

## The Netherlands

### Institutional and legal framework for ex situ conservation of AnGR

#### 20 years of gene bank development

First initiatives to start the development of a “prototype” gene bank started after a first inventory of the state of local breeds in the Netherlands in the late 1970's. This initiative was started by the deputy director of the Institute for Animal Research 'IVO Schoonoord', by storing semen of bulls and also some stallions, set aside by breeders and the breeding companies. In the early 1990's, the Dutch Foundation for Rare Domestic Animal Breeds (SZH), the Royal Dutch Cattle Syndicate (NRS) and AI organisation Holland Genetics (HG) took the initiative to formally establish a Foundation for the cryopreservation of AnGR, called the 'Foundation Gene Bank Farm Animals' (in Dutch: SGL). The goal of SGL was to contribute to the conservation and utilization of AnGR, taking into account the interest of the breeding industry and society interests.

In 2002, the Ministry of Agriculture, Nature Management and Fisheries published the policy document 'Sources of Existence: Conservation and the sustainable use of genetic diversity'. This policy document was the basis for the first national integrated plant and animal genetic resources programme, coordinated by the Centre for Genetic Resources, the Netherlands (CGN). CGN also became responsible for maintenance of AnGR gene bank collections, and started further development of gene bank collections for all major farm animal species in the Netherlands. Moreover, gene bank collections of SGL were transferred to CGN and - until today - breeding organisations contribute financially to maintenance of gene bank collections by CGN.

The gene bank collections of CGN are stored at two locations. Main storage is located at one of the locations of Wageningen UR Livestock Research (part of Wageningen University and Research Centre) in Lelystad. A duplicate storage site is located at the Veterinary Faculty of Utrecht University. The gene bank has a EU-certified storage site for cattle semen in Lelystad. Semen stocks of different species and of different veterinary health status are stored separately in different rooms and/or tanks. In addition, for security reasons we have duplicate collections in two separate locations (CGN Lelystad and Veterinary Faculty Utrecht). The storage of the duplicate collection also falls under the responsibility of CGN, and the Veterinary Faculty is contracted by CGN for this purpose.

#### Objective(s) of national cryopreservation programme/policy

CGN is collecting genetic material and maintains gene bank collections of cattle, pigs, horses, sheep, goat, chicken, duck and dogs. Both rare breeds and widely used/commercial breeds are represented in the gene bank. General objective of CGN is to conserve all rare domestic animal breeds in the gene bank and to stimulate animal breeders and breeding industry to back-up their commercial breeding populations in the gene bank of CGN.

For CGN, there is a number of reasons to conserve genetic material (*ex situ*) in the gene bank. Today or in the (far) future, gene bank material may be used:

- In breeding programs in case genetic problems (drift, inbreeding, genetic defects) occur in live populations
- To reconstruct a breed in case of extinction or loss of a substantial number of animals
- To create new lines or breeds or to quickly modify or reorient selection of a breed
- For research purposes to gain insight on genetic diversity of populations

#### Future plans

Apart from minor collections of embryos and oocytes, the Dutch national gene bank maintained by CGN currently contains relatively large collections of semen (Table 1). CGN will continue to expand semen collections, either to reach the required core collection size for native Dutch domestic animal breeds, or to further enlarge genetic variation stored for particular breeds. CGN aims to optimize cost efficiency of collecting new material to be added to the gene bank.

The aim of CGN is to lead or participate in scientific research towards improvement of cryoconservation protocols and reproductive techniques (different type of genetic material), and in conservation genetics research.

Currently, the Dutch government policy does not allow CGN to start freezing somatic cells as a parallel gene banking strategy. This because of the political sensitivity of reproductive cloning (and the discussions on cloning in the EU), where cloning often is directly associated with the storage of somatic cells.

### Collection categories

The first aim (but not the only aim) is to establish 'core collections' for all native Dutch breeds. The size of the core collection is based on the requirement to be able to reconstruct a breed in case of extinction. The number of samples (semen, semen and embryo's) needed depends on the species and several parameters (CGN developed the 'conservationplanner': planning tool available on [www.cgn.wur.nl](http://www.cgn.wur.nl)). For cattle and sheep for example we use as a 'rule of thumb' for semen collections that the core collection should consist of at least 25 (unrelated) donor animals, each contributing a minimum of 200 doses to the core collections.

In principle the core collection will not be touched, unless there is an emergency situation, or in specific cases the use of gene bank material may generate offspring that will add more genetic diversity to the core collections.

If possible and cost efficient the collections per breed will be further enlarged beyond the core collections. If the total collection exceeds the core collection requirements, there is material available to support breed conservation and breeding programmes or for research purposes.

