports Group
Imports & Exports Directorate
Standards Branch
MAF Biosecurity New Zealand
(pursuant to delegated authority)

Explanatory Note

These Export Requirements are the European Union Member States requirements for Bovine Embryos.

This OMAR shall be read in conjunction with the European Union Germplasm Export Requirements Part 1.

Part 3 Bovine Embryos

Note: to be read in conjunction with Part 1: General

3.1 Application

3.1.1 This Part applies to *in-vivo* derived embryos and *in-vitro* produced embryos of domestic bovine animals.

3.1.2 An embryo collection team must not carry out *in-vitro* fertilisation and/or *in-vitro* culture of embryos for the EU unless the team is approved by MAF and the EU as an EU-listed embryo production team.

The EU lists embryo collection teams and embryo production teams separately.

3.1.3 MAF may authorise the storage of semen in the premises approved for embryo storage as part of the embryos collection/production team's approval, provided that the semen complies with Part 2 of these Export Requirements. The semen must be stored in separate storage containers.

3.2 Facility requirements

3.2.1 An *in-vivo* embryo collection team must have access to either a permanently sited laboratory, or a mobile laboratory, as appropriate to their scope of operation.

3.2.2 A permanently sited laboratory must have:

a. a room where embryos can be examined and manipulated which is adjacent to but physically separate from the area used to handle the donor animals during collection

The permanent laboratory may be at another site.

b. a room or area equipped for cleaning and sterilising instruments and equipment used in embryo collection and manipulation, as appropriate.

A room or area is only required when it is applicable to the scope of the approval, and when instruments and equipment are not single-use.

3.2.3 Mobile laboratories must have a specially equipped part of the vehicle consisting of two (2) separate sections; one being a clean section for the examination and manipulation of embryos, and the other a section for accommodating equipment and materials used in contact with the donor animals.

3.2.4 Unless it uses only single-use equipment and commercially pre-packaged media, the mobile laboratory must 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 processing of embryos.

3.2.5 An *in-vitro* embryo production team must have a permanently sited laboratory which has:

a. adequate equipment and facilities, including separate rooms for:
   i. recovering oocytes from ovaries
   ii. processing oocytes, ova and embryos
   iii. storing embryos.

b. a laminar-flow cabinet or other suitable facilities where specific sterile activities (processing of ova, embryos and semen) are carried out

Centrifuging of semen may be done outside the laminar-flow facility, as long as full hygiene precautions are taken.

c. in the case of ova and other tissues collected from a slaughterhouse, suitable equipment for the collection and transport of ovaries and other tissues to the processing laboratory in a hygienic and safe manner.

3.2.6 The design and lay-out of the embryo team's processing and storage facilities must ensure that cross-contamination of embryos is prevented.

3.2.7 An embryo storage facility must comprise of at least one (1) lockable room for the storage of ova or embryos. The construction must be such that the storage room can be readily cleaned and disinfected.

3.3 Operational requirements

3.3.1 Approval of an embryo collection team requires:

a. the collection, processing and storage of embryos must be carried out either by a team veterinarian, or by one or more team technicians under the responsibility of the team veterinarian
b. the team technicians must be competent and trained by the team veterinarian in methods and techniques of hygiene and in techniques and principles of disease control
c. the team must be under the general supervision and authority of the recognised person
d. there must be access by the team to permanent or mobile laboratory facilities where embryos can be examined, processed and packed, consisting of at least:
    i. a work surface
    ii. an optical or stereo microscope
    iii. cryogenic equipment, where necessary.

3.3.2 In order to be given approval, the team members of an *in-vitro* embryo production team must also have received adequate training on disease control and laboratory techniques, particularly in procedures for working in sterile conditions.

3.3.3 Embryo teams must be inspected by a recognised person at least twice a year, to verify the conditions of approval and supervision.

The verification would normally include records, standard operating procedures and internal audits, as well as compliance with the sanitary conditions regarding collection, processing and storage of embryos. Further inspections can occur in addition to the audits carried out twice a year.

3.3.4 Any major change in the organisation of the team is to be notified to MAF.

3.3.5 The embryo team must be re-approved whenever the team veterinarian is replaced, or whenever any major changes are made in its organisation or the laboratories at its disposal.

3.3.6 The team veterinarian must inform the recognised person of any failure of compliance with these Export Requirements as soon as possible, and before affected embryos are exported to the EU.

3.3.7 Embryos must be collected in a place that, at the time of collection, is isolated from other parts of the premises or property, and which must be in good repair and easy to clean and disinfect.

