

Research on the Survival Rate of Vitrification Transplantation of Frozen Embryos in Mutton Sheep

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Abstract

Cryopreservation of embryos is indispensable in the field of assisted reproduction. At present, many countries use the freezing method to breed meat sheep. Methods Save mutton sheep embryos and obtain certain embryo survival rates, implantation rates, and pregnancy rates. Gestation rates and delivery rates have become mature methods of embryo freezing. But its disadvantages are time-consuming, high cost, and inevitable freeze and thaw. Damage to the embryos is caused by the formation of ice crystals during this process. Therefore, it is necessary to discuss it. An alternative method of freezing the embryos. Vitrification preserves embryonic energy. It can overcome the disadvantages of program freezing and is expected to achieve a higher embryo survival rate. Therefore, more and more scholars are paying attention to it, and become one of the hot issues in the field of assisted reproduction at home and abroad. Vitrification technology mainly uses a high concentration of antifreeze, and then places the rings in frozen tubes immersed in liquid nitrogen. This method is the technique of freezing embryos at a cooling rate in a very short time. At present, the cryopreservation technology of mutton sheep embryos has gradually matured, mainly used for the production and research of mutton sheep. Vitrification is a simple and efficient freezing method developed after programmed freezing and does not require a slow sequential cooling process.

Keywords

Mutton sheep, vitrification of frozen embryos, conception rate, transplantation, survival rate, influence

Introduction

After the cells are treated in vitrification liquid for a short time, they are directly put into liquid nitrogen for freezing, that is, the vitrification freezing phase can be used for sequential freezing, which greatly reduces the time consumption of expensive freezing devices purchase and the freezing process and facilitates the popularization and application. In recent years, the quality improvement of local mutton sheep has gradually increased, which is conducive to embryo transfer with local mutton sheep as recipients, producing high-quality breeding sheep, and providing high-quality and low-cost breeding sheep for farmers and herdsman. The aim of this study was to investigate the role of the recipient sheep. The influence of the level of feeding management, embryo age, and embryo freezing method on the pregnancy rate of mutton sheep embryo transfer was explored through experiments, which provides a reference for the promotion of embryo transfer pregnancy rate in this area [1].

1. The basic principles and significance of vitrification

The basic principle of vitrification is to use science to put a high concentration of antifreeze in an ultra-low temperature environment for curing, so as to obtain irregular glassy solid substances, which can preserve the normal distribution of

antifreeze molecules and ions under liquid and play a protective role in vitrification. After the preserved cells are dehydrated in the cryoprotectant solution, the rapidly cooled cells will play a protective role when endogenous cytoplasmic macromolecules such as proteins and cryoprotectants are concentrated in the cell.

Embryo reservation is one of the key technologies in animal reproduction and engineering, with high-quality embryos. At the same time, combined with the post-estrus embryo transfer technology, it can not only preserve the species of endangered animals, but also make the introduction of animals no longer limited by regional and time conditions, so embryo transfer should be promoted. Vitrification is a simple and efficient freezing method developed after programmed freezing and does not require a slow sequential cooling process. After short treatment in vitrification liquid and directly frozen in liquid nitrogen, that is, the vitrification freezing phase can be used for sequential freezing, which greatly reduces the time-consuming of expensive freezing instruments and facilitates the popularization and application. In recent years, the quality improvement of local mutton sheep has gradually increased, which is convenient for local mutton sheep as recipients, to produce high-quality breeding sheep, and to provide high-quality and low-cost breeding sheep for farmers and herdsman [2].

The results showed that there were no statistically significant differences in embryo survival rate, transfer rate, actual pregnancy rate, and live birth rate between the two systems. In order to reduce the risk of cross-contamination and disease transmission during long-term cryopreservation, closed glass freezing of sheep embryos has been widely used by embryo laboratories around the world, but the long-term effect requires more experimental data. The formation of ice crystals during freezing and thawing is the main cause of embryonic injury in mutton sheep. Vitrification partly compensates for this deficiency by directly converting the fluid into a glassy state, but the large amount of fluid contained in the blastocyst cavity theoretically increases the risk of cell damage. Artificial contraction (AS) technology can remove the fluid contained in the capsule before vitrification, thus improving the permeability of CPAs, and freezing the survival rate of blastocysts and pregnancy outcomes [3].

2. Effect of graft survival

This paper aims to explore the breeding management level, embryo age, embryo freezing method, and other factors on the influence of mutton sheep embryo transfer pregnancy rate, for the region of embryo transfer technology, and the egg nest corpus luteum does not affect the transplantation effect, the recipient mutton sheep plasma progesterone concentration is related to the number of corpus luteum, so achieve pregnancy [4]. The main role of agonist hormones is to promote embryo implantation and maintain pregnancy. Effect of an ovarian number of the yellow body on embryo transfer pregnancy rate in recipient sheep.