Table 1. Semen collections of CGN in the Netherlands (April 2012)

Species	Number of breeds/lines by species	Number of males per breed (range)	Total number of insemination doses	Number of doses per donor (range)
<b>Cattle</b>	18	1 - 4.716	214.419	2 - 1.861
<b>Sheep</b>	7	13 - 69	22.548	1 - 616
<b>Goat</b>	2	5 - 24	3.965	15 - 364
<b>Horse</b>	6	1 - 26	1.983	1 - 88
<b>Pig</b>	27	1 - 47	16.298	4 - 251
<b>Chicken</b>	31	1 - 20	18.805	1 - 241
<b>Duck</b>	3	4 - 11	72	1 - 9
<b>Dog</b>	2	3 - 7	158	8 - 27

### Stakeholder participation

The mandate of CGN for its gene banking activities is based on the Dutch national policy on genetic resources (Sources of existence, 2002; Dutch country report on AnGR, 2002) and the commitment of the Dutch government to implement the Global Plan of Action for AnGR (FAO, 2007) and the implementation of the Convention on Biological Diversity (CBD, 1992).

There is not a specific national law or regulation for genetic resources, but CGN of Wageningen UR is requested to carry out so called Statutory Research Tasks for the Ministry of Economic Affairs, Agriculture and Innovation. CGN carries out these Tasks within 5 year contract periods for the Ministry of

Economic Affairs, Agriculture and Innovation, and in close collaboration with a variety of stakeholders. For plants, forestry and animals together there is a national committee governing the 5-year work plan of CGN. Specifically for farm animal gene banking there is a steering group, to advise CGN on objectives, priorities and access to the gene bank. There is a special relationship with the Dutch Foundation for Rare Domestic Animal Breeds (SZH), with a large variety of breed societies and breed interest groups, and with the cattle and pig breeding industry.

### Quality management system

The management of the national gene bank for AnGR is ISO certified (NEN-EN-ISO9002-2008). This is to guarantee good quality level of our work to the Ministry of Economic Affairs, Agriculture and Innovation, and to our collaborators, stakeholders and users. The primary processes of gene bank management include:

- selection and sampling of genetic material for the gene bank
- collection and transfer of genetic material to the gene bank facilities
- cryopreservation and storage of genetic material
- documentation and maintenance of the gene bank collections
- handling of applications to use gene bank material and distribution

We identified a number of critical points in our processes in terms of performance indicators. The following performance indicators are regularly monitored:

- cost efficiency of collection of genetic material by species
- correct and complete documentation of legal and sanitary status
- quality of genetic material before and after freezing
- number of publications and presentations per employee

### Sanitary regulations relevant for gene banking

The Dutch gene bank works in conformity with the national legislation for production, storage and trade of semen and embryos. For bovine and porcine semen national standards are equal to EC standards. The national standards for equine semen are slightly lower than EC Intra-Community trade and import standards. For ovine and caprine semen there are no specific national standards for production, storage and trade of semen. The latter is also the case for embryos.

Given the high national standards for production, storage and use of bovine semen, CGN applied for a derogation in national law, to be able to better support the preservation of rare domestic cattle breeds. For this objective there is sometimes the need to use old semen (1960 – 1980) that does not comply with the current national veterinary standards for distribution and use of bovine semen. Furthermore, sometimes bulls of rare cattle breeds do not meet the high health requirements for the herd of origin to enter semen collection centres (AI stations). In those cases, semen collection on farm usually can solve the problem, but distribution of semen collected on farm to other farmers (distributed by CGN) does not comply with national veterinary legislation for bulls semen. The derogation in national legislation for bovine semen includes a detailed protocol, designed specifically for CGN, for collection of semen on farm for the gene bank, and for distribution of semen that does not comply with the highest veterinary EC standards. The specific derogation for CGN will minimize veterinary risks, since CGN will carry out the same health tests as applicable for certified semen collection centres. For semen collection on farm the number of bulls per rare cattle breed is maximized to 5 per year. The number of semen doses distributed from the gene bank is approximately 200 (2011), and CGN keeps track of all semen doses distributed directly from the gene bank to the user/farmer, within the Netherlands. CGN will be audited every year by official national veterinary authorities.

## Ownership and access

It is important for a national gene bank to be clear about ownership of genetic material in the gene bank (including related data) and – even more important – about the criteria or conditions to get access to gene bank material. The starting point is the moment of collection of genetic material or transfer of genetic material to the gene bank (physically or not). CGN is using a model “Material Acquisition Agreements” (MAA) for this purpose. For access to gene bank material CGN is using a set of access criteria and a model “Material Transfer Agreements” (MTA). MAA or MTA usually have specific adaptations depending on the species and the results of negotiations with the provider or user.