3.3.8 Embryos must be collected and processed by an approved collection team, without coming into contact with any other consignment of embryos not meeting these Export Requirements.

3.3.9 Embryos must be processed (examined, washed, treated and placed in identified and sterile containers) in either a permanent laboratory facility or a mobile laboratory facility, which is not situated on a property or in a zone subject to animal health prohibition or quarantine measures.

Embryos/oocytes collected on farm may be transferred to a permanent laboratory at another site.

3.3.10 All equipment which comes into contact with the embryos or the donor animal during the collection and processing must either be single-use disposable, or must be disinfected or sterilised prior to use in accordance with the current IETS manual.

3.3.11 Any biological products of animal origin used in the media and solutions for collection, processing, washing or storage of embryos must be free of pathogenic micro-organisms.

3.3.12 Media and solutions used in the collection, processing, washing or storage of embryos must be sterilised by approved methods in accordance with the current IETS manual, and handled in such a manner to ensure that sterility is maintained.

Pre-packaged, commercially prepared media and solutions that are sterile do not need to be re-sterilised.

3.3.13 Antibiotics may be added, where appropriate, to media and solutions in accordance with the current IETS manual.

3.3.14 Only embryos from a single donor can be washed together. In addition, only ten (10) or fewer embryos from a single donor can be washed together.

3.3.15 Each embryo must be washed at least ten (10) times, with some of the washes containing trypsin, in accordance with the current IETS manual. Each wash must be at least a hundred (100) -fold dilution of the previous wash and a sterile micro-pipette must be used to transfer the embryos on each occasion.

3.3.16 After the last wash, each embryo must 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. If defects in the zona pellucida are noted after washing, the defective embryos should be removed and the washing procedure must be repeated for the remaining embryos using fresh washes.

3.3.17 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.

3.3.18 Each flush of embryos that has successfully undergone the microscopic examination must be placed in a sterile container (straw/ampoule) marked in accordance with clause 3.3.19 and must be sealed immediately.

3.3.19 Each frozen embryo container (straw/ampoule) and the containers in which they are stored and transported (goblets and canes) must be labelled in accordance with the standardised system in the current IETS manual.

This means being code-marked in such a way that the date of collection of the embryos, the breed and identification of the donor sire and donor dam, and the registration number of the embryo collection team can be readily established.

3.3.20 Storage and transport containers must be disinfected or sterilised before the commencement of each filling operation.

3.3.21 The cryogenic agent used for the preservation and storage of embryos must not have been previously used for other products of animal origin.

3.3.22 Each embryo must, where appropriate, be frozen as soon as possible and stored in a facility that is under the control of the team veterinarian, and which is subject to regular inspection by a recognised person.

3.3.23 Each collection team must store routine samples of flushing fluids, washing fluids, disintegrated embryos, non-fertilised ova etc., resulting from its activities, and have these available to send to a laboratory recognised or approved by MAF for examination for bacterial and viral contamination, as required.

These samples must be stored at or below -70°C for a minimum of twelve (12) months from the date of collection.

The samples should include: - at least one 0.5 ml straw of pooled uterine flushing fluid (*in-vivo*) or maturation media (*in-vitro*) - at least one 0.5 ml of pooled embryo washing fluid from the last four (4) washes - at least one 0.5 ml straw of non-fertilised ova, disintegrated embryos or non-transferable embryos, if available. These samples should be stored separately (i.e. in a separate tank) from EU-eligible germplasm.

3.3.24 Each collection team must keep a record of its activities in respect of embryo collection during the twelve (12) months before and twelve (12) months after storage including:

a. the breed, age and identification of the donor animals concerned

In this context, the donor animals include sires.

b. a record of the health history, all diagnostic tests and results, and all vaccinations and treatments carried out on donor animals
c. the place of collection, processing and storage of embryos collected by the team
d. the identification of the embryos together with details of their destination, if known
e. details of micro-manipulation of embryos which involve penetration of the zona pellucida, if applicable.

3.3.25 The operational requirements in the clauses above apply as appropriate to the collection, processing, storage and transport of ovaries, oocytes and other tissues for use in *in-vitro* fertilisation and/or *in-vitro* culture. In addition, when ovaries and other tissues are to be collected at a slaughterhouse:

a. the donor animals must be traceable to the property of origin
b. the slaughterhouse must be officially approved and under the control of an official veterinarian whose responsibility it is to ensure that ante-mortem and post-mortem

inspection of donors is carried out, and certify that the donor animals were free of clinical signs of relevant contagious diseases transmissible by embryos (as per 3.4)

c. materials and equipment coming into direct contact with ovaries and other tissues must be sterilised before use. Separate equipment must be used to handle oocytes and embryos from different batches of donor animals
d. only embryos from the same batch of donors may be stored in the same ampoule/straw.

