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Opinion Article

Procedures and Clinical Manifestations of Cryopreservation

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DESCRIPTION

Organelles, cells, tissues, extracellular matrix, organs, and any other biological constructs susceptible to damage from uncontrolled chemical kinetics are preserved by cooling to extremely low temperatures (typically -80°C or -112°F using solid carbon dioxide or -196 °C (-321 °F) using liquid nitrogen). This process is known as cryo-preservation or cryo-conservation. Any enzymatic or chemical activity that might harm the biological material in issue is effectively stopped at lowest temperatures. The goal of cryopreservation techniques is to attain extremely low temperatures without inflicting extra harm from ice crystal formation during freezing. The material to be frozen was often coated with a group of molecules known as cryoprotectants during traditional cryopreservation. As a result of the intrinsic toxicity of many cryoprotectants, new techniques are being researched.

The main idea behind cryopreservation procedures is to freeze sperms, eggs, or embryos in substances like liquid nitrogen at extremely low temperatures (below zero). Low temperatures stop all biological activity within cells, allowing them to last indefinitely. The sperm or embryos are fertilised or employed in a therapy cycle as needed.

The complete cryopreservation steps for keeping the collected biological samples safe are as follows:

Embryo selection

The woman is given medicinal stimulation to produce more eggs as part of the IVF procedure. Additionally, the man produces more sperm cells. The development of several embryos is common when both are combined. One to four embryos are often transferred into the woman by the doctor, who discards the remaining. The healthiest embryo or embryos are chosen in this case so they can be saved for later use.

Removal of the water content

Without replacing the water in the cells, the chosen embryo cannot be frozen immediately. This is due to the fact that when it is frozen, the water content within the cells also freezes and crystalizes. The crystallisation process may result in enlargement, which may cause the cell to burst open and die. Cryoprotectants are employed to refill the water content in the cells in order to stop this from happening.

Thawing

The thawing process is generally carried out by plunging the vials into a warm water bath with vigorous swirling. It also causes the vials to get transferred or move to another bath at 0 °C.

Washing and reculturing

The saved material is washed to remove the cryoprotectant. Then the material is recultured in a fresh medium.

Measurement of viability

Due to storehouse stress, there's a possibility of cell death. The presence of viability can be seen in utmost cases.

Toxicity

The major disadvantage of using low temperature that lead to crystallization of water therefore can produce unwanted physical and chemical event that may injure the cells that are being prepared. During cryopreservation process using vitrification or freezing, about half of the cell fluids were replaced with cryoprotectant molecules. These cryoprotectant molecules sometimes cause toxin while in warm temperature. For illustration as a cryoprotectant in warm condition, propylene glycol is nontoxic while ethylene glycol metabolized to poisonous elements. Newly it was set up that lipophilicity of cryoprotectant could help in deeper penetration of cryoprotectant into cells causes destabilization of cells. On the other hand, strong hydrogen adhesion also correlates with toxin by breaking up the hydration shell around macromolecules. The electrical properties of cryoprotectant also contain membrane toxins. Among all cryoprotectants, DMSO is severely poisonous, mostly DMSO/formamide result has been useful in vitrification process, but technique of biomedical toxin reduction is still unknown.

Applications

Now-a-days, cryoprotectants have some adaptable use. Primarily in conserving organ cells, in molecular biology, cryosurgery,

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blood transfusion, artificial insemination, bone marrow transplantation, *in-vitro* fertilization and most newly in relating unknown transmittable complication or pathogen. During 1996, first embryo preservation for fertility was done by using IVF cycle. IVF cycle was prerequisite before chemotherapy of breast cancer diagnosed cases. Conserving oocytes using cryoprotectants is formerly an established fashion. This approach causes no implicit side effects on labor. Same way, sperm and semen can be used after proper cryopreservation. Conserving testicular gonads using cryopreservation is still in immaturity, but in coming future cryopreservation can be used in vasectomy.

CONCLUSION

Cryobiology will have a significant inability in near future as a number of transplantation of vital organs of humans is growing more demanding in the present-day age. Yet, so numerous important complications are to be eased, like the exact medium of cryoprotectant, mode of action and cellular relations with implicit toxin. Further futuristic examination is demanded in respect of optimizing cryoprotectant concentration at slow and high freezing process. It was observed that macromolecular cells can effortlessly preserved by cryoprotectants but as far as large tissues or organs (kidney, liver, heart, lungs, and skin tissues) are concerned, the success rate of freezing isn't over to the mark by using cryoprotectant. Additional optimized examination on selection of specific concentration of cryoprotectants, solidifying and thawing rates, elegant infusibility, and equilibrium times in cryopreservation might lead to better cell survival during preservation. Ultramodern arising cryobank is a promising approach towards organ transplantation. But still, standardization and optimization ways are consent for best cryopreservation.