

Implementation of the first Italian semen cryobank of autochthonous turkey breeds: preliminary results

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INTRODUCTION

In the last few decades in Italy, Farm Animal Genetic Resources of avian species have rapidly declined. In addition to *in vivo* management, *in vitro* conservation is also a strategic method used in order to secure genetic diversity within a wide range of lines and breeds. Semen cryopreservation is the most adapt technology within *ex situ in vitro* conservation programs, to maintain the avian genetic resources in gene banks. Recently, thanks the financed project "TuBAvI" research groups from Molise and Milan have realized the first Italian semen cryobank of native chicken and turkey breeds.

AIM

The aim of this work is to describe all of the activities fundamental for:

- 1) the identification of a successful freezing protocols;
- 2) the write up of the standard operative procedures (SOP);
- 3) the storage of turkey semen doses in the cryobank and hence its implementation.

MATERIALS AND METHODS

The identified reference cryopreservation protocol guarantees at least 30% of live and motile spermatozoa after the freezing process and ensures an adequate fertility rate.

The reference freezing protocol is reported in the below figure. Briefly, the semen was collected by abdominal massage (step 1), pre-diluted with Lake diluent and cooled at 4°C for 25 min (step 2), extended with freezing extender to reach the final concentration of 10% dimethylsulfoxide, 0.5 mM Ficoll and 3×10⁹ sperm/mL (step 3). Then, the semen is loaded into 0.25 mL plastic straws (step 4), equilibrated at 4°C for 20 min (step 5), frozen by exposure at the height of 10 cm above liquid nitrogen (LN₂) vapor for 10 min and finally transferred into LN₂ (step 6).



Semen Collection

Pre-dilution and cooling of semen

Semen dilution with freezing extender

Semen loading into straw

Equilibration and Exposure above LN₂

Straw transfer into LN₂ for storage

RESULTS

The realization of a semen cryobank of native turkey breeds is a valid tool for the conservation and safeguard of the avian biodiversity in Italy. The doses stored in the cryobank could be useful in improving the genetic variability within farms, in correcting any selection errors and in reducing inbreeding problems. A second funding is foreseen to increase its consistency in term breeds and donators.



CONCLUSIONS

The draw up of the SOP results as important because there has been no regulation on the organization and management of an avian germplasm cryobank. However, the draw up of the SOP included the FAO indications and the following aspects: the choice of priority breeds, selection of semen donors, infrastructures and storage sites, cryopreservation process, doses traceability and management of cryobank. Currently, 85 turkey semen doses from Nero d'Italia, Romagnolo, Ermellinato di Rovigo and Bronzato breeds are stored in the first Italian cryobank.