3.3.26 Each embryo collection or production team must ensure that the embryos are stored at suitable temperatures in premises approved for the purpose by MAF as the competent authority.

3.3.27 From the time of collection until thirty (30) days after (or in the case of fresh embryos, until the day of dispatch) the embryos were stored in New Zealand in which there was no outbreak of bovine disease affecting the eligibility of the embryos for the EU.

3.3.28 The embryo storage premises must have permanent records of all incoming and outgoing movements of embryos, including the final destination.

3.4 Procurement of donor females

3.4.1 Immediately prior to the embryo/oocyte collection, the donor females must have spent at least the previous six (6) months in New Zealand and in a maximum of two (2) herds.

3.4.2 The herd(s) referred to in 3.4.1 must have the following health status:

a. officially Tb free (i.e. C2 or greater) during the previous six (6) months
b. officially brucellosis free during the previous six (6) months
c. free from EBL, or were from a herd which is not free but for which certification has been obtained that there has not been any clinical case of EBL during the previous three (3) years

In this context, the certification would be both an owner declaration and a veterinary declaration.

d. during the previous twelve (12) months they have not been present in a herd(s) which showed any clinical signs of IBR/IPV.

3.4.3 The donor females must have been present in the herd of origin for at least thirty (30) days prior to collection.

In this context, where the donors are transported to a permanent embryo collection facility, the herd of origin is the herd from which the donor animals came immediately prior to entering the facility. Once they have been resident for more than thirty (30) days, the herd on the collection facility becomes a herd of origin.

3.4.4 On the day of embryo/oocyte collection the donor female:

a. showed no clinical signs of disease

This excludes minor diseases which have no adverse effect or cannot be transmitted through embryos.

b. must come from a property that was not subject to animal health control measures
c. in the case of donors of ovaries and other tissues to be collected after slaughter in a slaughterhouse, must not be from animals that were designated for slaughter as part of a national disease eradication programme, and must not originate from a property which was subject to restrictions because of animal disease

d. in the case of donors of ovaries and other tissues to be collected after slaughter, the slaughterhouse was not situated in an area which was subject to animal health control measures.

3.5 Semen for fertilisation

3.5.1 Unless specifically authorised, EU eligible embryos must not be conceived as a result of natural mating.

3.5.2 Semen used for *in-vivo* fertilisation must be derived from:

a. semen collection or storage centres listed with the EU in accordance with Part 2 of these Export Requirements; or
b. semen collection or storage centres approved by MAF, which may include approved centres not subject to the requirements of Part 2 of these Export Requirements; or
c. semen collection or storage centres approved by the competent authority of the EU, the United States of America, Canada, Australia, Switzerland or Croatia.

If non-New Zealand produced semen is used, supporting documentation will be required from the competent authority from the EU, USA, Canada, Australia, Switzerland or Croatia that the semen was derived from an approved collection or storage centre. Supporting documentation in this context means a copy of the export certificate from the relevant country to NZ.

Note that the listed countries are those approved as of 2011. Appropriate enquiries should be made in advance to confirm the current situation.

3.5.3 Semen used for *in-vitro* fertilisation must be derived from:

a. semen collection or storage centres listed with the EU in accordance with Part 2 of these Export Requirements; or
b. semen collection or storage centres approved by MAF, which may include approved centres not subject to the requirements of Part 2 of these Export Requirements. These embryos are not eligible for intra-community trade.

In-vitro embryos fertilised with semen from non-EU listed semen centres may represent a potential disease risk, and so must be implanted into recipients present in the Member State of destination as indicated on the export certificate.

Appendix 1: Risk Analysis *Guidance Information*

Purpose / Scope
To identify the risk organisms relating to disease transmission that are reasonably likely to occur, and ensure that appropriate controls are included in the centre work manual so that the semen meets the EU Export Requirements.

