

## Editorial

# New Possibilities of Sperm Freeze-Drying

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A large number of strains of genetically engineered animals have been generated worldwide. Sperm preservation is an indispensable tool for maintaining these strains as future genetic resources. Furthermore, the risk of genetic and microbial contamination during breeding has been reduced by sperm preservation. Generally, sperm is preserved in liquid nitrogen. Although this is a gold standard method, it is costly since specialized equipment and a constant supply of liquid nitrogen is required.

Offspring can be produced from oocytes fertilized with sperm after freeze-drying [1], and successful results have been reported in mice, rats, hamsters and rabbits. Similar research is now being undertaken on domestic animals and primates. Preservation of sperm by freeze-drying is an innovative method because liquid nitrogen is not required. Additional advantages of freeze-dried sperm are that they can be stored at 4°C for a long time, and stored and transported for short periods at room temperature without the use of liquid nitrogen or dry ice as cooling agents. Furthermore, it has been demonstrated that mouse and rat sperm can be preserved for a long time after freeze-drying using a simple solution containing 10 mM Tris and 1 mM EDTA (Ethylenediaminetetraacetic Acid) adjusted pH ~8.0 [2,3]. Short-term preservation of freeze-dried sperm at room temperature also led to easier oversea transportation of preserved strains. In addition, valuable strains can be stored temporarily at room temperature even in the event of a power failure, interruption to the liquid nitrogen supply, or other emergencies caused by disasters such as earthquakes and typhoons. Freeze-drying of sperm, rather than cryopreservation, is expected to become a new simple preservation method for genetic resources.

Techniques to produce genetically engineered animals have been established. Although it became possible to produce knockout animals

by establish of Embryonic Stem (ES) cell, a simple and effective method was reported recently whereby knockout animals were produced by the introduction of Zinc-Finger Nucleases (ZFNs) [4] and Transcription Activator-Like Effector Nucleases (TALENs) [5] without using ES cell. Co-injection of freeze-dried sperm and exogenous DNA into oocytes is one of the methods to produce transgenic animals [6]. This means that freeze-dried sperm can be used to produce transgenic and knockout animals by co-injection of exogenous DNA/RNA.

Freeze-drying of sperm can contribute not only to the preservation method for genetic resources, but also to the simple and effective production method of genetically engineered animals. This suggests that in future liquid nitrogen should not be necessary to preserve sperm. These reports suggest re-consideration of the preservation method for sperm and subsequent applications.

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