The management of recipient sheep is very important, and the conception rate is directly related to the management of recipient sheep. When embryo transfer, the physical condition of the recipient sheep reaches a medium level, and most of the embryos can be implanted in the uterus, giving insufficient living conditions for the mutton sheep, which is not conducive to the growth of embryo implantation and postpartum lambs, and significantly affects the embryo transfer effect of the recipient sheep [5]. The conception rate of upper-upper sheep was obviously better than that of middle and lower recipient sheep, which is consistent with the research results of upper-upper sheep. Therefore, recipient sheep should be selected as much as possible to ensure a high pregnancy rate. The effect of embryo age on the pregnancy rate of frozen embryos is generally considered to be the embryo transfer before the morula embryo. Good effect, high pregnancy rate. Blastocyst culture and transplantation have a great influence on the embryo implantation rate and pregnancy rate. The control of multiple pregnancy rates is significant. Therefore, it is necessary to keep the remaining blastocysts effectively frozen and preserved. However, the programmed cryopreservation of blastocysts was never ideal. Recently, the application of vitrification has enabled the freezing of blastocysts. Save the new steps. So far, the application of vitrification has been successfully used to freeze embryos in many mammals such as cattle, pigs, and rats [6].

Pregnancy and birth rates were higher after resuscitation and transplantation compared to fresh blastocysts. There was no significant difference observed. In addition, vitrified mutton sheep blastocysts were obtained. Great progress has been made. Choi et al. [8] first preserved human cysts by vitrification. Successful embryo transfer occurred with a survival rate of 51.6%. Cho et al. [9] improved on this complex. The survival rate of the vitrification-frozen embryos was 50.6% to 82.6%, and the clinical pregnancy rate was 34.1%. Thereafter, 725 blastocysts were cryoprotected using frozen rings as vehicles by Mukaida et al. The survival rate was 80.4%, and the clinical pregnancy rate was 37%. Recently, the first amp quot f1 l1 artificial contraction of expanded blastocysts was achieved by Hiraoka et al. Vitrification, blastocyst survival, and clinical pregnancy rates as high as 98%, respectively. In summary, the blastocysts are preserved by vitrification. Encouraging results have been achieved, and vitrification has replaced this procedure. Methods are gradually becoming a development trend of blastocyst freezing.

Studies have shown that the implantation rate of mulberry embryo transfer is higher than that of blastocyst transfer. Some experiments have proved that the blastocyst stage is the best time interval for embryo transfer, the pregnancy rate is not very high; the blastocyst embryo conception rate is significantly higher than the mulberry embryo. The pregnancy

rate of goat blastocysts was significantly higher than mulberry embryos and slightly higher than transplanted embryos. The pregnancy rate in mulberry embryos. However, there was no significant difference, indicating that conventional freezing of mutton sheep embryos has been widely used in production. Study of vitrification: it is also reported that the survival rate of expanded blastocysts can be high after the seventh day of freezing, while the survival rate of morula embryos is relatively low. The pregnancy rate and contrast of fine-tube vitrification were not significant, but the embryo tube was directly thawed and transplanted, yielding a difference. The advantage of glass embryo vitrification is that it can be done at room temperature of 20-25 without the use of a cryostat. The freezing process is manual, but the embryo freeze method takes a long time with the help of a freezer, and the cooling process takes a long time to complete. In 1970, the researchers found the antifreeze protein in the serum of deep-sea fish and achieved a good antifreeze effect [7].

Due to the high concentration of vitrification cryoprotectant, mutton sheep embryos, and mutton sheep, embryos need a large amount of time for cryopreservation. The toxic effect will be enhanced accordingly, so it needs to be strictly screened to prepare for low toxicity. Efficient vitrification solution is an important factor in embryonic vitrification. All kinds of antifreeze according to different concentrations, and different balance mix. Can be arranged and combined into a variety of vitrification frozen liquids. Methods to find (achieve) experts have done a lot of research on low-toxic and efficient vitrification. Systematic research and exploration, the solution is to choose low-toxicity antifreeze. Preparation and combined application of two or more antifreeze agents (including at least permeable cryoprotectant), etc. AFP is a spherical culture cassette found mainly in Lepidopteran fish. Embryo freezing can reduce the formation of ice crystals during cooling and act as cryoprotection. The addition of sucrose as a cryoprotectant has been widely used in vitrification of mouse embryos. Traditional antifreeze is mainly synthetic objects and antifreeze proteins with antifreeze ability in nature. Its discovery provides a new idea for the improvement of frozen liquids. There are four kinds of antifreeze proteins, among which the fish antifreeze protein was studied relatively much because it was found earlier. AFP III Is one of the fish antifreeze proteins and has been shown to be suitable for embryo freezing. We indicated that the survival and hatching rates of thawed frozen blastocysts were 83, respectively. 7% and 75. The survival and hatching rates of frozen blastocysts after thawing were 56, respectively. 3% and 64. 4%, significantly higher than that in the EFS 40 group 391-393. (51.7% and 45. Therefore, AFP) can serve as a good protective agent to improve the vitreous freezing efficiency of embryos [8].