Identification of Biological Hazards from Inputs and Process Steps

Process step	Inputs	Hazard reasonably likely to occur	Justification	Control Measures	Reference
1. Procurement of donors	Donor cow	Tb, EBL, IBR miscellaneous pathogens	Sporadic incidence of infection may occur	• Resident in NZ 6 months, and in a maximum or 2 herds • Come from herds of known disease status • Present in herd of origin for 30 days prior to collection	(a) Procurement of donors 3.4.1 3.4.2, 3.4.4 3.4.3
2. Embryo fertilisation	Semen	Tb, EBL, BVD, IBR, *Campylobacter*, *Trichomonas* miscellaneous pathogens	Sporadic incidence of infection may occur	• For in-vivo, semen from either EU semen centre, MAF approved semen centre, or imported semen • For in-vitro, semen from either EU semen centre or MAF approved semen centre	(b) Semen source 3.5.2 3.5.3
3. Embryo collection	Donor cow	Tb, EBL, BVD, IBR miscellaneous pathogens	Sporadic incidence of infection may occur	• Donors show no clinical sign of disease on day of collection • Facilities able to be cleaned and disinfected • Trypsin wash of embryos	(c) Control of collection 3.4.4 3.3.7 3.3.15

Process step	Inputs	Hazard reasonably likely to occur	Justification	Control Measures	Reference
				• Embryos held 30 days post collection to ensure donor health status not changed	3.3.27
	Other animals	Tb, EBL, BVD, IBR miscellaneous pathogens	Direct contact	• Donors isolated during collection • Facilities able to be cleaned and disinfected • Trypsin wash of embryos	3.3. 3.3.7 3.3.15
	Equipment	Tb, EBL, IBR miscellaneous pathogens	Disease transmission may be indirect	• Equipment must be single use disposable, or disinfected prior to use	3.3.10
	Media	None	Free of pathogenic organisms	• Media and solutions sterile	3.3.12
4. Embryo processing	Embryos	IBR miscellaneous pathogens	Disease may be present in donor	• Equipment must be single use disposable, or disinfected prior to use • Trypsin wash of embryos • Intact zona pellucida	(d) Control of processing 3.3.10 3.3.15 3.3.16
	Equipment	None	Single use disposable, or disinfected prior to use	• Equipment must be single use disposable, or disinfected prior to use	3.3.10
	Media	None	Free of pathogenic organisms	• Media and solutions sterile	3.3.12
	Packaging	None	Single use disposable, or disinfected prior to use	• Equipment must be single use disposable, or disinfected prior to use	3.3.10
5. Storage	Packaging	None	Disinfected prior to use	• Storage and transport containers must be disinfected prior to use	(e) Control of storage 3.3.20
	Cryogenic agent	None	New	• Must not have been used for other products of animal origin	3.3.21

Control measures
(a) Procurement of donors Risks associated with donor cows is managed by: • The donor cows must be resident in NZ for 6 months, and in a maximum of 2 herds • Come from herds of known disease status for Tb, EBL, and free of clinical signs of IBR • Present in herd of origin for 30 days prior to collection • Animals show no clinical sign of disease on day of collection • Bovine brucellosis not considered a risk organism as NZ has disease freedom.
(b) Semen source Risks associated with semen from donor bulls is managed by: • For in-vivo collection, semen from either EU semen centre, MAF approved semen centre, or imported from specified countries • For in-vitro production, semen from either EU semen centre or MAF approved semen centre.
(c) Control of collection Risks associated with donor cows is managed by: • Donor cows isolated from other cows during the physical collection • Facilities able to be cleaned and disinfected – non-porous surfaces on structures in direct contact with donors during collection (i.e. wood painted/sealed, concrete in good condition), flooring material in collection area either easily washable or able to be replaced (i.e. sand/bark) • Donor cows must show no clinical disease on the day embryos are collected • Donor cows adequately prepared – vulval area free of obvious faecal material • Equipment must be single use disposable, or disinfected prior to use • Media and solutions used are commercially prepared and sterile, so do not represent an animal health risk • Risk of diseases additionally managed by use of trypsin wash during embryo processing • Embryos held 30 days post collection to ensure donor health status not changed.

(d) Control of processing
Processing area physically separated from collection area
Equipment must be single use disposable, or disinfected prior to use
Media and solutions used are commercially prepared and sterile, so do not represent an animal health risk
Risk of diseases additionally managed by use of trypsin wash during embryo processing
Embryos examined to ensure they have an intact zona pellucida
Equipment for packaging, including straws, must be single use disposable, or disinfected prior to use
For in-vitro production, access to laminar flow cabinet and staff trained in sterile techniques.
(e) Control of storage
Storage area physically separated from collection and processing areas
Storage and transport containers must be disinfected prior to use
Cryogenic agent must not have been used for other products of animal origin.