

France commits in the preservation of its horses and donkeys genetic resources

CRB-Anim project :

A Network of Biological Resources Centers for domestic animals



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The CRB Anim project

*A Network of Biological Resources Centers
for domestic animals*

CRB Anim

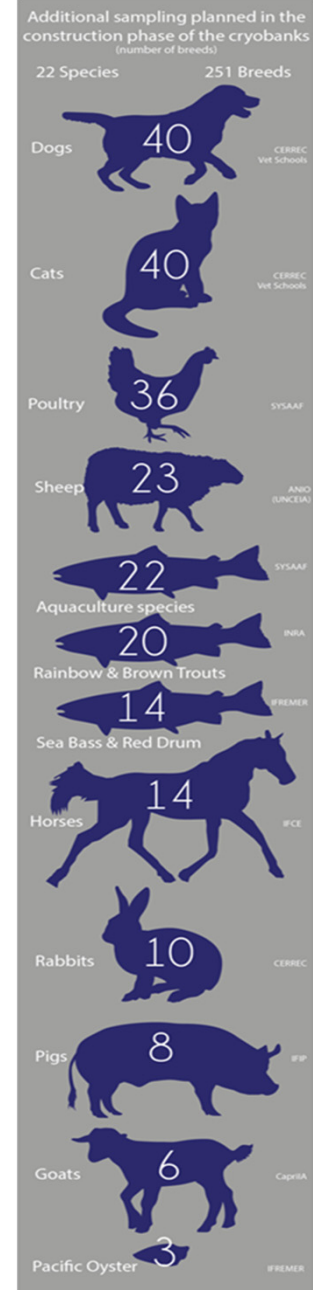
A multi-species project



Most of the national institutes (INRA, CNRS, VetAgroSup) and technical professional organisations (IFREMER, IFCE, SYSAAF, ...) are involved and have already contributed to the **French National Cryobank**.

A single project grouping 22 species supported by the National Agency of Research (ANR)

Financial support of 11M€ !
A 8 years project : 2013-2020



The CRB Anim project

Principal aims



- ❖ Improvement of biotechnologies for conservation of reproductive cells or tissues
 - Overcome actual technical locks
- ❖ Improvement of practices and synergies between the existing Centres of Biological Resources (CRBs),
- ❖ Enrichment of the collections : collection, development of new biotechnologies of reproduction
- ❖ Safety of collections, and identification
- ❖ Potential European extension of the French network

The French National Cryobank

Aims

CRYOBANQUE NATIONALE
Groupement d'Intérêt Scientifique

- ❖ **Patrimonial** conservation of biological resources
(long and safe storage)
- ❖ Easy use of biological samples in case of breed extinction
→ **Sustainable** use of genetic resources

Actually in the National Cryobank:

- **5477** donors from **14** species and **198** breeds
- Mainly **sperm cells** but also **embryos**, **tissues**
(depending on the species)



02

Equids in the CRB Anim project

Collaboration between INRA and IFCE

The CRB Anim project in the equine species

Two main objectives

- ❖ Overcome technical looks in cryopreservation techniques:



→ sperm cells



→ embryos

- ❖ Enrich the actual collections in the French National Cryobank

→ Be able to maintain the genetic diversity of equine breeds and help in the preservation of “endangered” species

Selection of 7 horse breeds (local and draught) and 7 ass breeds

↳ Funding : 455K€ for biological samples collection / 176K€ for research

The CRB Anim project in the equine species

1- Overcome technical looks in cryopreservation techniques



→ sperm cells

Actually, quite efficient method to freeze donkey sperm but:

- ❖ High variability in freezing methods and composition of extenders (containing animal products : egg yolk and milk)
- ❖ In all *in vivo* experiments, very low fertility rates (*Vidament et al., 2006; 2009*):
 - ❖ Pure-breeding → donkey x jenny (0% to 13%)
 - ❖ Crossbreeding → donkey x mare (36% to 60%)

Except in a recent paper from Italy (*Rota et al., 2012*)

Pure-breeding → donkey x jenny (20% to 60%)

The CRB Anim project in the equine species

1- Overcome technical looks in cryopreservation techniques



→ sperm cells

Our hypothesis : the freezing extender could be the key factor of the success

Chemically defined extender to reduce:

- ❖ presence of animal products in the extender
toxicity against the jenny genital tract / sperm cells ?
- ❖ interactions between components of the extender
- ❖ interactions between components and sperm cells

→ INRA Freeze extender (formulated for stallion sperm) seems to be efficient but surprising data in sperm quality characteristics.....to be confirmed

See: *Milon et al* , EAAP2014 Copenhagen, S32 Discovery session Horse Commission

The CRB Anim project in the equine species

1- Overcome technical looks in cryopreservation techniques



→ embryos

Advantages to freeze embryos:

- ❖ Easier to recreate a breed
 - to genotype (sex, genetic diseases...)
 - to increase national and international exchanges
- ❖ Reduce the costs of cryobanking
- ❖ Reduce the costs and management of embryo transfer
 - actually transfer requires a mares' herd (fresh or 24h cooled embryos)*

→ Low fertility rate after transfer using the “traditional” freezing technique
....except very new data using vitrification of embryos (F. Guignot et al.)

