Chapter 1

Introductory Chapter: Application Fields of Cryopreservation Biotechnology

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http://dx.doi.org/10.5772/intechopen.82229

1. Cryopreservation biotechnology

Cryopreservation is the process of freezing the biological material at a temperature of liquid nitrogen (LN_2) (−196°C). This means biological activities discontinue including the biochemical reactions creating cell death and DNA damage at these low temperatures. In this way, it is possible to store the biological materials unchanged for centuries with the capability of recovering of the cell functionality following the thawing process.

The cells that were chosen for the early studies on the effects of freezing and thawing on cell viability were gametes. First cell material was "sperm cells" because of their availability, small size, and motility that was a simple marker of viability. Second cell type was "oocytes" due to their size large enough to allow for simple morphological evaluation.

Spallanzani [1] for the first time reported successful sperm freeze-thaw application on stallion semen. Nowadays, there are numerous cryopreservation protocols varying in terms of extenders, storage temperatures, freezing/thawing periods, and biological samples, which are suitable for the cryopreservation process.

The success in cryopreservation of biological materials has been gradually increasing every year with the understanding of physical and chemical process occurring during the freezing and thawing cycle. From this point of view, it is well known that intracellular ice formation, especially, is an important issue that has to be controlled to keep the cell membrane undisturbed and the cells lively. The critical issues for prevention of ice formation are the freezing rate and freezing medium composition. The freezing medium is known as a cryoprotectant containing extender solution. The choice of cryoprotectant and its concentration show differences between cells and species and influence the cryoprotection results.

Finally, cryopreservation biotechnology focuses on preservation of cells that have many applications in the fields of human and veterinary medicine, agriculture, and aquaculture. In addition, this biotechnology has many applications in biomedical researches, specifically in the areas of immunology, virology, neurobiology, toxicology, and pharmaceutical industry.

2. Application of cryopreservation biotechnology in medical sciences

In human medicine, cryopreservation has gained its importance when its usage in infertility treatment was realized. Since then, gamete cryopreservation has been developed to solve fertility problems in this field.

Sperm was the first type of reproductive cell successfully frozen and still remains the easiest cell to freeze because of containing low amounts of cytoplasm and consequently low quantity of water. Furthermore, sperm nuclear material is compressed and preserved against physical injuries. For these reasons, cryopreservation of sperm cells gives excellent results in terms of viability and fertility and is widely used in human medicine today.

In recent years, protocols regarding freezing of semen and embryos were established successfully and live births from assisted reproductive cycles using frozen semen or embryos were recorded. In addition, researches have also focused on the cryopreservation of human oocytes and ovarian tissues. While there are still insufficient researches especially for the oocytes, studies on the cryopreservation of immunological memory lymphoid cells, aortic root allografts, and osteoblasts for bone banking are going on. Cryopreservation of cornea, umbilical cord, and hematopoietic cells and sperm banking procedures are performed routinely in the field of human medicine [2].

In veterinary medicine, preservation of gametes is closely connected with the development of artificial insemination. Today, reproductive biotechnologies such as artificial insemination used in breeding programs are well developed.

During the last 100 years, many hundreds of species have been lost and a third of the breeding animals are threatened with extinction. Concerning with the threatened species, cryopreservation of genetic material (sperm, egg, and embryo) is used for the genetic management programs and genetic resource banking [3].

Since the first successful cryopreservation of bull semen [4], it is being used to reproduce scarce and threatened species. Numerous bovine calves have been produced via transferring of the cryopreserved embryos into cow, which are artificially fertilized with the frozen-thawed bull sperm greater than 25 million each year [5]. Nowadays, tissues, cultured cell lines, DNA, and serum samples could be frozen and stored in cryogenic banks.

3. Application of cryopreservation biotechnology in biological sciences

Cryopreservation is one of the most reliable methods for long-term conservation of plant genetic resources, because all metabolic processes and physicochemical changes are suspended at the cryogenic temperature (−196°C).

This biotechnology mostly interested with the germplasm cryopreservation is used for the genetic improvement of domestic varieties and their well adaptation to environmental changes in the field of agriculture. In spite of preservation of plant germplasm in cryogenic conditions is comparatively a new practice, a range of cryopreservation techniques for the conservation of plant cells and tissues were developed by the scientists more than 40 years [6]. Nowadays, it is a feasible application of using these techniques for the plant genotypes. A large number of laboratories are constituting this biotechnology for the aim of preservation of genetic resources. Recently, new cryogenic procedures using cryoplates (the V cryoplate and D cryoplate) have been developed. These methods provide some advantages such as ease of handling during the application and high regrowth rates, following cryopreservation process.

In addition, gamete, embryo, and embryonic cell cryopreservation has become a tremendous value in aquatic biotechnologies, which provide an important tool for the propagation of economically important species and also in protection of the endangered species and genetic diversity in aquatic species.

Since the first work of Blaxter [7] with Atlantic herring spermatozoa, sperm cryopreservation protocols are now available for over 200 finfish and shellfish species.

According to results of the researches, cryopreservation of sperm from marine fish species is more successful when compared to those obtained from the freshwater fish, and fertilization rates are similar to those obtained with mammalian species [8].

4. Conclusion

Cryopreservation has many biotechnological applications in different fields. This situation has been increasing the importance of cryobiology as a science, examining the effect of ultralow temperatures on cell, tissue, organ, and organisms and also the freezability of these structures maintaining their viability [9]. It is possible to underline that better understanding of functional properties of thawed cells following freezing process has been accelerating development of the cryopreservation biotechnology.

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References

[1] Spallanzani L. Observations, and Experiments around Freezing of Sperm and Eggs in Humans and Animals. Rome: Moderna; 1776

[2] Depalo R, Loverro G, Selvaggi L. *In vitro* maturation of primordial follicles after cryopreservation of human ovarian tissue: Problems remain. Medical and Pediatric Oncology. 2002;**38**:153-157

[3] Holt WV. Alternative strategies for long-term preservation. Reproduction Fertility and Development. 1997;**9**:309-319

[4] Polge C, Smith AU, Parkes AS. Revival of spermatozoa after vitrification and dehydration at low temperatures. Nature. 1949;**164**:666

[5] Mapletoft RJ, Hasler JF. Assisted reproductive technologies in cattle: A review. Revue Scientifique et Technique. 2005;**24**(1):393-403

[6] Engelmann F. Plant cryopreservation: Progress and prospects. In Vitro Cellular and Developmental Biology—Plant. 2004;**40**:427-433. ISSN: 1054-5476

[7] Blaxter JHS. Sperm storage and cross-fertilization of spring and autumn spawning herring. Nature, London, UK. 1953;**172**:1189-1190

[8] Tsvetkova LI, Cosson J, Linhart O, Billard R. Motility and fertilizing capacity of fresh and frozen-thawed spermatozoa in sturgeons *Acipenser baeri* and *Acipenser ruthenus*. Journal of Applied Ichthyology. 1996;**12**:107-112

[9] Bozkurt Y. The Role of Cryobiology in Conservation of Aquatic Genetic Resources. Saarbrücken, Germany: Lambert Academic Publishing; 2017. 94 p. ISBN: 978-3-330-05346-5