### Material Acquisition Agreements (MAA)

Before genetic material will be collected or transferred to the gene bank, an agreement will be signed between CGN and the owner of the donor animal, or between CGN and the owner of the genetic material (MAA).

The MAA between the two parties includes the following components/conditions, which can be slightly adapted depending on the species or specific situation:

- Parties want to contribute to conservation of genetic diversity of the <breed> through cryopreservation of <type of genetic material> of <donor animal>.
- The <owner> of the <donor animal> agrees to make the <donor animal> available for collection of <type of genetic material> for the gene bank.
- Specific costs related to collection of <type of genetic material> will be covered by CGN.
- Ownership of genetic material collected for the gene bank will be transferred to CGN, unless agreed otherwise.
- CGN is responsible for cryopreservation of the <type of genetic material> collected, and for storage and documentation.
- Future applications to use the genetic material will be judged by CGN, based on a set of access criteria for use of gene bank material. Genetic material from the gene bank will only be issued after consultation of the relevant breed society.
- Users of gene bank material shall not claim intellectual property rights as a result of use of this material
- Owner of the <donor animal> declares that <donor animal> does not show any sign of diseases, and the owner of the <donor animal> will provide all relevant information about the disease status of the <donor animal>, and is willing to collaborate further in veterinary research on the <donor animal>.
- Together with the <type of genetic material> a blood sample will be taken from the <donor animal> and will be stored and owned by CGN.
- The owner of the <donor animal> declares that he/she has the right to make the <donor animal> available for collection of <genetic material> for the gene bank.
- The owner of the <donor animal> declares that he/she has the right to make available the <genetic material> for the purpose of this agreement and CGN will be protected against potential claims of third parties.

### Handling of applications to use gene bank material

Although *ex situ in vitro* conservation of genetic material has a long term insurance objective (“in case of extinction of a breed”), CGN gene bank material can also be used to support breeding of endangered populations and for research purposes. For this reason it is important to have clear access criteria. CGN will always receive written requests, including information about the applicant/user, the intended use and expected benefits. Use of gene bank material shall always contribute to the (long term) objectives of CGN, to contribute to maintenance of genetic diversity and conservation of rare breeds, or to promising research projects.

### Criteria for access to gene bank material

Requests to use gene bank material will have to fulfil the following main criteria:

- The contract between the original provider/owner of genetic material and the gene bank shall be respected. The contract may include specific restrictions, such as ownership issues or an embargo period for use of the material.
- There shall be a clear need to use gene bank material to support breeding of an endangered breed, in particular to minimize inbreeding rates.
- The respective breed society of the breed, will advise positively about the use of gene bank material for the intended purpose to support breed conservation.
- The requested material shall not be available commercially. CGN will provide genetic material free of charge and shall not compete with the commercial breeding and AI business.
- Genetic material of a particular animal of a particular breed can be issued if the total number of doses available in the gene bank exceeds the minimum "core collection" (species specific).
- If the number of available doses of a particular animal in the gene bank has reached a minimum threshold (e.g. for cattle: 50 doses), genetic material will not be provided, unless there is a serious calamity. In that case genetic material can be provided under specific conditions, and using the best available reproduction techniques or practices.
- For each requested mating, CGN will provide a maximum of 2 insemination doses. Applicants shall never distribute the genetic material from the gene bank further to other interested parties.
- Distribution of genetic material shall comply with the relevant national veterinary regulations (species dependent), including specific derogations for the gene bank.

For research requests the situation is different. In case of requests for research purposes, CGN will ask for a detailed research project proposal and will first judge whether i) it is a promising research project, ii) the use of gene bank material will be an essential contribution to the research project, iii) alternative options have been explored to get the same or similar genetic material. After this initial judgement, CGN will ask it's steering group if they agree with the judgement of CGN.

### Material Transfer Agreements (MTA)

If the decision about use of genetic material from the gene bank is positive, CGN will distribute the genetic material, including an MTA. The MTA is actually combined with the application form. The applicants signs the application and the MTA at the same time. The details of the MTA conditions depend on the species and the application, but will be based on the following set of components:

- Identification of the type and quantity of genetic material distributed and reference to the intended use of the material.
- In case expensive reproductive technologies are needed, CGN will agree with the user who will cover the additional costs.
- Liability statement related to the eventual sanitary risks of the material and to the (low) quality of the genetic material.
- CGN claims the right to collect genetic material from the resulting offspring of the application in the future.
- The applicant shall inform CGN about the results of the mating, about offspring born, and about 'left over' genetic material. User shall never distribute the genetic material further to other interested parties.
- User will not claim Intellectual Property Rights on the material or the offspring.