

Abstract

Intracytoplasmic sperm injection, or ICSI, is a commonly used method in assisted reproduction centers and is also an effective tool for a fertilization research. The interspecific ICSI is the suitable method for production of zygotes, which are formed by association of gametes of different, distantly unrelated species. It can bypass natural interspecies barriers and create so-called interspecific zygotes. This can be used to evaluation properties of genetic material. In this work, this method was used for evaluation of the quality of stored sperm originating from male goats of the white shorthair goat genetic resource.

The aim of this work was to evaluate the effect of cryopreservation on the quality of goat sperm, which was frozen in three different ways at two different breeding stations. Freshly collected semen served as a control group. The heads of xenogeneic goat sperm were injected into mature mouse oocytes. The injected oocytes were allowed to develop to the zygote stage, in which both female (mouse) and male (goat) pronuclei was formed. In male pronuclei, DNA damage was assessed by immunofluorescence staining with anti- γ H2AX antibody, a marker of DNA breaks.

The results obtained by measuring the level of γ H2AX fluorescence intensity in the paternal pronuclei show that the values differ between the individual groups of semen. The lowest values of fluorescence intensity, and thus the lowest DNA damage, were found in freshly collected semen (11.93 ± 8.06). Variability was also demonstrated between the experimental groups of frozen semen. The influence of different methods of semen collection and its freezing and the influence of the composition of the freezing medium were confirmed. The highest values of fluorescence intensity were found in the freezing medium with the additioned egg yolk (39.23 ± 8.06).

This method of assessing the quality of gamets of genetic resources animals and, in the future, endangered animal species, is a relatively simple and inexpensive method that can replace previous sperm quality assessment techniques with the advantage of replacing hard-to-reach oocytes of endangered species with readily available mouse oocytes.

Key words

ICSI, biodiversity, gene sources, sperm