3. The development prospect of vitrification embryo technology in mutton sheep

The freezing technology of mammalian embryos has been developing, and experiments in various experimental animals and most domestic animal embryos have been successful with satisfactory development. In particular, the cryopreservation technology of cattle and sheep embryos has been basically matured and applied in livestock production. But in general, the protective agents currently used are toxic to the embryos, and the effect of in vitro culture and transplantation after freezing and thawing is not satisfactory, and the procedure of freezing and thawing is very complicated. Fortunately, research in this area has begun, and embryo freezing technology is gradually combined with in vitro fertilization, embryo segmentation, sex identification, and other techniques [9]. It is expected that in the near future, embryo vitrification and freezing technology will be further improved and standardized. It will be combined with other embryonic engineering technologies, and will also make new breakthroughs in the field of livestock production and technology. Embryonic freezing plays an increasingly important role in assisting reproduction and preserving reproductive capacity. The cryopreservation protocol was optimized to further avoid freezing injury of embryos. It is hoped that by improving the frozen embryo medium, reducing oxidative stress and apoptosis of cells, and improving the vitrification embryo cryopreservation device, the freezing process will be safer and more efficient, and make breakthroughs in the vitrification freezing of mutton sheep embryos in the near future [10].

Frozen early embryos of mutton sheep can accumulate the pregnancy rate, reduce the multiple fetal rates, and avoid the ovarian hyperfunction of mutton sheep. In the field of assisted reproduction, the stimulation danger to patients is indispensable. The less important part. Vitrification refers to a high concentration of cryoprotection. During the rapid cooling process, the reagent forms an extremely viscous liquid. The glass does not have any form of ice crystal shape between the body and the crystal., Thus avoid various damage caused by icing [4]. Compared with the conventional programmed freezing technique, vitrification has simplified the operation steps. The freezing time is shortened, eliminating expensive procedures to cool the instrument to avoid it. Avoid consuming large amounts of liquid nitrogen. The vitrification technology should be carefully operated, and only rational cooperation can effectively achieve its success rate.

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References

- [1] Wang Yanlin & Zhu Guijin. Effects of three frozen storage tubes on vitrification and freezing of human tripronuclear cleavage stage embryos [J]. *Reproductive and Contraception*, 2006, 26 (8): 500-503.
- [2] Li Jinsong, Sun Qiang, Chao Qimei, et al. Establishment of the rapid freezing method for HBV-C transgenic mice [J]. *Chinese Journal of Experimental Zoology*, 2001, 11 (1):
- [3] Mukaida T, Wada S, Takahashi K., et al. Vitrification of human embryos based on the assessment of suitable conditions for 8-cell mouse embryos [J]. *Hum Reprod*, 1998, 13(10): 2874.
- [4] El-Danasouri I & Selman H. Successful pregnancies and deliveries after a simple vitrification protocol for day 3 human embryos [J]. *Fertil Steril*, 2001, 76(2): 400.
- [5] Cuello C, Gil MA, Parrilla I, et al. In vitro development following one—step dilution of OPS—vitrified porcine blastocysts [J]. *Theriogenology*, 2004, 62(6): 1144-1152.
- [6] Dattena M, Accardo C, Pilichi S, et al. Comparison of different vitrification protocols on viability after transfer of ovine blastocysts in vitro produced and in vivo derived [J]. *Theriogenology*, 2004, 62(3-4): 481-493.
- [7] Choi DH, Chung HM, Lira JM, et al. Pregnancy and delivery of healthy infants developed from vitrified blastocysts in IVF—ET program [J]. *Fertil Steril*, 2000, 74(4): 838-839.
- [8] Cho HJ, Son WY, Yoon SH, et al. An improved protocol for dilution of cryoprotectants from vitrified human blastocysts [J]. *Hum Reprod*, 2002, 17(9): 2419-2422.
- [9] Mukaida T, Nakamura S, Tomiyama T, et al. Vitrification of human blastocysts using cryoloops: clinical outcome of 223 cycles [J]. *Hum Reprod*, 2003, 18(2): 384-391.
- [10] Hiraoka K, Hiraoka K, Kinutani M, et al. Blastocoele collapse by micro pipetting prior to vitrification gives excellent survival and pregnancy outcomes for human day 5 and 6 expanded blastocysts [J]. *Hum Reprod*, 2004, 19(12): 2884-2888.