See: Guignot, F., EAAP2013 Nantes, S03 New advances in Biotechnology of Reproduction

The CRB Anim project in the equine species

1- Overcome technical looks in cryopreservation techniques



→ embryos

Equine embryo specificities:

- ❖ The presence of a capsule reduces the penetration of cryoprotectants
- ❖ The size of the embryo (# 250-300µm) at Day 6.5 - 7 in uterus
 - Both reduce the success of cryopreservation

Objective (M.Caillaud, IFCE):

- ❖ Get smaller embryos in the uterus to improve the success of freezing
 - Accelerate the descent of the embryo by prostaglandins (PGE2)

In parallel F.Guignot (INRA) project on vitrification

→ birth of 4 foals from frozen embryos , collaboration INRA – IFCE

The CRB Anim project in the equine species

2 - Enrich the collections of the Cryobank

Today in Cryobank

- 18 horse breeds
- 2 ass breeds



7 horse breeds and 7 ass breeds have been identified as “endangered”

→ Increase the number of donors in each breed

Actual collections in Cryobank of “endangered” breeds

Stallion/donkey Breed	Nb of donors'	Nb of doses
Auxois	3	79
Boulonnais	5	119
Castillon	0	0
Cob Normand	4	242
Landais	0	0
Poitevin Mulassier	3	260
Trait du Nord	1	39
Âne du Cotentin	0	0
Âne Grand Noir du Berry	1	3
Âne Normand	0	0
Âne Bourbonnais	0	0
Âne de Provence	0	0
Âne des Pyrénées	0	0
Baudet du Poitou	6	192

The CRB Anim project in the equine species

2 - Enrich the collections of the Cryobank

To be able to reintroduce a breed :

↳ 13 stallions per breed are needed

→ CRB-Anim funding : 5 stallions or donkeys per breed

1 - Reasoned choice of the breeds according to:

❖ Genetic aspects as:

- major ancestors (to limit consanguinity in the breed),
- breed composition (to preserve the original characteristics of the breed)

❖ Originality of some Sires:

- particular phenotypes (hair and coat colours) or disease

❖ Quality of semen

2 - Selection of approved Freezing Centres (precise specifications)

Thanks to.....



European Federation of
Animal Science

and



EAAP Horse
Commission



Network of Biological Resources
Centers for domestic animals



Management of the project



- Selection of breeds
- Relations with ass breeders



.....Thanks for attention !

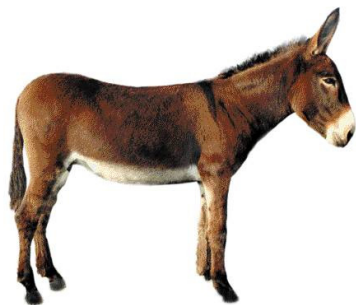


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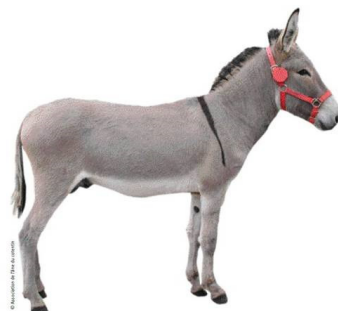


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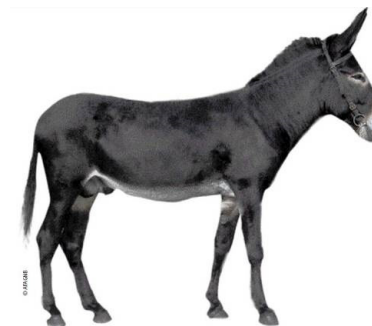
7 endangered ass breeds



Âne du Bourbonnais



Âne du Cotentin



Âne Grand Noir du Berry



Âne Normand



Baudet du Poitou



Âne de Provence



Âne des Pyrénées

7 endangered horse breeds



Auxois



Boulonnais



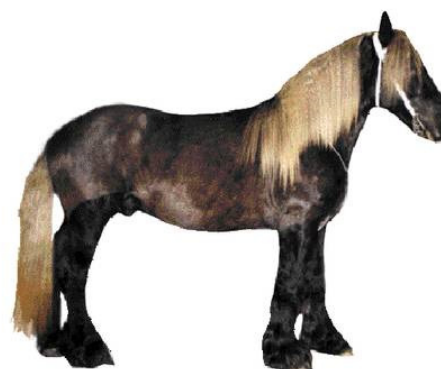
Cob Normand



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